

Safety and efficacy of sonothrombolysis using bilateral TCD monitoring by diagnostic 2 MHz probes – a pilot study

Petr Bardon^a, Martin Kuliha^b, Roman Herzig^c, Petr Kanovsky^c, David Skoloudik^{b,c}

Introduction. Sonothrombolysis is a new treatment method for patients with acute ischemic stroke (IS). Various ultrasound frequencies and intensities are being tested these days. The aim of this pilot study was to assess the safety and efficacy of sonothrombolysis using 2 diagnostic probes and bilateral monitoring in patients with acute occlusion of the middle cerebral artery (MCA).

Patients and Methods. Twelve consecutive IS patients (7 males; age 47 – 78, average 64.1 ± 9.4 years) with acute MCA occlusion and contraindication of thrombolysis were included in the study. 60-min bilateral 2-MHz pulsed-wave Doppler monitoring of the area of occlusion was performed in all patients (Group 1). The control group consisted of 37 IS patients (20 males; age 32 – 78, average 62.2 ± 12.1 years) treated with standard sonothrombolysis and selected from the Thrombotripsy Study database (Group 2). The differences in number of recanalized arteries after a 1 h treatment, independent patients (modified Rankin scale [mRS] value of 0 – 2) after 90 days and symptomatic intracerebral hemorrhages (SICH) were statistically evaluated.

Results. Complete recanalization was found in 4 (30.0%) Group 1 and in 12 (32.4%) Group 2 patients. Seven (58.3%) Group 1 and 22 (59.5%) Group 2 patients were independent after 90 days. SICH was found in none of Group 1 patients and in 1 (2.7%) of the Group 2 patients ($P > 0.05$ in all cases).

Conclusion. In this pilot study, sonothrombolysis using 2 probes and bilateral monitoring is safe but not more effective than standard sonothrombolysis in acute IS patients with MCA occlusion.

Key words: sonothrombolysis, ischemic stroke, recanalization, therapy

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^aDepartment of Neurology, Hospital Trinec-Sosna, Trinec, Czech Republic

^bComprehensive Stroke Center, Department of Neurology, University Hospital Ostrava and Ostrava University, Ostrava

^cComprehensive Stroke Center, Department of Neurology, Faculty of Medicine and Dentistry, Palacky University Olomouc and University Hospital Olomouc

Corresponding author: David Skoloudik, e-mail: skoloudik@email.cz

INTRODUCTION

In vitro and in vivo studies on animals performed from the 1970's have confirmed acceleration of thrombus dissolution using ultrasound with various intensities (0.2 up to 2.0 W/cm^2) and frequencies (20 kHz up to 2 MHz) (ref.¹⁻⁶). In the last ten years, several studies have found this effect also in patients with ischemic stroke (IS) and middle cerebral artery (MCA) occlusion⁷⁻¹².

Alexandrov et al. (2004) proved the safety and efficacy of sonothrombolysis using transcranial Doppler (TCD) monitoring of the occluded MCA over 2 h in the largest randomised CLOTBUST trial. These results were also confirmed in the Thrombotripsy study using diagnostic duplex transcranial probe with a Doppler frequency of 2 MHz¹⁰. When comparing the safety of sonothrombolysis alone and in combination with intravenous thrombolysis (IVT), a higher occurrence of symptomatic intracerebral hemorrhage (SICH) was found in the latter¹¹. Besides the transcutaneous form via the temporary cranial window, sonothrombolysis can be performed also endovascularly using the EKOS system and the high frequencies of emitted ultrasound (2.05 – 2.35 MHz) directly in the area of thrombus through intra-arterially inserted catheter¹³.

In vitro and *in vivo* studies on animal models as well as on IS patients with volunteers proved that ultrasound with low frequencies and high intensity has a primarily mechanical effect, quickly destroying the thrombus into microscopic fragments, whereas using high frequencies (around 2 MHz) causes activation of the endogenous fibrinolytic system^{3,14,15}.

The aim of this pilot study was to assess if local increase of ultrasound intensity (spatial peak temporal average, SPTA) in the area of the occluded MCA using 2 diagnostic transcranial Doppler probes with 2-MHz frequency during bilateral monitoring increases the percentage of MCA recanalizations and improves the clinical outcome of IS patients with occlusion of the MCA arterial trunk or branch.

PATIENTS AND METHODS

Inclusion and exclusion criteria

All consecutive patients with acute IS and occlusion of the main trunk (M1 segment) or the branch (M2 segment) of the MCA contra-indicated for IVT meeting the inclusion and exclusion criteria shown in Table 1 were

included in the study during a 4-month period (November 2010 – February 2011) – Group 1. IVT treated patients were excluded due to higher risk of SICH occurrence when combined with sonothrombolysis.

Diagnostics

All patients underwent physical and neurological examination included in the National Institutes of Health Stroke Scale (NIHSS) evaluation, collection of biochemical samples and coagulation, blood count, electrocardiogram record, heart and lung X-ray, brain computed tomography (CT), duplex sonography of cervical arteries and transcranial colour-coded sonography (TCCS). Sonothrombolysis using bilateral monitoring of symptomatic MCA was initiated after detection of occlusion of MCA main trunk (M1 segment) or branch (M2 segment) (ref.¹⁶).

Treatment

All patients were treated according to accepted recommendations with early commencement of secondary prophylactic treatment¹⁷. Mini-heparinization (low-dose heparin, low molecular weight heparin) was administered as a prevention of deep venous thrombosis according to accepted recommendations. All patients were treated at a stroke unit.

Sonothrombolysis

A 60-min bilateral Doppler monitoring of the area of occlusion was performed in all Group 1 patients. The area of the arterial occlusion was detected from both the ipsilateral and contralateral cranial bone window using a TCD probe which was bilaterally fixed with monitoring helmet. Sample volume was set at 10 mm. Correct position of the probe was confirmed using the detection of preocclusive flow in the MCA and a typical “click” in Doppler mode. Continual bilateral monitoring was commenced without delay using the maximum diagnostic intensity of ultrasound and was carried out for a period of up to 60 minutes. Artery recanalization was evaluated every 5 min and, in case of the detection of recanalization with continuous flow (TIBI III – V), the sonothrombolysis was terminated. Echocontrast agents were not used in any patient.

The TCD instrument DWL MultiDop T2 (DWL Elektronische Systeme, Sipplingen, Germany) with the option of bilateral monitoring using 2 probes with 2.0 MHz frequency was used for sonothrombolysis.

Control group

The control group (Group 2) consisted of patients contraindicated for IVT and treated with standard sonothrombolysis selected from the Thrombotripsy trial database¹⁰.

Statistical analysis

The Kolmogorov-Smirnov and Shapiro-Wilk test were used for the testing of normal distribution in all monitored parameters. Data with normal distribution are presented

as average \pm standard deviation. Other data are presented as average, median and interquartile range.

The following data were statistically evaluated: age, sex, time to commencement of sonothrombolysis, occurrence of risk factors (arterial hypertension, diabetes mellitus, hyperlipidemia, smoking, excessive alcohol intake [defined as > 36 g of pure alcohol per day for females and > 54 g of pure alcohol for males¹⁸], atrial fibrillation), difference in the number of recanalized arteries after 1 h of treatment and 24 h from symptom onset, difference in the number of independent patients after 90 days (defined as a modified Rankin scale [mRS] value of 0 – 2) and the number of SICH, using the non-parametric Kruskal-Wallis test, chi-square test and Wilcoxon signed ranks test. $P < 0.05$ was considered significant.

Statistical analysis was performed using the SPSS 14.0 software (SPSS Inc., Chicago, IL, USA).

Ethics

The whole study was conducted in accordance with the Helsinki declaration of 1975 (as revised in 2004 and 2008) and it was approved by the local ethics committee of the University Hospital in Ostrava. Informed consent was obtained from each patient.

RESULTS

During a 4 month period, 12 patients (7 males, 5 females; age 47 – 78, average 64.1 ± 9.4 years) were included in the study (Group 1). The control group (Group 2) consisted of 37 patients from the Thrombotripsy trial database (20 males, 17 females; age 32 – 78, average 62.2 ± 12.1 years). Patient demographic data are presented in Table 2.

Complete recanalization within 60 min from the start of sonothrombolysis was detected in 4 (30.0%) of Group 1 patients and in 12 (32.4%) of Group 2 patients ($P > 0.05$). After 24 h since symptom onset, complete recanalization was detected in 11 (91.7%) of Group 1 patients and in 36 (97.3%) of Group 2 patients ($P > 0.05$). Independence after 90 days (mRS 0 – 2) was achieved in 7 (58.3%) of Group 1 patients and in 22 (59.5%) of Group 2 patients ($P > 0.05$). SICH was detected in none (0%) of Group 1 patients and in 1 (2.7%) of Group 2 patients ($P > 0.05$). Re-occlusion of the MCA during 24 h was detected in none of the Group 1 patients and in 1 (2.7%) of Group 2 patients ($P > 0.05$).

DISCUSSION

The results show no superiority of sonothrombolysis using bilateral TCD monitoring over standard sonothrombolysis. Nevertheless, the safety of the sonothrombolysis of an acute intracranial artery occlusion using a diagnostic 2-MHz probe, and its potential positive effect on the acceleration of the early arterial recanalization were confirmed. The results also indicate a probable biological

Table 1. Inclusion and exclusion criteria.

Inclusion criteria	<ul style="list-style-type: none"> • Acute ischemic stroke • Detection of the occlusion of the trunk (M1) or the branch (M2) of the MCA according to TIBI criteria using transcranial Doppler sonography up to 6 hours since symptoms onset • National Institute of Health Stroke Scale score 4 – 25 points • Appropriate ultrasound temporal cranial window with detection of the flow in the arteries of the circle of Willis • Sonothrombolysis start within 6 h since symptoms onset • Age 18 – 80 years • Signed informed consent
Exclusion criteria	<ul style="list-style-type: none"> • Patients with modified Rankin scale value > 1 point before symptoms onset • Signs of ischemia in extent larger than 1/3 of the MCA territory, acute ischemia in other area than that of MCA, intracranial bleeding or brain tumour detected by CT • Patients indicated to intravenous or intraarterial thrombolysis or mechanical recanalization of the MCA or participation in a clinical study within last 3 months

CT – computed tomography, MCA – middle cerebral artery, TIBI – Thrombolysis in Brain Ischemia

Table 2. Patients demographic data.

	Group 1 (patients treated with bilateral sonothrombolysis)	Group 2 (control group from the Thrombotripsy trial)	<i>P</i> value
Age, mean ± SD (years)	64.1 ± 9.4	62.2 ± 12.1	0.34
Males, n (%)	7 (58.3%)	20 (54.1%)	0.40
NIHSS on admission, mean, median, IQR	14.2, 15, 8 – 20	14.7, 15, 9 – 20	0.43
Time to the commencement of sonothrombolysis, mean ± SD (min)	133.8 ± 58.4	142.3 ± 56.6	0.22
Arterial hypertension, n (%)	10 (83.3%)	31 (83.8%)	0.49
Diabetes mellitus, n (%)	3 (25.0%)	12 (32.4%)	0.32
Hyperlipidemia, n (%)	6 (50.0%)	20 (54.1%)	0.41
Atrial fibrillation, n (%)	8 (66.7%)	26 (70.2%)	0.41
Smoking, n (%)	4 (33.3%)	17 (45.9%)	0.23
Excessive alcohol intake, n (%)	6 (50%)	24 (64.9%)	0.20
Occlusion / stenosis > 50% of the ICA, n (%)	0 / 3 (0 / 25.0%)	2 / 6 (5.4 / 16.2%)	0.42
Left hemisphere lesion, n (%)	5 (41.7%)	20 (54.1%)	0.24

ICA – internal carotid artery, IQR – interquartile range, min – minutes, SD – standard deviation

effect of diagnostic TCD monitoring which may facilitate spontaneous recanalization of the arterial occlusion.

In this study, the pulsed wave (PW) Doppler was used. In contrast to continual wave (CW) Doppler where the interference of ultrasound waves is significant and can lead to constructive or destructive interferences¹⁹, in PW Doppler mode these interferences are extremely rare and do not have any significant influence. The use of 2 probes in PW mode leads predominantly to increase in SPTA.

Although a potential effect of the ultrasound on the thrombus dissolution has been shown by in vitro studies and studies on animal models since the 1970's (ref.^{1,2,4,6,14,20-23}), the effect of TCD monitoring in acute IS patients was reported for the first time in 2000 (ref.²⁴) and the results of the first randomized study, testing the effect of sonothrombolysis showed the safety and efficacy of sonothrombolysis using the diagnostic 2-MHz TCD probe⁷. The present study confirmed these results but in contrast to the results of the Clotbust study⁷, no re-occlusion was detected during the first hours in either group.

The mechanism of ultrasound effects on thrombus is still not clearly known. Firstly, mechanical effect caused by the thrombus vibration may lead to the acceleration of the penetration of fibrinolytics³. Secondly, non-thermal mechanisms – increasing the transport of fibrinolytic agents into the thrombus by mechanical disruption of its structure²⁴, direct activation of fibrinolytic enzymes^{14,15}, transient peripheral vasodilatation caused probably by increased production of nitrite oxide in the endothelium^{21,26} also play a role in clot dissolution. However, confirmation of different effects on thrombus dissolution using various ultrasound frequencies and intensities are still missing.

Few serious adverse events have been reported in studies using diagnostic transcranial ultrasound with monitoring to accelerate the recanalization of the MCA occlusion in acute IS patients and these did not exceed control groups values⁷⁻¹². In the present study, no SICH or symptomatic brain oedema was found in patients treated by sonothrombolysis using bilateral TCD monitoring. In the control group, the SICH was detected in only 1

(1.9%) patients. These results are even better than the Clotbust study where the occurrence of SICH was 4.8% in the target group⁷. In contrast to studies using diagnostic transcranial probes, two studies using a non-diagnostic 300 kHz probe were stopped^{27,28}. The Trumbi study²⁷ was terminated prematurely because of excessively increased number of intracranial hemorrhages (92.8% intracranial haemorrhages, 35.7% SICH) and the results of the study presented by Reinhard et al. showed a disruption of the blood-brain barrier after sonothrombolysis²⁸.

Several limitations of the presented study should be mentioned. Firstly, this is only a prospective case-control study with historic controls from the observational Thrombotripsy trial. Secondly, in the Thrombotripsy trial, TCCS-guided TCD using a TCCS probe was used, while in the present study, pure TCD technique using a TCD probe was applied. The ultrasound beam in TCD mode differs in the probes. This might affect the comparability of the groups. Thirdly, although clinical evaluations were performed by a blinded neurologist, TCCS/TCD evaluation of the artery recanalization was assessed by the same sonographer who performed the sonothrombolysis. Finally, a relatively small number of patients is another cause of potential bias.

Regarding the second mentioned limitation, experimental measurement of the intensity of ultrasound beam using both one and two TCCS-guided TCD and TCD probes with various energies would be worthwhile to find the optimal approach for sonothrombolysis

CONCLUSION

Sonothrombolysis using diagnostic 2-MHz transcranial probe is a safe method of treatment of IS in patients with MCA occlusion. However, the increase of ultrasound intensity in the area of arterial occlusion using bilateral monitoring probably does not lead to increase in the number of recanalized arteries and subsequent improvement in patients clinical outcome.

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