ORIGINAL CONTRIBUTION

Prognostic Value of Proton Magnetic Resonance Spectroscopy in Ischemic Stroke

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Objective: To determine the correlation between metabolite concentrations and clinical outcome during the acute or subacute phase of ischemic stroke by using single-voxel localized proton magnetic resonance spectroscopy (^{1}H-MRS).

Setting: A university hospital neurologic department.

Patients and Methods: Combined single-voxel ^{1}H-MRS and magnetic resonance imaging were performed on 26 patients with a recent ischemic stroke (on 8 patients during the first 24 hours after the stroke and on 18 during the first week). For all patients, the signals from *N*-acetylaspartate, choline-containing compounds, and creatine-phosphocreatine were compared with those on the contralateral side as peak area ratios. The data for ^{1}H-MRS were related to scores on the Scandinavian Stroke Scale and the Barthel Index at a 6-month clinical follow-up.

Results: The signals from *N*-acetylaspartate, choline-containing compounds, and creatine-phosphocreatine were significantly reduced in all infarcted areas ($P<.001$, $P<.001$, and $P=.003$, respectively, Wilcoxon signed rank test). A lactate signal was present in 19 patients. The statistical analysis showed a significant positive correlation between *N*-acetylaspartate signals and Scandinavian Stroke Scale scores and between reduction of *N*-acetylaspartate signals and Barthel Index scores (Spearman rank correlation test). Patients in whom lactate was present had Scandinavian Stroke Scale scores significantly lower than patients in the group without lactate (Mann-Whitney *U* test).

Conclusion: Single-voxel ^{1}H-MRS performed during the acute or subacute phase of ischemic stroke may provide prognostic information.

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THE DEVELOPMENT of new therapeutic strategies for acute stroke requires that clinicians have a better understanding of the pathophysiological mechanisms operating during severe cerebral ischemia as well as of the improvements in the diagnostic tools available for the management of patient care. Magnetic resonance imaging (MRI) is highly sensitive for the early detection of ischemic brain lesions.[1,2] Nevertheless, clinical evolution probably depends on the extent of neuronal loss, which cannot be determined directly from the images. New magnetic resonance techniques, such as diffusion-weighted imaging[3] and proton magnetic resonance spectroscopy (^{1}H-MRS), may, however, have considerable prognostic value.

In recent years, single-voxel ^{1}H-MRS and spectroscopic imaging have been widely applied to the study of acute ischemic cerebrovascular disease to provide information about neuronal viability and the metabolic state of the infarcted tissue.[4-14] Reduced *N*-acetylaspartate (NAA) levels and elevated lactate levels are readily detectable in the ischemic lesions. Loss of NAA reflects neuronal death and can begin as early as 2 hours after the onset of focal infarction, as shown in studies of animals.[15,16] Moreover, in some studies of humans, the levels of NAA showed additional decline during the subacute phase, probably related to further neuronal loss in the ischemic penumbra.[4,5,17] High lactate levels can persist a long time after the occurrence of the stroke, but only during the first 24 to 48 hours are they ascribable to anaerobic metabolism.[18]

Few studies have compared ^{1}H-MRS findings with clinical measures to evaluate their potential prognostic value.[5,11,19] In a previous study, with fewer patients, Federico et al[10] found a positive correlation between reduced levels of NAA and the clinical condition, evaluated by using the Scandinavian Stroke Scale (SSS) in which higher scores indicate better func-

PATIENTS AND METHODS

The study included 26 patients (11 women and 15 men; mean±SD age, 61.0±14.6 years; range, 26-78 years) who had an ischemic stroke in the area of the middle cerebral artery. All had cortical or subcortical lesions greater than 25 mm in diameter. All patients had a new motor or speech deficit and were first examined by combined MRI and single-voxel ^{1}H-MRS between 8 hours and 7 days after the onset of symptoms (mean±SD, 3.5±2.1 days). Of the patients, 20 underwent another examination from 8 to 121 days after the stroke (mean±SD, 59.6±38.5 days). The patients judged unable to participate in the examination protocol were excluded, and a few patients were given mild sedation by an anesthetist. Informed consent was obtained from all patients or from the closest relative, and the experimental protocol was approved by the Neurology Department Ethics Committee of the University of Bari, Bari, Italy. A neurologic examination was performed by 1 of us (G.L.), who was unaware of the spectroscopic results from the first and second ^{1}H-MRS examinations and the 6-month follow-up, except for 2 patients who died of cerebral causes, 1 after 7 days, and the other after 24 days. Measurement of the neurologic deficit was assessed by means of the long-term SSS.The functional outcome after the stroke was determined at the 6-month follow-up by using the Barthel Index score in which lower scores indicate the worst disability.[20,21]

The MRI and ^{1}H-MRS were performed with a whole body 1.5-T iron-shielded system (Magnetom 63 SP, Siemens AG, Erlangen, Germany) using a standard circularly polarized head coil. The imaging protocol consisted of sagittal and coronal T_1-weighted spin-echo sequences (repetition time [TR], 600 milliseconds; echo time [TE], 15 milliseconds) and transverse T_2-weighted sequences (TR, 2200 milliseconds; TE, 80 milliseconds). The slice thickness was 5 mm and the matrix, 256×256. After global shimming performed with a standard nonselective shimming sequence, the volumes of interest (VOI) were localized in the ischemic area, taking care to avoid the inclusion of normal tissue or cerebrospinal fluid, and in the corresponding nonaffected contralateral region. The VOIs, ranging between 8 and 16 mL, were targeted from T_2-weighted scans. Local shimming within the selected VOI was required to obtain a spectral width of half of the maximum of the water proton peak of 3 to 6 Hz. The water proton signal was suppressed by a preceding chemical shift–selective radiofrequency pulse.[22] The proton spectra were acquired by means of a double spin-echo sequence with TE, 135 milliseconds; TR, 1500 milliseconds; and 256 acquisitions (necessary to obtain 1 spectrum). The total examination time for the MRI and the ^{1}H-MRS was less than 60 minutes. The signals in the time domain were multiplied by a half gaussian function with a half-width of 256 milliseconds and by a factor of 100. After Fourier transformation and zero-order phase correction, the areas under the peaks were obtained by numerical integration. Baseline correction was performed for the purpose of presentation. Postprocedure processing was always performed by the same investigator (D.M.M.). At subsequent examinations, anatomic landmarks in the images were used to place the VOI in the same location as before. Resonances were assigned as follows: choline-containing compounds (Cho), 3.2 ppm; creatine-phosphocreatine (Cr), 3.0 ppm; NAA, 2.0 ppm; and lactate, 1.3 ppm.[23] It is difficult to measure the absolute values with our technique; rather, results are obtained as ratios of metabolite signals. Nevertheless, with the same measurement variables (TR and TE), the results of clinical ^{1}H-MRS are relatively reproducible. As previously observed, the levels of all metabolites are reduced during the acute and subacute phases of ischemic stroke.[4,6,8,24,25] This effectively limits the usefulness and reliability of ratios obtained comparing metabolites from the infarcted area. Thus, we compared the peak areas of NAA, Cr, and Cho from infarcted areas with those of the contralateral normal regions in the same patients. Therefore, the spectrum from the contralateral region served as the control for the patient. We also included 9 healthy volunteers (mean±SD age, 60.7±15.3 years; range, 27-75 years) as control subjects in the study for comparison of the mean metabolite signals between patients and healthy persons. In the healthy volunteers, the ratios for the metabolite signals from corresponding regions in the right and left hemispheres were almost identical. We chose a long TE (135 milliseconds) to minimize potential contamination of the signal by lipids, which have a very short T_2. In addition, the long TE allows acquisition of a signal from the lactate methyl groups in an antiphase condition doublet (spin-spin coupling constant, 7.35 Hz). Some of the ^{1}H-MRS data, analyzed in a different manner, have been reported.[10,24]

The results were evaluated statistically by using a 5% significance level for the following nonparametrical methods: the Wilcoxon signed rank test for comparison of paired samples, the Mann-Whitney *U* test for comparison of unpaired samples, and the Spearman rank correlation test.

tion, during the first week after the stroke and at follow-up. We conducted the present study to evaluate the correlation between metabolite variations during the acute and subacute phases of stroke and the clinical outcome at 6 months.

RESULTS

The ^{1}H-MRS and data are shown in the **Table**. **Figure 1** and **Figure 2** show T_2-weighted images and the ^{1}H-MRS spectra for 2 of the patients.

At the first examination, the peak area signals for NAA, Cr, and Cho were significantly reduced in the infarcted areas compared with the contralateral regions ($P<.001$, $P<.001$, $P=.003$, respectively; Wilcoxon rank sum test). The peak area ratios for NAA, Cr, and Cho between the ischemic (i) area and the contralateral (c) side (iNAA/cNAA, iCr/cCr, and iCho/cCho) showed a positive correlation with the SSS scores ($P<.001$, $P=.02$, $P=.01$, respectively.

At the second examination, a significant reduction of the NAA and Cr signals persisted ($P<.001$ and $P=.01$, respectively), but with a significant positive correlation between only the SSS scores and the iNAA/cNAA ratio ($P=.01$). Nevertheless, the iNAA/cNAA ratio, obtained during the first examination, was positively and significantly correlated with the SSS scores at the 6-month follow-up ($P=.01$) (**Figure 3**). The Barthel Index scores

Proton Magnetic Resonance Spectroscopy (^{1}H-MRS) and Clinical Data*

Patient No./ Sex/Age, y	First Examination						Second Examination						Clinical Follow-up	
	^{1}H-MRS Data					SSS Score	^{1}H-MRS Data					SSS Score		
	Day	iNAA/cNAA	iCr/cCr	iCho/cCho	Lac		Day	iNAA/cNAA	iCr/cCr	iCho/cCho	Lac		SSS Score	BI Score
1/F/71	1	0.01	0.01	0.01	+	0	†	NA	NA	NA	NA	NA	NA	NA
2/M/77	1	0.01	0.43	0.62	+	10	8	0.05	0.07	0.04	+	20	34	55
3/M/55	1	0.05	0.07	0.07	+	32	95	0.26	0.47	0.90	+	51	51	90
4/M/56	1	0.37	0.40	0.70	+	20	67	0.70	0.46	1.52	–	48	48	100
5/M/55	1	0.53	0.68	0.64	+	37	120	0.87	0.79	0.33	–	48	48	100
6/F/72	1	0.57	0.77	0.93	+	12	78	0.01	0.25	0.50	+	21	21	25
7/F/71	1	0.61	0.90	0.79	+	21	71	0.50	0.61	0.83	–	44	44	100
8/M/61	1	0.80	1.21	0.93	–	36	57	0.97	1.17	1.09	–	42	51	70
9/F/55	2	0.96	1.04	1.22	+	51	NA	NA	NA	NA	NA	NA	56	100
10/F/33	3	0.31	0.76	0.74	+	34	120	0.24	0.66	0.89	+	44	54	100
11/M/61	3	0.33	0.51	0.52	+	32	59	0.20	0.43	0.27	+	44	44	100
12/M/50	3	0.69	1.05	1.17	–	30	12	0.87	1.05	1.10	–	37	37	30
13/M/57	3	0.94	0.85	1.21	–	56	NA	NA	NA	NA	NA	NA	58	100
14/M/78	4	0.18	0.19	0.61	+	29	121	0.35	0.61	0.48	+	38	38	85
15/F/26	4	0.21	0.45	0.41	+	11	110	0.15	0.52	0.56	+	33	33	60
16/F/54	4	0.43	0.50	0.54	+	43	NA	NA	NA	NA	NA	NA	50	60
17/F/28	4	0.58	1.00	1.15	+	34	28	0.43	1.27	1.88	–	41	51	70
18/F/65	4	0.68	0.29	0.47	+	29	29	0.62	0.52	0.76	–	46	46	100
19/M/67	5	0.32	0.73	0.50	–	38	39	0.21	0.83	0.92	–	51	53	75
20/F/74	5	0.52	1.03	1.06	–	27	25	0.36	0.93	1.68	–	31	40	55
21/M/71	5	0.80	1.09	1.40	–	50	12	0.93	1.14	1.47	+	52	58	100
22/F/57	6	0.29	0.40	0.63	+	33	55	0.14	0.20	0.70	+	37	39	70
23/M/78	6	0.45	0.51	0.77	+	37	69	0.27	0.66	1.01	–	36	36	40
24/M/76	7	0.01	0.01	0.14	+	10	†	NA	NA	NA	NA	NA	NA	NA
25/M/65	7	0.09	0.99	0.93	+	18	16	0.09	0.14	0.81	+	22	27	20
26/M/74	7	0.92	0.83	0.97	–	46	NA	NA	NA	NA	NA	NA	54	100

**iNAA indicates* N-acetylaspartate from infarcted area; cNAA, N-acetylaspartate from contralateral area; iCr, creatine-phosphocreatine from infarcted area; cCr, creatine-phosphocreatine from contralateral area; iCho, choline-containing compounds from infarcted area; cCho, choline-containing compounds from contralateral area; Lac, lactate; SSS, Scandinavian Stroke Scale; BI, Barthel Index; +, present; –, absent; and NA, not assessed. Data for the ^{1}H-MRS, except for lactate, are expressed as ratios.*

†*Deceased.*

at 6 months correlated significantly with the iNAA/cNAA ratio values (P=.01).

Lactate was present in 19 of 26 patients at the first examination. The SSS scores at 6 months were significantly lower in patients with lactate present than in patients without lactate (P=.049).

COMMENT

Several reports about the results of ^{1}H-MRS in human stroke have been published.* These studies sufficiently outlined the metabolic variations in the acute, subacute, and chronic phases of cerebral ischemia. In particular, a loss of NAA and reduced levels of Cho and total Cr have been found in acute infarctions. A marked increase in the lactate level was generally observed in the infarcted area.[4,6,8,14,17,25,26] Nevertheless, few studies have been performed to assess the prognostic value of ^{1}H-MRS metabolic information.[5,10,11,19] Ford et al[5] used chemical shift imaging to examine 8 patients after a stroke. They reported that the patients with the best recoveries had relatively preserved NAA, Cr, and Cho peaks. Gideon et al,[19] in a preliminary study, found no clear correlation between the levels of NAA and lactate during the acute phase of stroke and the clinical outcome. However, these studies involved too few patients to determine the significance of prognostic information. In a spectroscopic imaging study, Graham et al[11] stated that the lactate and NAA signals from the ischemic lesions were correlated with clinical measures of disability and functional outcome at the time patients were discharged from the hospital. These interesting results may be questionable because the time until examination after the stroke varied substantially (from 11 hours to 19 days), and the follow-up period, which did not exceed 35 days, may be too short for an adequate evaluation of the conditions of patients after a stroke. In a previous report about 14 patients who were serially examined during the first week and during the chronic phase, Federico et al[10] found a positive correlation between low NAA levels and the SSS scores.

The present study is the first performed in patients after stroke that uses ^{1}H-MRS with a clinical follow-up at 6 months. The metabolic changes in our patients were not different from those reported in previous studies.[4,6,8,12,13,27]

In the acute and subacute phases, we found a more marked reduction in the NAA level in the patients with the worst neurologic conditions. The reduction in the NAA level was related to an unfavorable clinical out-

* *References 4-6, 8, 10, 11, 14, 17, 19, 25, 26.*

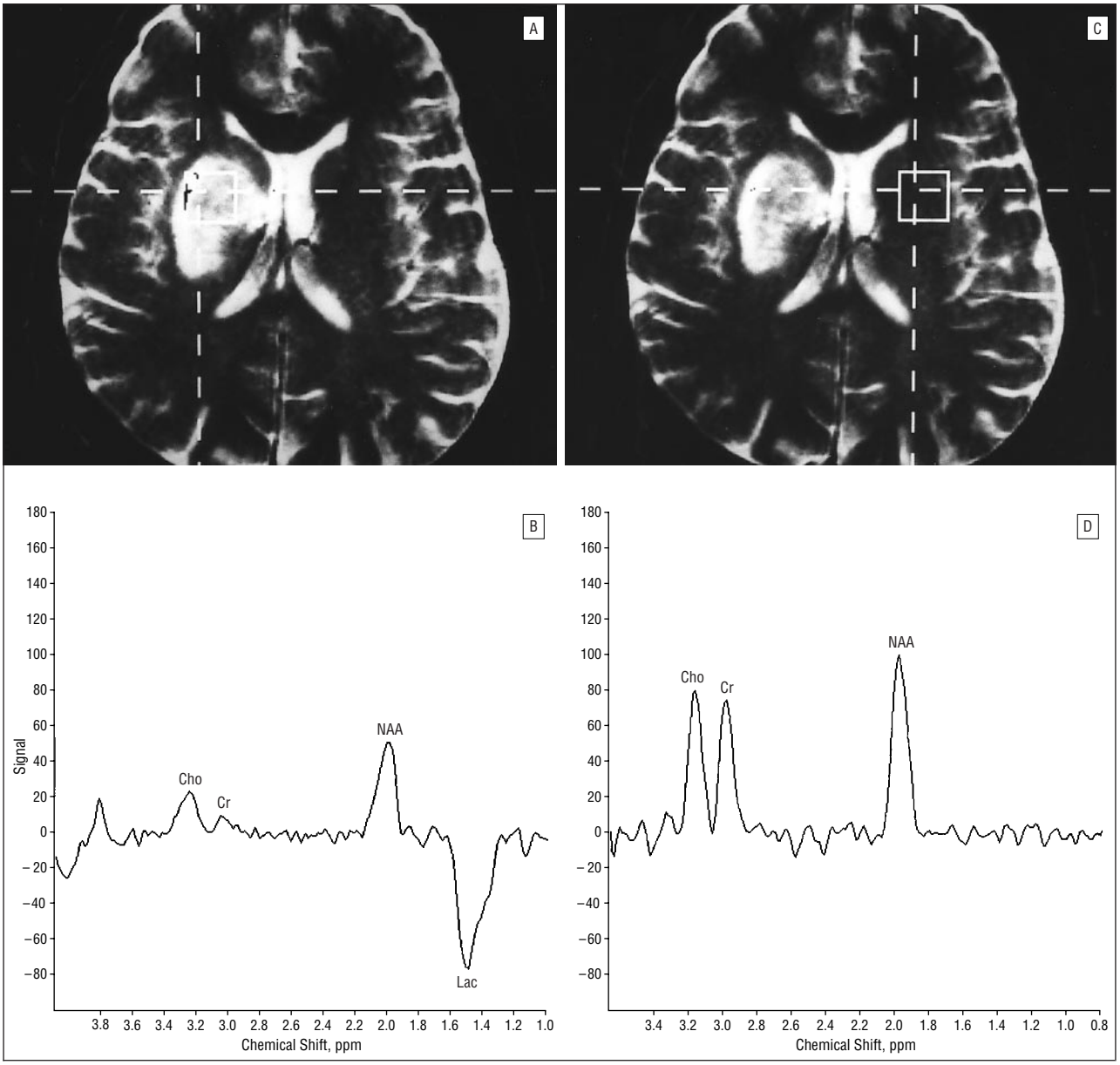

Figure 1. *A, T_2-weighted image of patient 18, 4 days after the onset of left hemiparesis, showing an area of increased signal in the right capsular region. B, Localized proton magnetic resonance spectroscopy (^{1}H-MRS) spectrum shows substantial lactate (Lac) and marked decreases in the levels of creatine-phosphocreatine (Cr), choline-containing compounds (Cho), and* N*-acetylaspartate (NAA) compared with the spectrum of a normal contralateral region (C, T_2-weighted image; D, ^{1}H-MRS result).*

come at 6 months. It is now generally accepted that the decrease in the NAA level indicates loss of neurons or neuronal function.[28-30] A more pronounced reduction of the level of NAA inside ischemic lesions, apparently evident to a similar degree in MRI, can suggest a more serious neuronal loss. Despite the presence of similarly sized MRI lesions, evident as T_2 hyperintensity usually found in the area of the middle cerebral artery, clinical evolution was greatly variable. One possible explanation for this variability is that while ^{1}H-MRS may detect only neuronal loss, MRI hyperintensity may be influenced by cellular edema.

At the first examination, the decrease in the level of NAA was accompanied by a decrease in the levels of Cho and Cr; the decreased levels of Cho and Cr had a positive correlation with the SSS scores. At the second examination, this correlation was no longer evident. We found an increase in the level of Cho in many patients who underwent a second examination; the increased levels may be caused by an elevated turnover of membrane lipids, as indicated by other authors.[2,17] The levels of Cr showed less marked variations, but the variations were probably sufficient to explain the absence of correlation with the SSS scores. Our data for the levels of Cho and Cr are similar to those reported previously.[4,12-14]

Lactate was present in only 19 of 26 patients at the first examination and in 10 of 20 patients at the second examination. The relatively low number of patients with lactate present during the acute and subacute phases needs explanation. Five patients (patients 12, 13, 19, 20, and

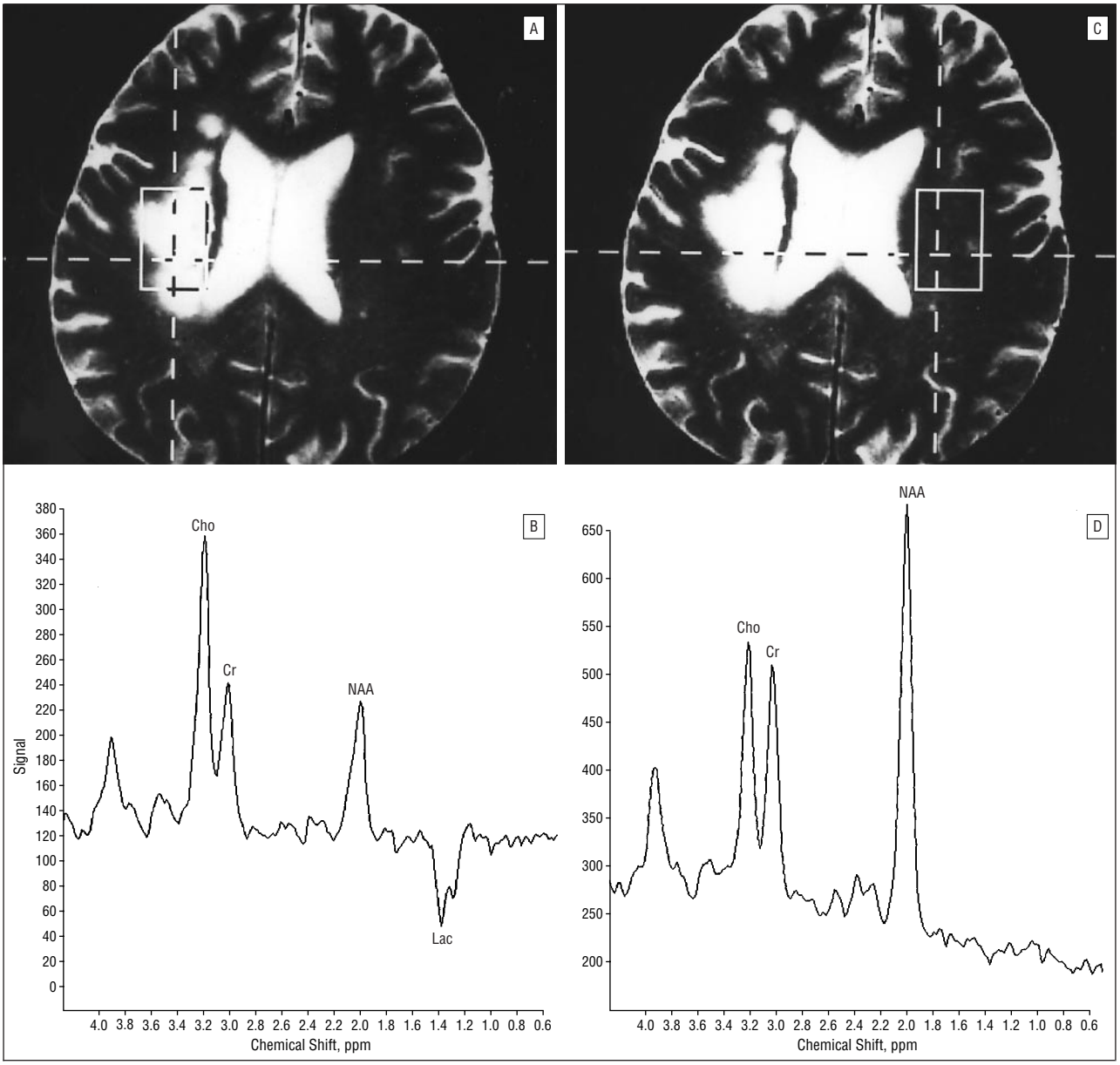

Figure 2. *A, T_2-weighted image at the second examination of patient 3, 3 months after the onset of severe left hemiparesis showing an area of increased signal in the right subcortical region. B, Localized proton magnetic resonance spectroscopy (^{1}H-MRS) spectrum shows a persistence of lactate (Lac) and a marked decrease of* N*-acetylaspartate (NAA) and creatine-phosphocreatine (Cr) compared with the spectrum of a contralateral region (C, T_2-weighted image; D, ^{1}H-MRS result). Cho indicates choline-containing compounds.*

21) were examined between the third and the fifth days, probably too late to show lactate. It has been reported that the lactate signal tends to reduce progressively after stroke.[12,13] Two other patients (patients 13 and 26) without a lactate signal had high levels of NAA and a complete neurologic recovery. Only 1 patient examined on the first day had no lactate signal (patient 8), but the same patient showed a relatively preserved NAA signal and a good recovery. In this study, we were unable to relate the presence of lactate with the long-term clinical outcome in the individual patient. However, the patients showing the presence of lactate during the acute and subacute phases had worse SSS scores compared with the SSS scores of the patients without lactate. This significant difference was no longer evident at the ^{1}H-MRS examination during the chronic phase. Our study results seem to confirm that lactate that is detected early can have a predictive value in patients who have had a stroke.

These results indicate that spectroscopy performed as soon as possible after stroke can have a prognostic value, especially when the reduction in the level of NAA and the presence of lactate are considered. It seems possible to recognize 2 groups of patients. The first group, characterized by lower NAA levels and a lesser likelihood for the presence of lactate, has an unfavorable neurologic outcome. The second group includes patients with NAA levels that are relatively preserved, no lactate signal, and a good recovery. This assertion is strongly supported by a recent experimental study in rats that seems to confirm the predictive value of these metabolic variations dur-

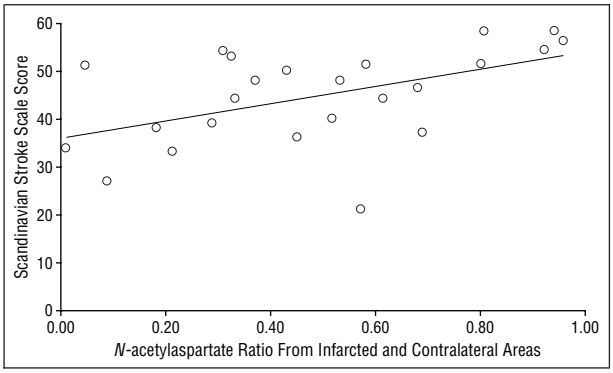

Figure 3. *Plot of the* N*-acetylaspartate ratio from infarcted and contralateral areas at first examination vs Scandinavian Stroke Scale scores at the 6-month follow-up.*

ing the acute phase of focal ischemia.[16] New studies, with a larger number of patients examined immediately after the stroke, are necessary to confirm our preliminary data.

Single-voxel ^{1}H-MRS performed during the acute or subacute phase of ischemic stroke may provide prognostic information and be useful for the evaluation of therapeutic interventions.

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