

Detection of cerebral ischemia in rabbits by magnetic induction phase shift spectroscopy comparison with laser Doppler flowmetry

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Abstract

Acute cerebral ischemia is one of the important clinical diseases that are usually detected and studied with magnetic resonance imaging, computed tomography, and positron emission tomography. Magnetic inductive phase shift (MIPS) is a new method used to detect cerebral ischemia which is non-contact, miniaturized, and low-cost. To obtain more useful information in broadband measurement of cerebral ischemia, we extended magnetic induction phase shift spectroscopy (MIPSS). A total of 25 rabbits were studied using a two-coil sensor within 0.3MHz to 200MHz frequency range in linear scanning mode, and all the subjects were measured for 1 hour. On the basis of a rabbit acute cerebral ischemia model by carotid artery occlusion, the rabbits were divided into unilateral ligation, bilateral ligation and non-ligation group. The characteristic frequency with the highest detection sensitivity was selected for further analysis and processing. Results showed that the average phase shift of transmission parameter S_{21} of non-ligation group, unilateral ligation group and bilateral ligation group was $-0.195 \pm 0.079^\circ$, $-4.873 \pm 1.042^\circ$, and $-9.165 \pm 2.862^\circ$ respectively. MIPSS can distinguish different severities of cerebral ischemia in rabbits with a statistical significance of $p < 0.05$. And the correlations of laser Doppler flowmetry and phase shift suggested that phase shift can reflect the blood flow changes in brain. Overall, the results suggest that the MIPSS detection method provides a potential for early detection and distinguishing different severities of acute global cerebral ischemia.

Keywords: acute cerebral ischemia, magnetic induction phase shift spectroscopy, phase shift, laser Doppler flowmetry.

Introduction

Ischemia of the brain is a medical condition in which a certain region of the brain is deprived of oxygen and nutrient-rich blood ^[1]. Ischemic stroke accounts for about 90% of all strokes and can occur in people of all ages, although the risk increases with age ^[2]. Cerebral ischemia can lead to severe damage of the central nervous system and eventually causes paralysis. Thus, early detection and real-time monitoring are the keys to emergency treatment and intensive care of cerebral ischemic stroke.

Magnetic induction phase shift (MIPS), which provides advantages of non-contact, noninvasive, inexpensive, small, and continuous bedside monitoring, is one of the potential methods of cerebrovascular disease detection. Gonzalez et al ^[3, 4] measured mouse abdominal cavity fluid and rat cerebral ischemia by using coaxial coils, and then they distinguish edema patients from hematoma patients and healthy volunteers by establishing a VEPS multi-frequency measurements system and a classifier ^[5]. Jin and Sun et al ^[6, 7, 8] detected rabbit acute cerebral hemorrhage by using a coaxial parallel double-coil. Li et al ^[9, 10] detected brain hemorrhage and cerebral edema in

rabbits based on MIPS method. Considering that the conductivity of biological tissues is very small, the magnetic field disturbance generated by the brain tissues is too weak. To improve the detection sensitivity of MIPS, a variety of coil structures have been introduced by numerous scholars^[11, 12]. However, the detection sensitivity of these methods is very limited and can be used only in laboratory models.

In this study, to observe the trend of MIPS changes in terms of ischemia severity and to improve the sensitivity of ischemia detection, we established a detection system based on magnetic induction phase shift spectroscopy (MIPSS) for acute cerebral ischemia. A total of 25 rabbits were studied by using a two-coil sensor within 0.3MHz to 200MHz frequency range. Characteristic frequency (CF) with optimal detection sensitivity within the entire band was selected for further analysis and research.

Materials and Methods

1. Detection Principle

In MIPS theory^[13], the measured sample is always placed between the excitation coil and the receiving coil, and a primary magnetic field (B) is generated by the sinusoidal excitation current ($Ie^{j\omega t}$) flowing in the excitation coil. This primary magnetic field induces induction currents in the sample, which consequently produce a second magnetic field (ΔB). The receiving coil receives the total magnetic field ($B+\Delta B$). The intensity of the induced magnetic field is related to the electrical conductivity of the measured sample, and the changes in electrical conductivity of the measured sample can be reflected by detecting the induced magnetic field intensity (ΔB). The phase shift of the total magnetic field is obtained through the voltage phase difference between receiving coil and excitation coil. MIPSS is a detection method based on the MIPS theory. MIPSS extends a single frequency to a limited frequency band and calculates the phase shift difference spectrum on the frequency band, then searches out the frequency with the highest detection sensitivity in the entire band.

2. Experimental System

A magnetic induction system to detect acute cerebral ischemia is shown in Figure 1. The system mainly includes the following: RF vector network analyzer (Agilent E5061B) was used to measure the amplitude and phase of signals, and the frequency range of input signal was 0.3MHz to 200MHz in linear scanning mode; a two-coil sensor was used to receive signals. We found that a frequency point with the maximum amplitude has the largest phase shift change, hence we defined the frequency point corresponding to the maximum amplitude as the characteristic frequency (CF). An intracranial pressure monitor was used to measure intracranial pressure (ICP) of rabbits. To reflect the change of blood flow in brain region, we used laser Doppler flowmetry (LDF) to observe the local cerebral blood flow perfusion of rabbits in this study. To observe the vital signs of animals, a multichannel physiological signal acquisition instrument was used to continuously monitor electrocardiogram (ECG) and heart rate variability (HRV) of rabbits.

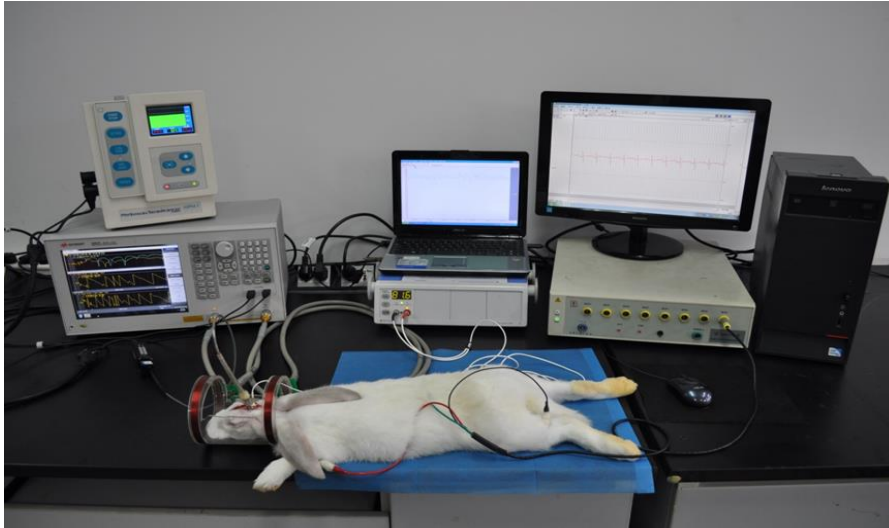


Figure 1. Photograph of experimental setup of cerebral ischemia magnetic induction detection system.

3. Experimental Design

Twenty-five rabbits (2.2 ± 0.2 kg) were randomly selected and divided into unilateral ligation group ($n=10$), bilateral ligation group ($n=10$), non-ligation group ($n=5$). The acute cerebral ischemia rabbit model was established by bilateral common carotid artery ligation method^[14]. The method started with anesthetization through an ear injection of urethane (25%, 5 ml/kg). Hair on the rabbit necks and the head was removed. The center of the neck skin was cut off after disinfection and the muscles were separated bluntly, thereby exposing and separating the bilateral common carotid arteries. A surgical ligation line was placed under common carotid arteries. The ICP probe was fixed on the brain “cross stitch” with 6 mm to the left of the coronal suture, 1mm below to the sagittal suture, and the LDF probe was placed on the brain “cross stitch” with 6 mm to the right of the coronal suture, 1 mm below to the sagittal suture. Then, the electrodes of the physiological signal acquisition instrument were inserted into the thighs subcutaneously to record the ECG and HRV. Finally, the surgical ligation line was fastened to block the unilateral or bilateral common carotid arteries in the process of experiment. The same operations were performed for three groups, and the detection time was 1 hour for all rabbits.

4. Data Analysis

To obtain the phase shift of transmission parameter S_{21} , we used the time-difference method to calculate the phase shift of every time point. Then, we drew the graph of the phase shift spectrum of each parameter within 0.3MHz to 200MHz by using MATLAB software. To demonstrate the significant difference among the eight cerebral ischemia levels, non-parametric statistical Friedman’s M-test and nonparametric statistical Kruskal–Wallis test were used to analyze the phase shift, Pearson correlation coefficients was used to analyze the relationship of phase shift and LDF measurement. Statistical analysis was conducted by SPSS12.0, and the significance level criterion p-value was 0.05.

Results and Discussion

1. Results

All the rabbits survived in this experiment. It's worth mentioning that the ICP data of cerebral ischemic rabbits basically remained unchanged. In one (No.14) of bilateral ligation group rabbits,

the ICP remained at 15mmHg $\pm$ 2mmHg, and the HRV decreased from 330 $\pm$ 5 to 280 $\pm$ 7 after 60min ligation time. Figure 2(a) showed the phase shift of the transmission parameter S_{21} in 0.3MHz to 200MHz frequency band during ligation time. Figure 2(b) illustrated that phase shift changed with ligation time around CF. The phase shift increased with the successive increase of ligation time in a negative way, indicating a strong correlation between phase shift and ligation time. This result is similar to the results reported by Gonzalez et al ^[4].

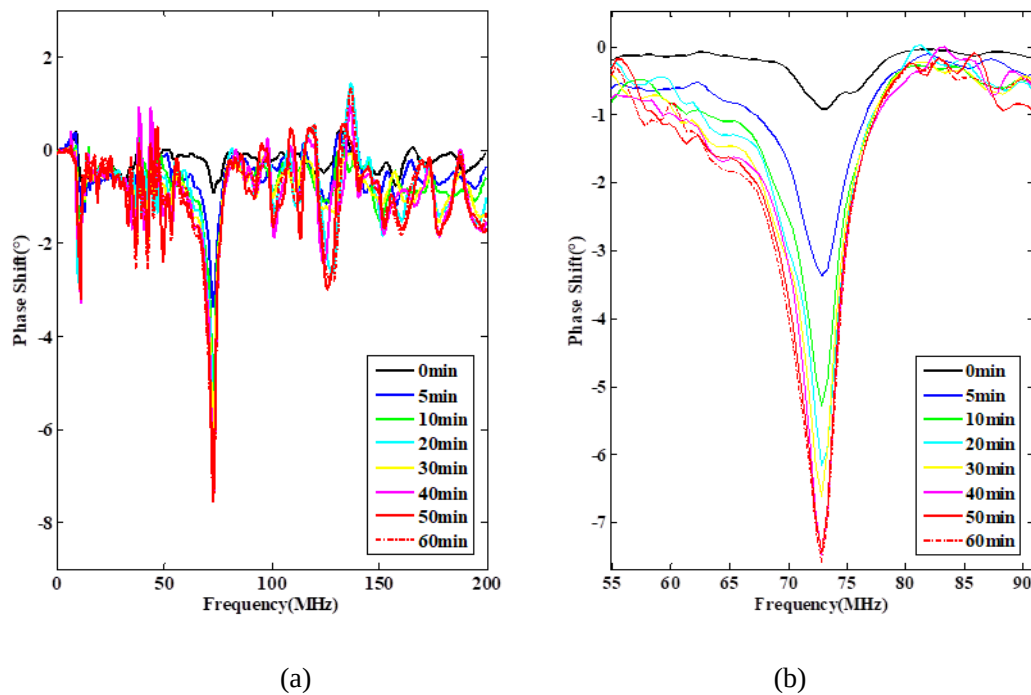


Figure 2(a) Spectra of phase shift of S_{21} as a function of frequency (0.3MHz to 200 MHz) for 60min measurement of rabbit No.14. (b) Phase shift around CF. The frequency is on a linear scale.

The descriptive statistical results of the phase shift at CF are summarized in Table 1. Five animals belonged to non-ligation group, and unilateral and bilateral groups comprised 10 rabbits respectively. The average phase shift in 60 min of non-ligation group was $-0.195 \pm 0.079^\circ$; the phase shift of unilateral ligation group was $-4.873 \pm 1.042^\circ$; and the phase shift of bilateral ligation group was $-9.165 \pm 2.862^\circ$. The nonparametric statistical Friedman's M-test was applied to the phase shift data analysis in three groups. The p-value of non-ligation group was 0.363 which was greater than 0.05, indicating no significant differences. And the p-value of unilateral ligation group and bilateral ligation group were both less than 0.05 which indicated that phase shift changed with ligation time significantly. Then, we used nonparametric statistical Kruskal–Wallis test to analyze the differences of average phase shift among three groups. Results revealed statistically significant differences among severe levels of cerebral ischemia with p-value < 0.05 . The preceding results suggested that we can distinguish different ischemia levels (non-ligation, unilateral ligation, and bilateral ligation group) with phase shift. Above all, MIPSS method provided a new method for early detection of rabbits' acute cerebral ischemia.

Table 1. Statistical analysis with Friedman's M-test of average phase shift of parameter S_{21} for three groups in which a statistically significant difference of p-value < 0.05 was found.

Ligation time	Non-ligation	Unilateral ligation	Bilateral ligation	p-level
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0min	-0.061 ± 0.046	-0.124 ± 0.209	-0.040 ± 0.437	1.214E-6
5min	-0.123 ± 0.055	-1.443 ± 0.522	-2.200 ± 0.567	2.628E-5
10min	-0.148 ± 0.039	-2.075 ± 0.689	-3.377 ± 0.913	2.381E-6
20min	-0.088 ± 0.023	-3.125 ± 0.590	-6.253 ± 1.573	3.718E-6
30min	-0.171 ± 0.113	-3.460 ± 0.545	-7.938 ± 1.754	2.174E-5
40min	-0.166 ± 0.022	-4.026 ± 0.825	-8.157 ± 2.179	4.835E-6
50min	-0.187 ± 0.029	-4.262 ± 0.985	-8.530 ± 2.269	5.738E-7
60min	-0.195 ± 0.079	-4.873 ± 1.042	-9.165 ± 2.862	6.428E-7
p-level	0.363	1.773E-5	1.255E-5	
Valid N	5	10	10	

Figure 3(a) showed the relationship between phase shift and LDF measurement of S_{21} of rabbit No.14. Phase shift of parameter S_{21} increased gradually with ligation time in a negative way. The LDF measurement decreased from 100% to 64% in 10 min, and remained dynamic stability for the next 50 min. Then, we draw the average LDF measurements of three groups in Figure 3(b). The average LDF measurement of three groups were very different, the average LDF measurement essentially remained unchanged for non-ligation group, decreased from 100% to 80% for unilateral ligation group, and dropped from 100% to 65% for bilateral ligation group.

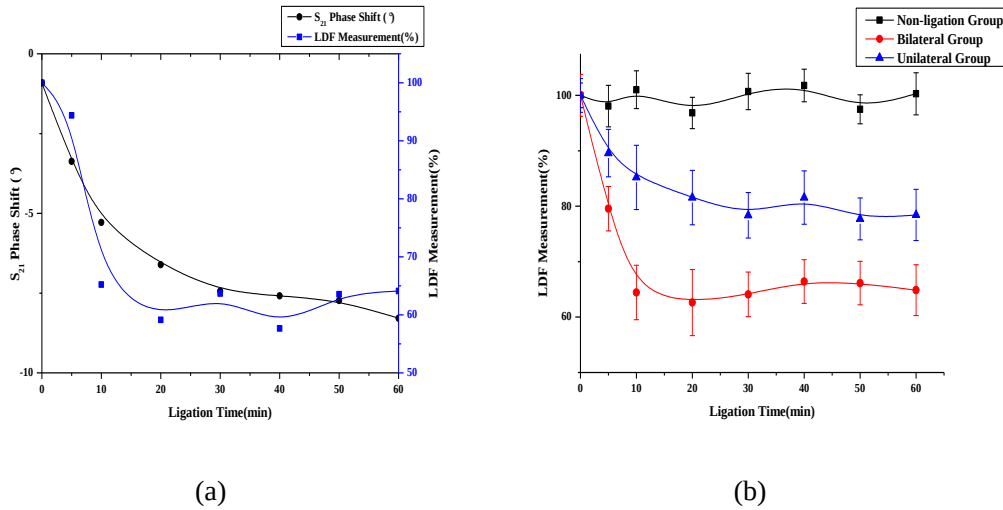


Figure 3. The relationship between phase shift and LDF measurement and average LDF measurement of three groups.

The cerebral blood flow is the primary variable in cerebral ischemia rabbits, and the change of cerebral blood flow will affect whole brain conductivity. This study analyzed the correlation between phase shift and LDF measurement by using Pearson correlation coefficients. Table 2 showed the correlations between phase shift and LDF measurement for parameter S_{21} of 20 rabbits. The correlation coefficients between phase shift and LDF measurement of S_{21} were positive. The correlation coefficients of bilateral ligation group were greater than that of unilateral ligation group. And p-value of bilateral ligation group were all less than 0.05, indicating that phase shift

was strongly correlated with LDF measurement of bilateral ligation group. Above all, the LDF measurement (blood flow data) confirmed the severity of cerebral ischemia.

Table 2. Correlation analysis of phase shift and LDF measurement from unilateral ligation and bilateral ligation group

Group	Sample	Correlation Coefficient	P-value
Unilateral Ligation Group	01	0.767	0.02
	02	0.732	0.03
	03	0.721	0.04
	04	0.620	0.10
	05	0.709	0.04
	06	0.679	0.06
	07	0.705	0.05
	08	0.698	0.05
	09	0.783	0.02
	10	0.723	0.03
Bilateral Ligation Group	11	0.867	0.01
	12	0.910	0.01
	13	0.849	0.01
	14	0.934	0.01
	15	0.899	0.01
	16	0.861	0.01
	17	0.900	0.01
	18	0.933	0.01
	19	0.871	0.01
	20	0.860	0.01

Discussion

The change of phase shift mainly relates to the electromagnetic properties (mainly electrical conductivity) of brain tissues in MIPSS measurement. The coil structure (coaxially paralleled dual-coils) used in this study determined that the measured phase shift reflected conductivity changes in the whole rabbit head which was placed in the space covered with the excitation magnetic field. The conductivity changes in the whole rabbit head were related to the severity levels of cerebral ischemia. In this study, we controlled the ligation time as 1 h to simulate the early rapid cerebral ischemia. The ligation of common carotid artery significantly reduced the cerebral blood flow (CBF) and resulted in shortage of CBF supply. Ischemia caused changes in the electromagnetic properties of the brain tissues^[15].

The results of Figure 3 indicated that LDF measurement varied distinctly within 10 minutes, and remained largely unchanged in following 50 minutes. We thought that's due to the LDF measurement is very sensitive to the changes of cerebral blood flow after ligation, whereas the LDF measurement showed no remarkable change once autonomic regulation of cerebral blood flow started. According to the LDF measurement, the phase shift of both parameters and LDF measurement were strongly correlated especially in bilateral ligation group which demonstrated that phase shift change mainly due to cerebral ischemia, the ligation of common carotid artery

significantly reduced the cerebral blood flow (CBF) and resulted in the change of conductivity in whole brain.

The experimental results of MIPSS data analysis and processing at the CF completely demonstrated the feasibility of our method for early detection of acute cerebral ischemia in rabbits. By using this method, we can clearly distinguish the differences in healthy rabbits (non-ligation group), general-ischemia rabbits (unilateral ligation group), and severe-ischemia rabbits (bilateral ligation group) to achieve the rapid detection and diagnosis of acute cerebral ischemia.

Conclusion

In this study, we developed an MIPSS method to detect acute cerebral ischemia in rabbits. Our experimental results showed the possibility of early and non-contact detection of global acute cerebral ischemia and the potential to distinguish the severity level of cerebral ischemia in rabbits. This preliminary study is mainly based on the animal model of the global acute cerebral ischemia, and further research remains necessary for local cerebral ischemia and clinical trial. Although cerebral ischemia in clinical cases is a highly complex process, this preliminary study provides the feasibility of detecting global acute cerebral ischemia by MIPSS method.

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