Oral Choline Administration to Patients with Huntington’s Disease

*John H. Growdon and **Richard J. Wurtman

*Tufts-New England Medical Center Hospital, Boston, Massachusetts 02111; and **Massachusetts Institute of Technology, Cambridge, Massachusetts 02139

Soon after the demonstration that choline increased brain acetylcholine levels in experimental animals (8,9,22), neurologists and psychiatrists began to examine choline’s ability to ameliorate the clinical manifestations of disease states thought to result from inadequate central cholinergic tone. In general, the decision to test choline (and the corollary belief that inadequate acetylcholine release characterizes the disease state) has been based on the results of clinical pharmacologic testing and on biochemical measurements in brains obtained at postmortem examinations. These studies indicated that anticholinergic drugs such as benztropine exacerbated chorea, whereas physostigmine, a drug that blocks the enzyme acetylcholinesterase that degrades acetylcholine, generally suppressed it (11,25). Direct biochemical measurements also implicated cholinergic deficiencies, since it was found that levels of choline acetyltransferase (ChAc), the enzyme located in cholinergic neurons that catalyzes the conversion of choline to acetylcholine, were reduced in brains of patients who died of Huntington’s disease (HD) (5, 27,38). These observations prompted investigators in several laboratories (2,11), including our own (18), to attempt to treat HD patients with oral doses of choline. This chapter describes the evidence that choline administration increases brain acetylcholine synthesis, levels, and release in rats and summarizes the published data on the use of choline in HD.

RATIONALE

The synthesis of acetylcholine from choline and acetyl coenzyme A is catalyzed by ChAc.

\[
\text{Choline} + \text{acetyl coenzyme A} \underset{\text{ChAc}}{\longrightarrow} \text{acetylcholine}
\]

The brain is apparently unable to synthesize choline de novo and must obtain it from the systemic circulation (1,15). Choline in the blood derives from two sources: Some is synthesized in the liver by the step-by-step methylation of ethanolamines (7), and some comes from dietary sources. Foods that are particu-
larly rich in choline include eggs, meat, fish, and beans (16). Choline levels in
blood vary widely, depending on the quantity of choline consumed in the diet
(24,44). The transport of choline into the intact brain is mediated by a low-
affinity uptake system located at the blood-brain barrier within the endothelial
cells of cerebral capillaries (31). Since plasma choline levels normally fall well
below the \( K_m \) of the system that transports choline into the intact brain (0.22
mM), this uptake system normally is highly unsaturated (32). Thus, any signifi-
cant variation in plasma choline levels should generate corresponding changes
in brain choline uptake, and ultimately in brain choline levels. Furthermore
the \( K_m \) of ChAc for choline (0.4 mM) and for acetyl coenzyme A (18 \( \mu \)M)
are both well above the normal brain concentrations of these substances. There-
fore, changes in the brain levels of either of these precursors for acetylcholine
might be expected to modify the rate at which the neurotransmitter is synthesis-
ed. A high-affinity choline uptake system (\( K_m = 1.5 \mu \)M) has also been observed
in synaptosomes prepared from cholinergic nerve terminals (21,45); this carrier
is sodium-dependent and is blocked by hemicholinium. The high-affinity system
may affect the distribution of choline within the brain by shunting it preferentially
to sites of acetylcholine synthesis; it may also allow neurons to recapture and
reutilize choline formed from the hydrolysis of acetylcholine released into syn-
apses. These relationships are diagrammed in Fig. 1. Acetylcholine synthesis
is apparently under "open-loop" control, since treatments that increase the
amount of substrate choline will also accelerate brain acetylcholine synthesis.
Thus, the systemic administration of choline to laboratory animals by injection
(8,22), dietary supplementation (9), or gastric installation (41) increases brain
choline and acetylcholine levels. In our laboratory, choline administration in-
creased brain choline and acetylcholine levels in all brain regions examined
(9), including the hippocampus, which contains cholinergic presynaptic nerve
terminals (23). Thus, the increased acetylcholine levels induced by choline admin-
istration are, by virtue of their intracellular location, available for release. The
increased levels of acetylcholine induced by choline administration apparently
result from increased de novo synthesis rather than from slowed inactivation.
We gave rats an acetylcholinesterase inhibitor, physostigmine, in conjunction
with choline. The resulting increase in brain acetylcholine levels was equal to
the sum of the effects of either agent alone; this indicates that choline acts by
enhancing acetylcholine synthesis (9).

The increase in neuronal acetylcholine levels caused by choline administration
to animals probably causes increased acetylcholine release as well. To examine
the relationship between precursor-induced increases in acetylcholine levels and
the amounts of the transmitter released into synapses, we utilized indirect experi-
mental approaches that involve measuring biochemical changes in cells that
are postsynaptic to the cholinergic neurons. Two such cells are the dopaminergic
neurons that terminate in the caudate nucleus, and the chromaffin cells of the
adrenal medulla, both of which contain the enzyme tyrosine hydroxylase, which
is rate-limiting in catecholamine synthesis. Tyrosine hydroxylase in the caudate
FIG. 1. Model illustrating sources of choline for brain acetylcholine synthesis. Choline enters the blood either from dietary sources (primarily lecithin) or hepatic synthesis. It is transported into the brain's extracellular fluid by a low-affinity transport system (L) localized within the capillary endothelium of the blood-brain barrier. It then can (a) be taken up within terminals of cholinergic neurons either by a high-affinity (H) or low-affinity (L) transport system; (b) be taken up within other brain cells by a low-affinity mechanism (L); or (c) pass into the cerebrospinal fluid or back into the bloodstream. The choline within nerve terminals is converted to acetylcholine through the action of choline acetyltransferase (CAT); once the transmitter is released into the synaptic cleft, it is hydrolyzed to choline by the enzyme acetylcholinesterase (AChE), after which choline may again enter the presynaptic neuron via the high-affinity uptake system. Under some conditions, a low-affinity system present within cholinergic nerve terminals also may participate in choline uptake. Moreover, a part of the choline taken up within cholinergic terminals may be utilized for functions other than acetylcholine synthesis (e.g., production of lecithin for incorporation into membranes).

nucleus undergoes a rapid and short-lived activation when the neuron containing it is depolarized (29); the enzyme in chromaffin cells apparently does not exhibit short-term activation, but is induced 24 to 48 hr after the cells are subjected to prolonged cholinergic stimulation (40). Choline administration (60 mg/kg i.p.) increased striatal tyrosine hydroxylase activity by 25% within 2 hr of injection (43). This effect was dose-related and was blocked by pretreatment with atropine, a muscarinic antagonist (Table 1). Choline administration also caused a parallel increase in the accumulation of DOPA, the product of this enzyme,
in animals that were treated with an inhibitor of DOPA decarboxylase. Choline administration also induced tyrosine hydroxylase activity in the adrenal medulla. The time course of this induction differed from that of the increase in adrenal acetylcholine levels; these returned to normal within 16 hr of choline administration, whereas a significant elevation in tyrosine hydroxylase activity (+30%) was first observed 24 hr after choline administration (41). When choline was administered daily by stomach tube for 4 days, adrenal tyrosine hydroxylase activity increased by 50%. This increase did not occur in control rats given saline or ammonium chloride; it was blocked by pretreatment with cyclohexamide, an inhibitor of protein synthesis, and by prior adrenal denervation (41). Choline administration also significantly increased urinary epinephrine levels (34). In another test situation, choline administration increased acetylcholine release from isolated hearts during electrical stimulation of the vagus nerve (13). These observations are all compatible with the view that choline is taken up into presynaptic terminals of cholinergic neurons, that choline accelerates the rate at which acetylcholine synthesis proceeds, and that it also accelerates acetylcholine release.

Although these findings indicate that choline administration increases acetylcholine release, they do not differentiate between two possible mechanisms, i.e., increased rate of firing versus increased amount of transmitter released per impulse. To distinguish between these two possible actions, we again utilized adrenal medullary tyrosine hydroxylase responses to acetylcholine. In these experiments, groups of rats received two treatments—choline, which presumably acts by increasing presynaptic acetylcholine levels, and an additional treatment (such as hypotension induced by reserpine, systemic 6-hydroxydopamine, or phenoxybenzamine; doses of insulin insufficient to produce severe hypoglycemia; or prolonged exposure to a cold environment) designed to accelerate the firing rate of the cholinergic splanchnic nerve to the adrenals (42). In all cases, administration of both choline and the other treatment had significantly greater effects

### Table 1. Effect of atropine on the choline-induced increase in striatal tyrosine hydroxylase (TOH) activity

<table>
<thead>
<tr>
<th>Treatment</th>
<th>TOH activity (nmoles hour⁻¹mg⁻¹)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>2.23 ± 0.09</td>
<td>(8)</td>
</tr>
<tr>
<td>Choline chloride</td>
<td>2.68 ± 0.10*</td>
<td>(9)</td>
</tr>
<tr>
<td>Atropine sulfate</td>
<td>2.29 ± 0.08</td>
<td>(8)</td>
</tr>
<tr>
<td>Atropine sulfate + choline chloride</td>
<td>2.36 ± 0.05</td>
<td>(7)</td>
</tr>
</tbody>
</table>

*Different from control groups or from atropine-treated groups at p < 0.05.

Animals received choline chloride (60 mg/kg) and atropine sulfate (40 mg/kg) intraperitoneally 2 hr before they were killed. Values are expressed as mean ± SEM. The number of samples assayed is indicated in parentheses. Data were analyzed by analysis of variance.

From Ulus and Wurtman (43).
or adrenal medullary function than the sum of the effects produced by either treatment alone. These observations suggest that choline acts primarily by increasing the amount of acetylcholine released per nerve firing, and not by changing the rate of firing. Thus, current evidence indicates that choline administration increases acetylcholine synthesis, acetylcholine levels, and also the amount of acetylcholine released from cholinergic terminals. These observations form the scientific basis for choline administration to patients with diseases, such as HD, in which physicians may wish to increase central acetylcholine tone.

**MATERIALS AND METHODS**

Ten patients with previously established HD took choline according to a single-drug open-label protocol (18). Each patient had a family history of HD consistent with an autosomal dominant mode of transmission, and each displayed neurologic features that included personality change, memory loss, dysarthric speech, involuntary movements (chorea), gait disturbances, and poor balance (Table 2). All patients were hospitalized for 1 to 2 weeks at the MIT Clinical Research Center, where complete histories and physical examinations were obtained with particular emphasis on the neurologic features of the illness. A clinical scoring system (0 = normal; 1 = mild; 2 = moderate; 3 = severe) was used to estimate disability in each of 16 areas: speech, memory, intellect, mood, strength, muscle tone, reflexes, tongue chorea, face chorea, upper extremity chorea, lower extremity chorea, akathisia, tremor, myoclonus, balance, and gait. Speech, memory, intellect, and mood were evaluated during the mental status examination but formal psychologic tests were not performed. The severity of chorea was estimated by counting the number of involuntary movements in a body part over a 30-sec period with the body part both at rest and in action. Observations were made daily by the same person (J.H.G.) in a comfortable, private room in order to increase consistency and to control for the effects of stress. Balance was tested by having the patient stand unsupported on one foot, and by measuring the patient's postural reaction to an unexpected tilt while seated in a chair. Gait was evaluated while the patient was walking, running, and tandem-walking. The scores for each of these variables were summed, and the resultant total score was considered to reflect the extent of the patient's disease. The minimal score (normal) would be 0; the score associated with maximal severity would be 48. Eight patients had combined surface electromyogram (EMG) and accelerometer recordings to quantitate involuntary movements further (46). In addition, moving pictures of each patient were taken under standardized conditions in which the patient sat quietly at rest, talked, protruded the tongue from the mouth for 30 sec, held the arms outstretched, wrote, stacked blocks, walked, ran, stood on one foot, hopped, and was tilted off balance while sitting in a chair. More moving pictures of each patient were taken when he or she left the hospital and returned for outpatient visits. The patient's performance, as recorded in the moving pictures, was graded by the authors according
TABLE 2. Clinical features in 10 patients with HD

<table>
<thead>
<tr>
<th>Patient no.</th>
<th>Sex</th>
<th>Age (yr)</th>
<th>Duration of HD (yr)</th>
<th>Concurrent medication (per day)</th>
<th>Some clinical characteristics*</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>M</td>
<td>32</td>
<td>7</td>
<td>Haloperidol, 3 mg; lithium, 1,200 mg; diazepam, 15 mg</td>
<td>++ + - +</td>
</tr>
<tr>
<td>2</td>
<td>M</td>
<td>59</td>
<td>6</td>
<td>Haloperidol, 4 mg; chlordiazepoxide, 20 mg; lithium, 1,200 mg</td>
<td>++ + + ++</td>
</tr>
<tr>
<td>3</td>
<td>M</td>
<td>56</td>
<td>10</td>
<td>Lithium, 1,500 mg; diazepam, 20 mg</td>
<td>++ ++ +++ +++</td>
</tr>
<tr>
<td>4</td>
<td>F</td>
<td>30</td>
<td>3</td>
<td>Haloperidol, 15 mg</td>
<td>- +++ - ++</td>
</tr>
<tr>
<td>5</td>
<td>M</td>
<td>50</td>
<td>17</td>
<td>Flupentixol, 2 mg; lithium, 900 mg; amitriptyline, 60 mg</td>
<td>+ +++ + +++</td>
</tr>
<tr>
<td>6</td>
<td>F</td>
<td>41</td>
<td>3</td>
<td>Haloperidol, 4 mg</td>
<td>+ ++ - ++</td>
</tr>
<tr>
<td>7</td>
<td>M</td>
<td>33</td>
<td>6</td>
<td>Haloperidol, 4 mg</td>
<td>+ ++ -</td>
</tr>
<tr>
<td>8</td>
<td>F</td>
<td>43</td>
<td>3</td>
<td>- +++ - ++</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>M</td>
<td>31</td>
<td>3</td>
<td>Meprobamate, 400 mg</td>
<td>- - + +</td>
</tr>
<tr>
<td>10</td>
<td>M</td>
<td>47</td>
<td>17</td>
<td>Haloperidol, 6 mg</td>
<td>+ ++ +</td>
</tr>
</tbody>
</table>

*Rating scale: -, normal; +, mild; ++, moderate; ++++, severe.
From Growdon et al. (18).
to the clinical scoring system and by global assessment: improved, deteriorated, or unchanged during choline therapy.

During hospitalization patients received a diet that provided 2,200 to 2,500 kCal/day, including 85 to 95 g protein, 240 to 260 g carbohydrate, and 100 to 120 g fat. This was chosen to approximate a normal diet, and no effort was made to increase the supply of foods known to contain high quantities of choline. The actual quantity of food consumed was not measured, nor were its content of free choline and choline bound to lecithin. Five patients (numbers 3,4,6–8) received 1 mg of physostigmine intravenously 30 min after an oral dose of 30 to 45 mg of propantheline, which was given to block the effects of peripheral cholinergic activation. Speech, writing, drawing, chorea at rest, and gait were tested 5 min after the physostigmine injection and at 15-min intervals for 1 hr thereafter. Patients continued their previously prescribed medications (Table 2) and then took choline, either the chloride or bitartrate salt, mixed in sweetened beverages; the initial dosage, calculated as the base, was 1 to 2 g four times a day, and the daily dose was increased every 2 days. The maximum dose varied among patients but ranged from 2 to 5 g four times a day (Table 3). Once a maximal nontoxic dose of choline had been established, the clinical testing, EMG-accelerometer recordings, and moving pictures were repeated. In addition, choline levels in serum and CSF were measured before and during choline ingestion by a radioenzymatic method (35).

RESULTS

Serum choline levels at 9:00 A.M. on the day prior to choline ingestion ranged between 10.7 and 15.2 nmoles/ml with a mean ± SEM of 13.6 ± 1.7 nmoles/

| TABLE 3. Total disability scores in 10 patients with HD before and during treatment with choline |
|---|---|---|---|
| Patient no. | Choline dose (g/day) | Duration of treatment (days) | Score * |
| 1 | 20 | 38 | 12 | 11 |
| 2 | 20 | 15 | 13 | 11 |
| 3 | 20 | 125 | 25 | 24 |
| 4 | 8 | 27 | 14 | 13 |
| 5 | 20 | 29 | 20 | 23 |
| 6 | 12 | 21 | 14 | 16 |
| 7 | 12 | 64 | 10 | 10 |
| 8 | 12 | 124 | 12 | 12 |
| 9 | 20 | 37 | 4 | 4 |
| 10 | 8 | 19 | 10 | 12 |

*The sum of sixteen clinical variables, each graded on a scale of: 0, normal; 1, mild; 2, moderate; and 3, severe (see text for details). There was no statistical difference in the scores before and during treatment (p > 0.1, sign test).

From Growdon et al. (18).
ml (Table 4). Serum choline levels rose in every patient during choline administration: On the second day of treatment (at 9:00 A.M., 1 hr after a 2-g choline dose) they ranged between 22.9 and 27.4 nmoles/ml with a mean of 25.8 ± 1.7 nmoles/ml (p < 0.001). The increase in serum choline levels was directly proportional to the amount of choline ingested. Five patients took choline doses of 2, 4, and 5 g four times a day on separate days, and serum choline was determined each day 1 hr after one of the choline doses. There was a dose-related linear increase in serum choline levels (r = 0.90). Choline levels were measured in the CSF 1 hr after maximal doses of choline (150 to 200 mg/kg/day) in eight patients. The mean choline level before treatment was 1.8 ± 0.1 and it rose 74% to 3.1 ± 0.3 nmoles/ml during treatment (p < 0.1).

Choline administration, either alone (patients 7 and 8) or in combination with other drugs (all other patients) did not suppress the signs of HD in any of the 10 patients (Table 3). Although the total scores did not change significantly, there were some minor benefits in some patients. Speech was smoother and more distinct in one patient (number 4) and balance and gait were steadier in five patients (numbers 1-4, and 8). These improvements did not last more than several weeks, however, despite continued choline administration. Choline did not suppress chorea in any patient (based on clinical examination, EMG-accelerometer recordings, and review of moving pictures) and choreic movements actually increased in two patients (number 5, with choline doses greater than 16 g/day, and number 10, with doses greater than 8 g/day). Physostigmine did not relieve any features of HD in the five patients tested (patients 3, 4, and 6-8).

Choline chloride has a bitter taste and was unpleasant to take, despite the use of sweetened beverages. Most patients who ingested choline chloride developed the aroma of rotten fish in their urine, sweat, and on their breath; this

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### TABLE 4. Effect of oral choline administration on blood choline levels (nmoles/ml) in patients with HD

<table>
<thead>
<tr>
<th>Patient</th>
<th>Before treatment</th>
<th>During treatment with 2 g choline 4 x/day&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>10.8</td>
<td>27.4</td>
</tr>
<tr>
<td>2</td>
<td>15.0</td>
<td>27.4</td>
</tr>
<tr>
<td>3</td>
<td>13.8</td>
<td>25.0</td>
</tr>
<tr>
<td>4</td>
<td>14.0</td>
<td>27.1</td>
</tr>
<tr>
<td>5</td>
<td>14.8</td>
<td>23.7</td>
</tr>
<tr>
<td>6</td>
<td>10.7</td>
<td>25.0</td>
</tr>
<tr>
<td>7</td>
<td>14.1</td>
<td>22.9</td>
</tr>
<tr>
<td>8</td>
<td>13.8</td>
<td>27.4</td>
</tr>
<tr>
<td>9</td>
<td>15.2</td>
<td>26.1</td>
</tr>
<tr>
<td>Mean ± SEM</td>
<td>13.6 ± 1.7</td>
<td>25.8 ± 1.7&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Blood samples were collected 1 hr after a choline dose on the second day of treatment.

<sup>b</sup>p < 0.001, Student's t-test.

From Growdon et al. (17).
disappeared when choline ingestion was discontinued. The odor apparently derives from the trimethylamines produced by the action of intestinal bacteria on choline (26), and does not occur after lecithin administration, which also elevates serum choline levels (24,44). High doses of choline chloride (>250 mg/kg/day) produced lacrimation, anorexia, vomiting, and diarrhea but no changes in pupil size or heart rate. These effects were transient and disappeared when the choline dose was lowered or discontinued.

DISCUSSION

Choline is the physiologic precursor of acetylcholine (1,15); the discovery that choline administration produces sequential elevations in blood choline, brain choline, and brain acetylcholine levels in rats led to its use in treating human diseases in which there may be deficient central cholinergic tone. Shortly after the publication of the animal data, Davis et al. (10) reported that 16 g/day of choline chloride suppressed choreic movements in a single patient with tardive dyskinesia. Subsequently, the usefulness of choline chloride or lecithin, the naturally occurring dietary source of choline, in treating patients with tardive dyskinesia was confirmed in numerous studies (11,19,20,39). The ability of choline and lecithin to suppress tardive dyskinesia illustrates a new mode of medical therapy in which a naturally occurring dietary substance (choline) that is a precursor for a neurotransmitter (acetylcholine) may be used to treat a nonnutritional brain disease (tardive dyskinesia). Thus, choline administration provided an opportunity, for the first time, to test the long-term effects of chronic cholinergic stimulation in patients with HD. The 10 patients who participated in this study were all ambulatory and living at home, and in some instances were still working; it was anticipated that choline administration might improve these milder patients more than those who were severely demented and bedridden. Even though oral doses of choline increased blood choline levels in all patients, choline administration did not suppress chorea in any patient. These results are similar to those reported by Aquilonius and Eckernas (2), who gave choline to five patients with HD. They reported minor changes in chorea during choline ingestion, but concluded that choline did not significantly suppress the movements. They measured serum choline levels and also found a linear increase over a range of 3 to 15 g of choline per day. Davis et al. (12) obtained the most favorable results thus far published with choline treatment in HD. They treated eight patients with 12 to 25 g/day of choline chloride and reported that chorea decreased significantly in three and was unchanged in five patients. The three patients who improved with choline had also improved transiently during physostigmine infusion. These investigators suggested that a patient’s response to physostigmine may help to predict the potential benefit of choline therapy (11).

The results of these three studies (Table 5) suggest that most patients with HD do not improve significantly during choline administration. Choline ingestion
significantly increases choline levels in blood, but it may not increase acetylcholine levels in brain to a clinically useful extent if there has been extensive presynaptic cholinergic destruction. Under normal conditions, ChAc is not saturated with its substrate, and increasing the amount of available choline will accelerate acetylcholine synthesis (9). In HD, however, cholinergic interneurons in the striatum are damaged (27,28), and may be less responsive to increases in brain choline levels. Choline administration would also be ineffective if there were extensive postsynaptic cholinergic destruction as well. For example, the number of striatal cholinergic receptors could be so reduced (14) that any newly synthesized acetylcholine, even if it were released, would be less likely to combine with a receptor site. This may also explain the failure of arecholine, a direct muscarinic agonist, to improve chorea in patients with HD (30). In addition, the loss of cholinergic neurons is only a part of the pathologic process, since other neurons, including those that contain glutamic acid decarboxylase (33) and angiotensin-converting enzyme (3), are also known to be destroyed in HD. These observations may explain why treatments that increase central cholinergic tone are relatively ineffective in altering the course of this disease. In contrast, choline has been used successfully to suppress tardive dyskinesia (11,20,39); its administration may also enhance memory in normal subjects (37) and improve social behavior (6) and learning (36) in patients with Alzheimer's disease. Further preliminary reports indicate that lecithin may also suppress tardive dyskinesia (19), and improve patients with Friedreich's ataxia (4). Perhaps the main determinant of whether a disease responds favorably to choline (or lecithin) is the extent to which the total neuronal pathology involves cholinergic neurons.

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ORAL CHOLINE TO HD PATIENTS

REFERENCES


