A Randomized Efficacy Trial of Citicoline in Patients With Acute Ischemic Stroke

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**Background and Purpose**—Citicoline (cytidine-5′-diphosphocholine; CDP-choline) may reduce central nervous system ischemic injury by stabilizing cell membranes and reducing free radical generation. A previous dose-comparison trial in patients with acute stroke found that 500 mg of citicoline appeared to improve neurological outcome with minimal side effects.

**Methods**—The current trial was a 33-center, randomized, double-blind, efficacy trial in 394 patients comparing placebo (n=127) with citicoline (n=267) (500 mg po daily) for 6 weeks, with a 6-week posttreatment follow-up period. Patients with acute (24 hours) ischemic strokes clinically assessed to be in the middle cerebral artery territory with National Institutes of Health Stroke Scale (NIHSS) ≥5 were enrolled.

**Results**—Mean time to treatment was 12 hours, and mean age was 71 for placebo and 70 for citicoline. Although mean baseline NIHSS were similar for both groups, there was a higher percentage of placebo patients with NIHSS <8 (34% vs 22%; P<0.01). The incidence and type of side effects were similar between the groups. The planned primary analysis (logistic regression: 5 categories Barthel) failed the proportional odds assumption and was rendered unreliable. There were no between-group differences seen on the planned secondary assessment analyses at 90 days, including the Barthel Index ≥95 at 12 weeks (last observation carried forward: placebo 40%; citicoline 40%) or mortality rate (placebo 18%; citicoline 17%). However, post hoc analyses in a subgroup of patients with baseline NIHSS ≥8 found that citicoline-treated patients were more likely to have a full recovery (Barthel ≥95): placebo 21%; citicoline 33%; P=0.05; whereas no difference was seen in patients with baseline NIHSS<8 (placebo 77%; citicoline 69%; P>0.1).

**Conclusions**—The results of this study indicate that citicoline was safe but ineffective in improving the outcome of patients with acute ischemic stroke who were enrolled in this trial. Post hoc analyses indicate that there may be a subgroup of patients with moderate to severe strokes who would benefit. (*Stroke*. 1999;30:2592-2597.)

**Key Words:** citicoline ▪ stroke ▪ treatment

Citicoline (cytidine-5′-diphosphocholine or CDP-choline) is an essential precursor for the synthesis of phosphatidylcholine, a key component of cell membranes. Although citicoline is a naturally occurring endogenous compound, the sodium salt of citicoline is synthesized for clinical use. During ischemia, phosphatidylcholine is broken down into free fatty acids, which in turn are used to generate free radicals that potentiate ischemic injury.1 The reversible synthetic pathway leading from choline to phosphatidylcholine is outlined in Figure 1A. Cerebral ischemia and hypoxia have been shown to reverse this synthetic pathway, leading to decreased phosphatidylcholine content in the cell membrane and increased concentrations of free fatty acids2–4 (see Figure 1B). The exogenous administration of citicoline has been shown in animal models to reduce this cell-membrane breakdown, leading to increased synthesis of phosphatidylcholine and decreased levels of free fatty acids5,6 (see Figure 1C).

The use of citicoline treatment has been shown to be beneficial in several animal models of ischemia or hypoxia,7–14 including recent studies with reversible focal occlusion13 and in an intracerebral hemorrhage model.16 These studies have found that citicoline treatment decreases free fatty acid concentration, improves neurological signs, decreases neurological deficits, restores animal learning performance, reduces glutamate-mediated injury, preserves phosphatidylcholine levels, and improves neuronal survival.

Citicoline has also been studied in several randomized clinical stroke treatment trials outside the United States,17–20 with initial time-to-treatment windows of 2 to 14 days. In these studies, doses of citicoline ranging between 250 to 1000 mg per day were found to improve global and neurological function at 90 days and promote earlier motor and cognitive recovery at 14 days. In a previous dose-finding, multicenter,
randomized trial in the United States, the effectiveness of 3 doses of citicoline (500 mg, 1000 mg, 2000 mg) versus placebo was evaluated in 259 patients with an acute ischemic stroke (“001A” trial). This trial found that both 500 mg and 2000 mg citicoline produced a significant treatment effect compared with placebo when the baseline National Institute of Health Stroke Scale (NIHSS) score was used as a covariate. Since the 500 mg group appeared to have less side effects, this dose was the one chosen for further efficacy evaluations.

The purpose of the current study (“007”) was to evaluate the safety and effectiveness of 500 mg citicoline versus placebo in patients who are examined within 24 hours of an acute ischemic stroke.

Subjects and Methods

This trial was a randomized, double-blind, placebo-controlled efficacy study of 500 mg of oral citicoline in patients with acute ischemic stroke. Patients for the trial were selected on the basis of eligibility criteria (see Table 1) from 31 stroke centers in the United States (See Appendix). The protocol used in this trial was approved for use by the institutional review board of each participating institution, and all patients or their legal representatives signed an informed consent.

The primary objective of this study was to determine the effects on recovery of 500 mg of citicoline given orally over a 6-week treatment period and a 6-week follow-up period in patients with acute ischemic stroke. It was hypothesized that citicoline would improve functional, global, and neurological outcome measures when compared with the placebo group.

To be eligible for the study, the patient must have been examined within 24 hours with symptoms consistent with ischemic stroke clinically assessed to be referable to the middle cerebral artery territory (angiographic confirmation not required). Patients were to have at least 5 points on the NIHSS with at least 2 of these points from the motor sections. For patients who awoke with their symptoms, it was determined that their onset time would start at the time that they awoke from sleep (not when they went to sleep). The rationale for allowing patients who awoke with symptoms to be enrolled were that there were no safety concerns with delayed therapy and that prior citicoline trials have effectively used time windows of up to 2 weeks. A baseline CT and/or MRI scan must have been consistent with a diagnosis of stroke. That is, although it did not have to be positive for a stroke, it could not show another diagnosis as a cause for the symptoms, for example, intracerebral hemorrhage or tumor.

All patients who qualified according to the inclusion/exclusion criteria and for whom informed consent was obtained from either the patient or family were randomly assigned to 6 weeks of treatment with either placebo or citicoline (500 mg) in a soft-gelatin capsule (identical in appearance and consistency). Patients were to be randomly assigned on a 2:1 basis to receive citicoline (267 patients) or placebo. (127) The patients were randomly assigned within each center with the use of a blocked randomization. No one at the local site was aware of the patient’s group assignment. The sample size was calculated on the basis of the primary outcome measure (Barthel Index) with ≥95 defined as a success using an α of 0.05 and a power of 80%. Predicted successful response rates based on the prior dose finding citicoline trial were 49% for citicoline and 33% for placebo. All patients were required to be inpatients at the start of the study medication treatment but could be discharged at any time with oral treatment continued through the remainder of the 6 weeks. Patients were seen by study personnel at baseline, 1 week, hospital discharge, 3 weeks, 6 weeks, and 12 weeks, at which time a side effect profile, drug accountability, and required efficacy measures were completed.

The primary outcome measure of this study was functional outcome as determined by the Barthel Index at 12 weeks. For the primary analysis, it was a priori determined that the Barthel Index would be classified into 5 strata (death or zero, 5 to 40, 45 to 60, 65 to 80, 85 to 100). Logistic regression analysis was to be used to determine whether there were significant differences between the 2 treatment groups in the distribution of patients within these 5 strata. It was further determined a priori that to decrease the impact of any baseline differences in stroke severity, the baseline NIHSS score would be used as a covariate in the primary analysis. Secondary prespecified outcome measures included (1) assessments of categorized Barthel Index at the other weeks, (2) percentage of patients who had a full recovery (Barthel Index 95 or 100 at 12 weeks), (3) assessment of treatment differences on the Modified Rankin scale, and (4) assessment of treatment differences on neurological,  

![Figure 1. A, Diagrammatic illustration of normal synthesis of phosphatidylcholine. B, Effect of ischemia on this reaction. C, Citicoline treatment reverses increased fatty acid formation and loss of phosphatidylcholine.](image-url)
behavioral, and cognitive function by use of the NIHSS and Mini-Mental State Examination, (5) assessment of mortality, (6) assessment of treatment differences in the percentage of patients who had a full recovery defined as an NIHSS score of \( \leq 1 \), (7) assessment of number of days from stroke to hospital discharge, (8) determination of the relative rate of improvement for the Barthel Index, Rankin scale, and NIHSS between the groups. Additional post hoc outcome analyses were performed on a subgroup of patients with moderate to large strokes.

All analyses were done with the last observation carried forward (LOCF) for those patients who discontinue early. All analyses were based on the intention-to-treat (ITT) sample. A patient qualified for inclusion in the ITT sample if he/she entered the double-blind phase of the study and was evaluated at least once or died since receiving double-blind medication.25

Figure 2 shows the trial profile for the study. All a priori treatment comparisons were declared statistically significant if the \( p \leq 0.05 \). All of the analyses presented used the Cochran-Mantel-Haenszel procedure, with center as the stratification variable.

**Results**

Three hundred ninety-four patients were enrolled in the study from June 30, 1996, to June 12, 1997. The randomization blinding was successful, with treatment unblinding occurring in only 1 patient. All analyses were based on the ITT population (see Figure 2). Table 2 gives the baseline characteristics for the 2 groups. There was no significant difference noted in terms of patient age, sex, hemisphere involved by the stroke, or time to treatment, stroke subtype, or preexisting medical condition. Although the mean baseline NIHSS was similar between groups, there was a significantly higher percentage of patients with mild strokes (baseline NIHSS <8) in the placebo groups (34%) compared with citicoline (22%) \( P<0.01 \). This baseline imbalance may have impacted the overall efficacy results in this trial.

### Table 2. Baseline Characteristics: Total Patient Population

<table>
<thead>
<tr>
<th></th>
<th>Placebo</th>
<th>500 mg Citicoline</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients, n</td>
<td>127</td>
<td>267</td>
</tr>
<tr>
<td>Age, y, mean</td>
<td>71</td>
<td>70</td>
</tr>
<tr>
<td>Sex, % men</td>
<td>49%</td>
<td>46%</td>
</tr>
<tr>
<td>Stroke hemisphere, % left</td>
<td>46%</td>
<td>48%</td>
</tr>
<tr>
<td>Time to treatment, h, mean</td>
<td>12.3</td>
<td>11.7</td>
</tr>
<tr>
<td>Weight, kg, mean</td>
<td>79.5</td>
<td>77.7</td>
</tr>
<tr>
<td>Baseline NIHSS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>12.7</td>
<td>13.3</td>
</tr>
<tr>
<td>4–7</td>
<td>34%</td>
<td>22% ( P&lt;0.01 )</td>
</tr>
<tr>
<td>8–11</td>
<td>20%</td>
<td>27%</td>
</tr>
<tr>
<td>12–18</td>
<td>23%</td>
<td>28%</td>
</tr>
<tr>
<td>19–32</td>
<td>23%</td>
<td>23%</td>
</tr>
<tr>
<td>Preexisting medical conditions, %</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypertension</td>
<td>72%</td>
<td>70%</td>
</tr>
<tr>
<td>Previous stroke/TIA</td>
<td>37%</td>
<td>35%</td>
</tr>
<tr>
<td>Heart disease</td>
<td>42%</td>
<td>40%</td>
</tr>
<tr>
<td>Stroke subtype*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Small vessel</td>
<td>26%</td>
<td>22%</td>
</tr>
<tr>
<td>Large vessel</td>
<td>43%</td>
<td>38%</td>
</tr>
<tr>
<td>Cardioembolic</td>
<td>24%</td>
<td>31%</td>
</tr>
<tr>
<td>Thromboembolic</td>
<td>5%</td>
<td>6%</td>
</tr>
<tr>
<td>Other</td>
<td>5%</td>
<td>6%</td>
</tr>
</tbody>
</table>

*TOAST criteria.31

The primary efficacy analysis was the Barthel Index score classified into 5 strata analyzed by use of logistic regression analysis, with the baseline NIHSS score used as a covariate. The distribution of Barthel Index scores at 12 weeks is shown in Table 3. Unfortunately, the distribution of patients failed the proportional odds assumption and the primary planned analysis was rendered unreliable and was therefore not completed. Table 4 shows results for some of the planned secondary end points for the study. In the overall population, no treatment benefit was seen on any of the planned secondary end points, even when the baseline NIHSS was used as a covariant. However, the study was only powered to detect differences on the basis of the primary measure and not these secondary outcome measures. To determine if citicoline may be beneficial for a subgroup of patients, post hoc analyses were performed by quartiles of the NIHSS scores. These post hoc analyses were not prespecified and involved multiple

### Table 3. Distribution of Patient Barthel Index Scores at Week 12: ITT Patient Population, Number (%) of Patients LOCF

<table>
<thead>
<tr>
<th>Categorized Barthel Index, 12 wks</th>
<th>Placebo (n=127)</th>
<th>500 mg Citicoline (n=267)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Death</td>
<td>18%</td>
<td>17%</td>
</tr>
<tr>
<td>0</td>
<td>5%</td>
<td>6%</td>
</tr>
<tr>
<td>5–40</td>
<td>9%</td>
<td>12%</td>
</tr>
<tr>
<td>45–60</td>
<td>8%</td>
<td>8%</td>
</tr>
<tr>
<td>65–80</td>
<td>15%</td>
<td>10%</td>
</tr>
<tr>
<td>85–100</td>
<td>45%</td>
<td>46%</td>
</tr>
</tbody>
</table>

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TABLE 4. Secondary Efficacy Analysis at Week 12: ITT Patient Population, % of Patients LOCF

|                          | Placebo (n=127) | 500 mg Citicoline (n=267) | P
|--------------------------|----------------|--------------------------|---
| Death                    | 18%            | 17%                      | NS
| Barthel ≥95              | 40%            | 40%                      | NS
| Rankin ≤1                | 25%            | 27%                      | NS
| NIHSS ≤1                 | 24%            | 27%                      | NS
| Hospital stay: d, median | 7.5            | 7.5                      | NS

Table 6 shows the effect of citicoline treatment on mortality and serious adverse events. There were no patient deaths reasonably attributed to the study medication in this trial. There were no serious adverse events (SAEs) reasonably attributed to the study medication and there were no between-group differences in the incidence of any of the SAEs. The higher incidence of central nervous system SAEs (11% vs 19%; P>0.1) reflected a few patients in the citicoline groups having recurrent strokes that were believed to be unrelated to the drug. In terms of other side effects, citicoline appears to be remarkably well tolerated with no adverse event being significantly more frequently seen in the citicoline group compared with placebo. The increased incidence of accidental falling and dizziness reported in the prior 001A dose-finding study was not seen in the current study.

Table 6. Serious Adverse Events: Total Patient Population, % of Patients

|                          | Placebo (n=127) | 500 mg Citicoline (n=267) | P
|--------------------------|----------------|--------------------------|---
| Death                    | 18%            | 17%                      | NS
| Cardiovascular           | 19%            | 16%                      | NS
| Central nervous system   | 11%            | 19%                      | NS
| Respiratory              | 12%            | 13%                      | NS
| Gastrointestinal         | 3%             | 4%                       | NS
| Metabolic                | 2%             | 2%                       | NS
| Urologic                 | 1%             | 5%                       | NS
| Heme/lymphatic           | 1%             | 2%                       | NS
| Musculoskeletal          | 1%             | 2%                       | NS
| Skin/appendages          | 1%             | 0%                       | NS
| Body as a whole          | 9%             | 9%                       | NS

Discussion

This study failed to find a benefit for citicoline on any of the planned efficacy end points in the overall study population. These results may have been confounded by a significant baseline imbalance in the baseline stroke severity between groups. This illustrates that despite proper randomization, important baseline imbalances can occur in studies when relatively small numbers of patients are used. The response rate (Barthel Index ≥95) in the control group overall was also higher than predicted in our sample size calculations (33% vs 40%); this may have increased the chance of a type II error occurring. We were also unable to perform the primary planned analysis in this study, a logistic regression based on a categorized Barthel Index, because of nonproportional distribution of patients. For this reason, future citicoline trials will instead utilize a responder analysis on the basis of the NIHSS. Overall citicoline was very well tolerated, with no reported side effect being significantly higher than in the placebo group.

A very high rate of spontaneous recovery regardless of treatment was seen in mild patients with NIHSS <8, suggesting that it would be very difficult to detect significant treatment effects in patients with mild stroke (high likelihood of a type II error). In patients with moderate-to-severe strokes defined by NIHSS ≥8, this study found potential treatment effects with citicoline on several of the efficacy end points measuring full recovery. Interestingly, the same <8 and ≥8 NIHSS cutoff has recently been found to be an important predictor of early stroke recovery.26 In this study, the investigators found that 45% of patients with initial NIHSS <8 were fully recovered (NIHSS 0,1) at 48 hours compared with only 2% of those with baseline NIHSS of ≥8. On the basis of the high spontaneous recovery results seen in these 2 studies, it appears that patients with an NIHSS of <8 probably should not be enrolled in clinical therapeutic trials until more sensitive measures of outcome are developed.

The current study failed to confirm the results of the previous citicoline dose-finding US trial in the overall population studied.21 That study (001A), involving 259 patients, found that both 500 mg and 2000 mg but not 1000 mg of citicoline produced a
significant treatment effect compared with placebo when baseline differences in the NIHSS score were used as a covariate. Because the 2000 mg and 500 mg groups appeared to have equal efficacy and because the 2000 mg group had a higher incidence of mild dizziness, it was decided to use the 500 mg group in the current study. However, after the importance of baseline differences in stroke severity was found in the current study, the previous 001A trial was reanalyzed. It was found that there was a significant imbalance in the 500 mg group; this time with the 500 mg treatment group having a higher percentage of mild strokes (47% vs 38%). In contrast, the stroke severity of the 2 groups was well matched for the 2000 mg group and placebo (35% vs 38%). It therefore appears that the 2000 mg dose may provide the greatest likelihood of therapeutic effect. For this reason, the 2000 mg dose has been chosen for use in a further confirmatory efficacy trial.

The safety and beneficial effects seen with citicoline in moderate-to-severe strokes in the current study are similar to those that have been seen in other randomized, non-US trials. Goyas et al17 studied the effect of intravenous citicoline (750 mg/d for approximately 10 days) within 48 hours of stroke onset in a double-blind, placebo-controlled trial. Citicoline patients showed a significant improvement on a quantified neurological assessment scale rating motor strength, muscular force, sensation, higher cortical function, and ambulation at 90 days. Patients treated with citicoline were significantly (P=0.02) more likely to be ambulatory compared with placebo-treated patients at 90 days (60% vs 24%). A weakness of this study is that it did not use currently accepted measures of neurological evaluation or functional outcome. A second double-blind, placebo-controlled trial evaluating intravenous citicoline (250 mg TID for 10 days) in patients treated within 48 hours of their symptoms was done by Boudouresques and Michel.18 Most of the patients were severely impaired at baseline. With the use of a 5-point recovery rating scale, this study found that a significantly higher percentage of patients had a very good to fairly good recovery with citicoline versus placebo treatment at 10 days after stroke (79% vs 44%). Both of these studies found significantly more fully recovered patients (52% and 48%, respectively) receiving citicoline as compared with placebo (24% and 9%, respectively). In a small double-blind, placebo-controlled study, Corso et al19 treated patients with acute stroke (48 hours) with either 1000 mg/d of intravenous citicoline or placebo for 30 days. In comparison to their baseline assessments, 76% of the citicoline-treated patients demonstrated improvement compared with only 31% of the placebo-treated patients (P<0.01). The largest multicenter, double-blind, placebo-controlled study investigating citicoline in ischemic stroke was completed by Tazaki et al20 in Japan. In this study, patients were treated within 14 days of onset of ischemic stroke symptoms with either 1000 mg/d of intravenous citicoline (n=133) for 14 days or placebo (n=139). A 6-category global improvement rating scale was used as a key end point. The study found that citicoline-treated patients had a significant improvement on day 14 in terms of the global improvement rating scale, with 52% of the citicoline treated patients showing a significant improvement compared with only 26% for those receiving placebo (P<0.01). A major weakness of this publication is that it did not describe in detail the long-term functional outcome results on these patients.

In the above trials, the incidence of side effects associated with citicoline treatment was low. In these placebo-controlled trials, only headache, vertigo, and dizziness were observed more frequently in the citicoline-treated patients compared with placebo (headache: 2.2% citicoline vs 0.3% placebo; vertigo: 1.2% citicoline vs 0.7% placebo; dizziness: 1.0% citicoline vs 0.2% placebo). A direct comparison of these trials with the current study is difficult because of differences in patient populations and efficacy end points. However, they appear to support the possible efficacy of citicoline in both functional and cognitive recovery after stroke.

It is interesting to compare the results and patient demographics of this study with other recent randomized stroke treatment trials. The baseline stroke severity in the current study (mean 13) is similar to that seen in the National Institute of Neurologic Disorders under 3-hour rt-PA trial (mean 14).27 The rate of spontaneous full recovery on the Barthel Index is also very similar, being 40% in our trial and 39% in the NINDS trial placebo group. Our baseline stroke severity was higher than that in the recently reported ECASS II and ATLANTIS rt-PA trials, both of which had an NIHSS mean of 11.28,29 The high rate of spontaneous full recovery (Barthel Index 53%) as the result of relative mild strokes has been offered as an explanation for the failure of these two trials to find a treatment benefit. Perhaps the most relevant comparison is that of the recent intra-arterial Prourokinase trial (PROACT II)30 to our >7 NIHSS subgroup. Both trials targeted middle cerebral artery (MCA) strokes, although, unlike the PROACT II trial, we did not confirm MCA occlusion. The baseline NIHSS severity was similar in the two populations (PROACT II 17; ≥8 group 15 as was the low rate of full recovery in the placebo groups: PROACT II 32%; ≥8 group 21%). Direct comparisons of treatment benefits are difficult, but both trials report an ~9% absolute potential increase in the rate of full recovery.

In summary, this study suggests that citicoline can be safely used in acute stroke treatment with little or no side effects. Overall, there were no significant differences between citicoline and placebo on the planned efficacy analysis in the total patient cohort treated in this trial. However, citicoline may be beneficial in a subgroup of patients with moderate-to-severe strokes. A large efficacy trial trying to confirm this promising effect in patients with moderate-to-severe stroke is currently in progress in North America.

Appendix

US Citicoline Stroke Treatment Study IP302-007

Participating Centers

Oregon Health Sciences Center, Portland: Wayne M. Clark, MD, Helmi L. Lutsep, MD, Anne Doherty, RN; 53 patients. Thomas Jefferson University, Philadelphia, Pa: Rodney D. Bell, MD, Jamie Strauss, RN; 9 patients. University of Rochester Medical Center, Rochester, NY: Curtis Benesch, MD, Justine Zentner, MSN, NP; 9 patients. Central Baptist Hospital, Lexington, Ky: William Brooks, MD, Patty Howard, RN, Cyndi Baxter, RN; 15 patients. Fairfax Hospital, Falls Church, Va: David B. Grass, MD, Maureen Burke, RN; 5 patients. Wayne State University, Detroit, Mich: Seemant Chaturvedi, MD, Bryan D. Bertasio, RN; 8 patients. Cox Medical Center, Springfield, Mo: James E. Duff, MD, Tracy Vaughn, RN; 3 patients. Southwestern Vermont Medical Center, Bennington: Keith R. Edwards, MD, Cindy Lewis; 11 patients. Genesis Health Care
Acknowledgments
The authors would like to thank Valerie Roska for her expert assistance in the preparation of the manuscript.

References
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Stroke. 1999;30:2592-2597
doi: 10.1161/01.STR.30.12.2592

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http://stroke.ahajournals.org/content/30/12/2592