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Cortical-evoked potentials and cerebral infarction in temporary middle cerebral artery occlusion in the cat

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Summary

Somatosensory-evoked potentials (SSEP) and cerebral infarct size were measured in nine cats in which the middle cerebral artery (MCA) was clip-occluded for 3 h followed by an equal period of recirculation. During MCA occlusion, there was a reduction in the mean amplitude of the major cortical wave (MCW) to less than 50% of the pre-occlusion value. There was no significant improvement in this amplitude after clip release. Cerebral infarction was detected in eight of the animals and the mean (± s.d.) infarct size was 27.8 ± 19.6%. A good negative correlation between the MCW amplitude and infarct size was apparent as early as 15 min into the period of occlusion ($r = -0.87$, $P < 0.01$). In this model of focal cerebral ischaemia, the 15-min MCW amplitude is a good predictor of cerebral infarction.

Résumé

Les potentiels somatosensoriels evoqués (PSSE) et la dimension d'infarctus cerebral ont été mesurés chez neuf chats dans lesquels l'artère cérébrale intermédiaire (ACI) était occlus avec l'attache pour 3 h suivi par une même durée de la ré-circulation. Pendant l'occlusion d'ACI, il y avait une réduction dans l'amplitude moyenne de l'onde corticale majeure (OCM) à moins de 50 pourcent de la valeur pré-occlusion. Il n'y avait pas d'amélioration importante dans cet amplitude après le dégagement de l'attache. L'infarctus cérébral était découvert chez huit des animaux et la moyenne (± s.d.) de dimension d'infarctus était 27.8 ± 19.6%. Une bonne corrélation négative entre l'amplitude de l'ACI et la dimension d'infarctus était évidente le plus tôt 15 min de la période d'occlusion ($r = -0.87$; $P < 0.01$). L'amplitude 15-min d'ACI est un bon prédicteur de l'infarctus cérébral dans cette modèle expérimentale de l'ischémie focale cérébrale.

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Introduction

The feline middle cerebral artery (MCA) occlusion model has been studied extensively, for the haemodynamic, biochemical and morphological accompaniments of stroke [1–6], and for the role of several putative protective agents [5–10]. However, information has only recently become available in this model on the usefulness of somatosensory-evoked potentials (SSEP) as a measure of the severity of cerebral infarction. In one study, occlusion of the MCA for 6 h or more was associated with marked reduction in the amplitude of the major cortical wave (MCW) complex and prolongation of interpeak latencies [11]. The size of the cerebral infarct was shown to be related to the severity of SSEP changes although it was only indirectly estimated by measurement of areas of the brain not stained by India ink perfusion.

The histological changes of cerebral infarction in cats have subsequently been correlated with SSEP by Steinberg *et al.* [12]. Complete failure of recovery of SSEP during 6 h of MCA occlusion occurred in animals with severe neuronal alterations, thus providing further evidence in this model that SSEP measurement is a reliable method of assessment of cerebral ischaemic damage. In both of these studies, MCA occlusion was permanent, the animals being killed without allowing re-perfusion of

ischaemic territory [11,12]. In the present investigation, we have examined the relationship between SSEP and cerebral infarction produced experimentally by temporary MCA occlusion. We measured the changes in the amplitude of the major cortical wave of the SSEP during 3 h of MCA occlusion and an equal period of re-perfusion in the cat, and estimated the size of infarcts by the tetrazolium histochemical method [13].

Materials and methods

The nine cats (2.5–4.0 kg in weight) reported in this study were also included in a larger project in our laboratory on cerebral ischaemic rescue. However, these animals were not subjected to any therapeutic interventions. They were prepared for transorbital occlusion of the middle cerebral artery. Anaesthesia was commenced with intraperitoneal ketamine hydrochloride (30 mg/kg) and atropine sulphate (0.1 mg/kg). After intubation, anaesthesia was maintained by mechanical ventilation with 0.5–1.5% halothane in an air/oxygen mixture (FIO_2 0.4) titrated to keep blood pressure at approximately 120/70. The animals were paralysed with gallamine triethiodide (6 mg/kg, i.v.). The core temperature was maintained and monitored by means of a thermal blanket and a rectal probe, respectively. The femoral artery and vein were cannulated for continuous monitoring of pulse and blood pressure and blood sampling. P_aCO_2 was maintained between 30 and 35 mmHg and pH between 7.35 and 7.45.

The head was rigidly immobilized, and under magnified vision the left middle cerebral artery was occluded with a Heifitz clip. The clip was removed 3 h later and the patency of the vessel was confirmed by inspection. The animals were killed 3 h after clip removal.

Somatosensory-evoked potentials

Each median nerve was stimulated in the distal forepaw with a pair of subcutaneous electrodes. The evoked response was recorded from a mid-parasagittal scalp electrode over the contralateral cerebral hemisphere. The two scalp electrodes were connected to a common fronto-nasal lead. The stimulus consisted of a square wave pulse of 2 mA and 0.2 msec, strong enough to twitch the forepaw before muscle paralysis, and delivered at a rate of 4.5/sec. The stimuli were generated and acquired by a Nicolet compact 4 averager (Madison, USA). Two hundred and fifty-six responses were averaged and displayed. For each recording, two sets of responses were averaged for each hemisphere. We analysed the evoked responses recorded at the following points during the experiment: during the 30 min prior to MCA occlusion; 15 min into the period of occlusion; at the end of the period of occlusion (before clip removal); 15 min and 3 h after clip removal. The amplitude of the major cortical wave was measured from the trough of the positive deflection to the peak of the negative deflection. The pre-occlusion value was regarded as the baseline and subsequent measurements were expressed as percentages of this value.

Measurement of infarct size

The experiments were terminated by euthanasia and transcardiac perfusion of the animals with 500 ml of physiological saline. A 5-mm thick coronal section of the cerebrum, obtained at the level of the optic chiasm, was incubated in 50 ml of a 2% solution of 2′ 3′ 5′ triphenyl-2H-tetrazolium chloride (TTC) for 30 min at 37°C. After incubation, the slices were photographed onto 35 mm slides. The areas of pallor corresponding to the areas of infarction [13] were mapped by projection and planimetry and calculated as percentages of the cross-sectional areas of the slices. The average of the values of the two surfaces of each slice was obtained.

The results were analysed with Student's *t*-test for paired samples and Pearson's correlation analysis.

Results

Blood pressure, pulse and temperature were maintained within physiological limits. The arterial P_aCO_2 was kept between 30 and 35 mmHg.

Somatosensory-evoked potentials

The distribution of the changes in MCW

amplitudes in the animals is illustrated in Fig. 1 and summarized in Table 1. In the left hemisphere, the mean amplitude of the major cortical wave at 15 min of occlusion was 41.2% of its baseline value. This severe decrement persisted and was observed at the end of the period of occlusion (35.5% MCW amplitude). The slight increase in mean amplitude after clip release was not statistically significant (Fig. 1). At the end of the period of re-perfusion, the mean MCW amplitude was only half of its value prior to MCA occlusion.

At each of the points of measurement, the amplitude averaged over the ischaemic (left) hemisphere was significantly less than that of the contralateral side, the maximum difference being noted at the end of the period of MCA occlusion ($P < 0.01$) (Table 1).

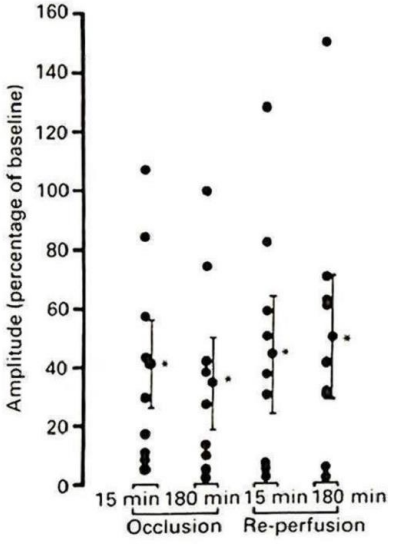

Fig. 1. Distribution of the amplitudes of the left MCW at each of the points of measurement. Each cat is represented by a single dot. Dot with vertical bar: mean ± s.d. These values were not significantly different ($P < 0.1$, *t*-test).

Infarct size and SSEP

An infarct was detected in eight of the cats. The size of the infarct varied considerably with a mean ± s.d. of 27.81 ± 19.57% (Table 2). The size of the infarct was inversely related to MCW amplitude. A significant negative correlation with infarct size was evident for amplitude measurements at 15 min ($P < 0.01$) and 3 h ($P < 0.001$) of occlusion, as well as 15 min ($P < 0.05$) and 3 h ($P < 0.01$) of re-perfusion (Fig. 2).

Discussion

The changes that occur in the somatosensory-evoked potentials during global and focal cerebral ischaemia have been described in several reports [14–18] and the utility of this measure of experimental ischaemia is generally accepted. SSEP amplitude has been correlated with cerebral blood flow. In primates, Branston *et al.* [19] demonstrated that below a threshold value (12–16 ml/100 g/min), diminution in SSEP amplitude was linearly related to blood flow. It has also been proposed that when regional hypoperfusion occurs during MCA occlusion

Table 1. Comparison of the left and right hemispherical SSEP at different times during the study

Study times	Left hemisphere	Right hemisphere	*P*-value*
15 min occlusion	41.2 ± 35.0	85.7 ± 30.9	0.002
180 min occlusion	35.5 ± 31.6	76.5 ± 39.8	0.01
15 min re-perfusion	44.6 ± 39.2	70.9 ± 44.0	0.05
180 min re-perfusion	50.5 ± 42.3	72.9 ± 53.1	0.05

Values represent mean ± s.d. of the MCW amplitude (percentage of baseline).

*For comparison between left and right hemispheres.

Table 2. Infarct size (percentage of hemispheric cross-sectional area) in the nine cats studied

Animal	1	2	3	4	5	6	7	8	9
Infarct size	0	10	21	21	34	37	40	50	56

Mean ± s.d. 27.8 ± 19.6

lasting more than 15 min, ischaemic anoxic damage to some neurons prevents full recovery of evoked potentials during recirculation [20]. This was based on data derived from experiments in which the period of occlusion was brief (15–65 min) and recirculation was limited to 90 min or less. The results of our experiments, in which the duration of temporary MCA occlusion and recirculation were much longer, support the proposition that amplitude decrement is directly related to the extent of morphological damage. This relationship has been described previously in cats [11,12] from studies in which the middle cerebral artery was permanently occluded for 6 h, and our results supplement these. Furthermore, it appears that the early (15 min) amplitude is a reasonable predictor of the eventual infarct size, at least in this model.

In the present study, the intercept of the regression line on the y axis (Fig. 2a) indicates that when the SSEP amplitude at 15 min is less than 90% of baseline, an infarct will supervene. In a previous study in the cat by Traupe *et al.* [3], persistent cerebral hypoperfusion occurred after temporary MCA occlusion exceeding 1 h, whereas cerebral perfusion was normalized after shorter periods (15 and 30 min). On the basis of these sets of observations, we would hypothesize that animals with less than 90% amplitude at 15 min may be prevented from developing cerebral infarction by restoration of flow in the occluded vessel. Such flow restitution would be most beneficial within 15–30 min of occlusion, a time period compatible with normal post-occlusion cerebral perfusion.

The decrease in SSEP amplitude after clip release is most probably due to post-ischaemic hypoperfusion. This has been shown to be due to obstructive changes in the vasculature down-

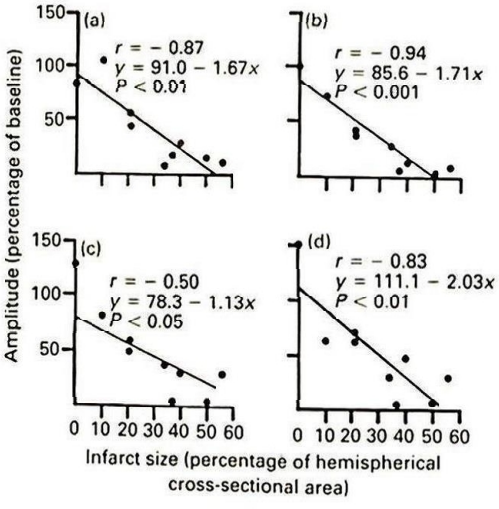

Fig. 2. Correlation between the size of infarcts and the amplitude of the left MCW after (a) 15 min occlusion, (b) 180 min occlusion, (c) 15 min re-perfusion, and (d) 180 min re-perfusion.

stream from an occluded vessel which prevent reflow after the vessel has been reopened [21–23].

In this acute study, there was some reduction in the amplitude of the major cortical wave in the contralateral (non-ischaemic) hemisphere, although this amplitude fared better than that in the ischaemic cortex. A similar reduction in amplitude was described in cats subjected to 6 h of permanent MCA occlusion [12]. This is at variance with the finding of Meyer *et al.* [11] of an increase in amplitude. However, it should be noted that in the latter study, the post-occlusion amplitudes were measured after the animals had been allowed to recover from anaesthesia. This distant effect of MCA occlusion may be due to the release of vasoactive substances from the ischaemic cortex [24] or may be explained by the concept of diaschisis [25,26]. In our preparations, the haemodynamic status, blood gases and temperature were maintained within physiological limits, and none of the infarcts was large enough to result in a shift of the midline.

It will be useful to study the neurological status of the animals which are allowed to recover after temporary focal cerebral ischaemia and to correlate neurological assessment with SSEP measurements and infarct size.

In conclusion, temporary middle cerebral artery occlusion in the cat often resulted in significant reduction in the amplitude of the major cortical wave of the somatosensory-evoked potential. The decrement persisted throughout the periods of occlusion and recirculation. There was a good negative correlation between the amplitude measured early during MCA occlusion and the eventual infarct size. A reduction in this amplitude in excess of 10% is likely to be associated with cerebral infarction.

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