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原　　著

Correlation of Cerebral Neuronal Activity and Oxygen Consumption During Ether and Pentobarbital Aneshesia in Dog

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Summary

In an attempt to verify the hypothesis that alteration of cerebral oxygen consumption ($CMRo_2$) by anesthetics is secondary to that of cerebral neuronal activity, effects of ether and pentobarbital on the cerebral electrical activities and $CMRo_2$ were studied in sixteen dogs. The cortical EEG and midbrain reticular multi-unit activity (R-MUA) were recorded, and changes in the $CMRo_2$ were plotted against those of R-MUA. Increments in the dose of pentobarbital initially induced slowing of the EEG, a gradual flattening followed and finally there was total silence. In contrast, ether initially induced rhythmic slow waves, followed by gradual flattening and finally by a high amplitude high frequency epileptiform EEG. The EEG changes induced by pentobarbital were associated with a gradual decrease in R-MUA, while those by ether were associated with initial increases followed by a progressive decrease. The changes in $CMRo_2$ closely paralled those of R-MUA: the enhancement and depression in the neuronal activities were associated with increase and decrease in $CMRo_2$. The rate of decrease in $CMRo_2$ per unit degree of depression of R-MUA was greater in the case of pentobarbital. When extrapolating the regression line in R-MUA-$CMRo_2$, the correlation indicated a significant quantity of residual $CMRo_2$ at the zero level of R-MUA, thereby suggesting that $CMRo_2$ consisted mainly of two components: the neuronal activity-dependent or electrical process-related, and the neuronal activity-independent or non-electrical one. The residual $CMRo_2$ in the case of ether was greater than that seen with pentobarbital. The greater $CMRo_2$ at a given level of R-MUA depression and the greater $CMRo_2$ at the zero level of R-MUA were both attributed to metabolic processes consumed for the cortical epileptiform EEG. A review of previous studies on action of anesthetics on the CNS electrical activities indicated that the present hypothesis could not be verified using available conventional neurophysiological techniques and anesthetics.

Key words: Anesthetics, Ether, Pentobarbital, $CMRo_2$, CNS electrical activity.
索引語：麻酔薬，エーテル，ペントバルビタール，脳酸素消費量，脳電気活動.
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The depth of anesthesia represents the degree of depression of the functional performance of the brain. This depression has been correlated by Faulconer and Bickford[1] to a progressive slowing of the cortical EEG and increments in arterial blood levels of anesthetic. In contrast, a correlation with changes in cerebral metabolism, for example oxygen consumption ($CMRo_2$), has not been confirmed. A dose-related depression of $CMRo_2$ has been confirmed in the case of barbiturates[9,11], halothane[8,9,21,23], enflurane[19] and isoflurane[22], but not with ether[8], cyclopropane[12] and ketamine[22]. These drugs have relatively little effect on cerebral metabolism, or may even enhance it, depending on the concentrations. When compared with regard to an equal degree of depression of the functional performance of brain, MAC, the depression seen with enflurane is reportedly greater than that produced by other inhalation anesthetics[21].

In previous studies, Mori and collegues found that although clinical doses of anesthetics induce cortical EEG slowing, in common, their actions on the neuronal firing of the brain stem reticular core, the putative substrate for anesthetic actions[2], were divergent: pentobarbital, halothane and enflurane significantly depressed the reticular neuronal firing, while ether[15,16], cyclopropane[13] and ketamine[14] increased the firing, to various degrees, particularly in case of clinical doses. These tendencies closely parallel the actions of these agents on the cerebral metabolic processes mentioned above.

The present study was an attempt to verify our working hypothesis, that the determinant of cerebral metabolic requirement, such as $CMRo_2$, is the level of neuronal activity of the brain, i.e., the anesthetic action is exerted primarily on the CNS neuronal activity and that alteration in metabolic requirements is a secondary phenomenon. For this purpose, pentobarbital and ether were chosen to represent the two extremes: pentobarbital depresses $CMRo_2$, in a dose-related manner, while ether produces no such depression, in clinical dose ranges. The dose ranges we used were thus extended far above the clinical ones. Some of the data were reported in a preliminary note[4].

Materials and methods

Sixteen mongrel dogs of either sex, weighing 10–15 kg, unmedicated and fasting, were used. Two weeks prior to the drug study, brain electrodes were implanted following administration of pentobarbital (20 mg/kg, iv initially, and then supplemented as required). The cortical electrode consisted of a stainless steel screw, 2.0 mm diameter, and was placed so as to reach the dura over the frontal cortex. A similar electrode placed in the frontal bone was used as a reference. Side-by-side parallel stainless steel wire electrodes, 0.2 mm diameter and insulated with epoxylite resin, except for the cut end, were implanted in the midbrain reticular formation of both sides at the point, rostral 12, lateral 4 and depth 20, according to the dog brain atlas of Lim et al.[7]. All electrodes were soldered to a miniature vacuum tube socket, which was fixed to the skull with dental cement. Anesthesia was induced with a sleeping dose of thiopental, 10–15 mg/kg, and maintained throughout the surgical procedure with halothane, 1% in 75% nitrous oxide in oxygen. Gallamine triethiodide, 100–150 mg iv, was administered prior to tracheal intubation. The femoral artery was cannulated for arterial blood pressure monitoring and for blood sampling.

The femoral and the cephalic veins were cannulated for replacement of blood from the sagittal sinus and for drug administration. Arterial P_{CO_2} and pH were adjusted in the ranges, 40–45 torr and 7.35–7.45, by adjusting the ventilatory volume and by administration of $NaHCO_3$. The rectal temperature was maintained at 37–38°C by use of a warm water blanket.

With the head fixed on a stereotaxic frame, the dog was placed in a prone position. A large craniotomy was made to expose the sagittal sinus and to isolate the emissary veins and anterior ethmoidal vein. After heparinization (300–400 units/kg iv), the posterior sagittal sinus was cannulated with a tapered catheter, posterior to which Oxycel® was packed. Blood flow from the sagittal sinus catheter was collected in a reservoir placed at the level of base of skull, and then returned via the femoral vein using a pump (Junken Co.). The arterial and sagittal sinus blood samples were analysed for P_{O_2}, P_{CO_2} and pH by direct reading electrode (I.L. ABL-1). Hemoglobin was measured by the cyan-methemoglobin method. Thus, the oxygen content was calculated from P_{O_2}, Hb, and percent saturation. $CMRo_2$ was calculated as the product of cerebral blood flow and the arterial-sagittal sinus blood oxygen content difference (otherwise see Michenfelder et al.[10]). At the end of each experiment the brain was removed and weighed to calculate cerebral blood flow (CBF) in ml/100 g of brain/min. Serial sections of the brain stem were made to confirm the reticular electrode positions.

The EEG activity was recorded by the conventional method. The reticular neuronal activity was measured by the method of recording multi-unit activity, as described previously[14,16], Briefly, the electrical activities obtained through the reticular electrodes were passed through a high frequency band-pass filter, centered at 1,300 Hz, and attenuated by 5-db at 900 and 3,500 Hz, and by 40-db at 400 and 8,000 Hz. Thus, the activities of EEG frequency range were filtered out, and only neuronal action potentials were picked up. The filtered activity was rectified and averaged with a smoothing rectifying circuit, was entered at the DC stage input of a polygraph (Nihonkoden) and was recorded simultaneously with the cortical EEG. In order to show the slow shift of the levels of multi-unit activity, a straight writing slow oscillograph (Sanei 8S) was also used. With this method, a rise in DC voltage indicated an increase and a fall a decrease in the neuronal firing of a population of units included in an area of approximately 1.0 mm radius around the electrode tip[3].

After completion of the surgical procedures (30–40 min), halothane was discontinued and anaesthesia was maintained with nitrous oxide, 75% in oxygen, for 20 min. Then, three sets of control measurements were made for $CMRo_2$, after which nitrous oxide was discontinued and either pentobarbital or ether administered immediately. Increments in either anaesthetic were given while confirming a definite change of 5–10% in the level of ongoing reticular multi-unit activity. The determinations of $CMRo_2$ were made at various levels of R-MUA. When the mean arterial pressure dropped below 70 mmHg, with deep anaesthesia, methoxamine, 0.2 mg/ml saline, was infused intravenously. The arterial blood concentration of ether was measured in 4 dogs using a gaschromatograph.

The mean values of multi-unit activity and $CMRo_2$ prior to the administration of either anesthetics served as controls, in each experiment, and the changes induces by anesthetics were

expressed by the percent of changes from the controls. These measurements were made when an apparent change of 5–10% in the multi-unit activity was noted on visual inspection. The percent changes of $CMRo_2$ against those of reticular multi-unit activity were plotted, and the regression lines were obtained by the minimum square method.

Results

In addition to the changes in cerebral electrical activities and $CMRo_2$, the various systemic values changed to various degrees during administration of either of the anesthetics (Table 1). At the end point of studies, statistically significant changes were observed in $Paco_2$, arterial blood pH and mean arterial pressure, despite vigorous adjustment of ventilatory volumes and administration of sodium bicarbonate.

Ether

The initial EEG change induced by ether was represented by desynchronization, and was followed by rhythmic slow waves of 2–3 Hz, very high amplitude slow waves of 0.5–1 Hz, irregular slow waves, and finally the so-called epileptiform activity of maximum amplitude repetitive spikes. These EEG changes were associated with intial increase by 11.7±2.3% of control ($p<.001$) and succeeding marked decrease, to various degrees, in the R-MUA levels (Fig. 1). When EEG

Table 1. Values for Certain Variables and $CMRo_2$ before and after Anesthetic Administration to dogs. The value of arterial ether concentration was studied in 5 dogs, the other values are those from 9 dogs for ether and 7 dogs for pentobarbital. (m±sem)

	before	end point	p
Ether			
Pao_2 torr	185±10	198±19	NS
$Paco_2$ torr	38±1	62±4	.001
pH	7.32±0.01	7.12±0.05	.001
Hb g/dl	11.6±0.6	11.5±0.7	NS
MAP torr	129±4	80±4	.001
Body temp.	38.7±0.5	38.4±0.5	NS
$CMRo_2$	4.53±0.34	3.78±0.32	.001
Final arterial blood concent. mg/dl		317±31	
Pentobarbital			
Pao_2	137±17	145±12	NS
$Paco_2$	37±2	44±3	.02
pH	7.32±0.03	7.29±0.05	.02
Hg g/dl	11.0±1.1	9.2±1.0	.001
MAP torr	108±11	93±10	.05
Body temp.	37.9±0.6	37.5±0.6	NS
$CMRo_2$	4.07±0.4	2.40±0.33	.001
Final cumulative dose mg/kg		39±3	

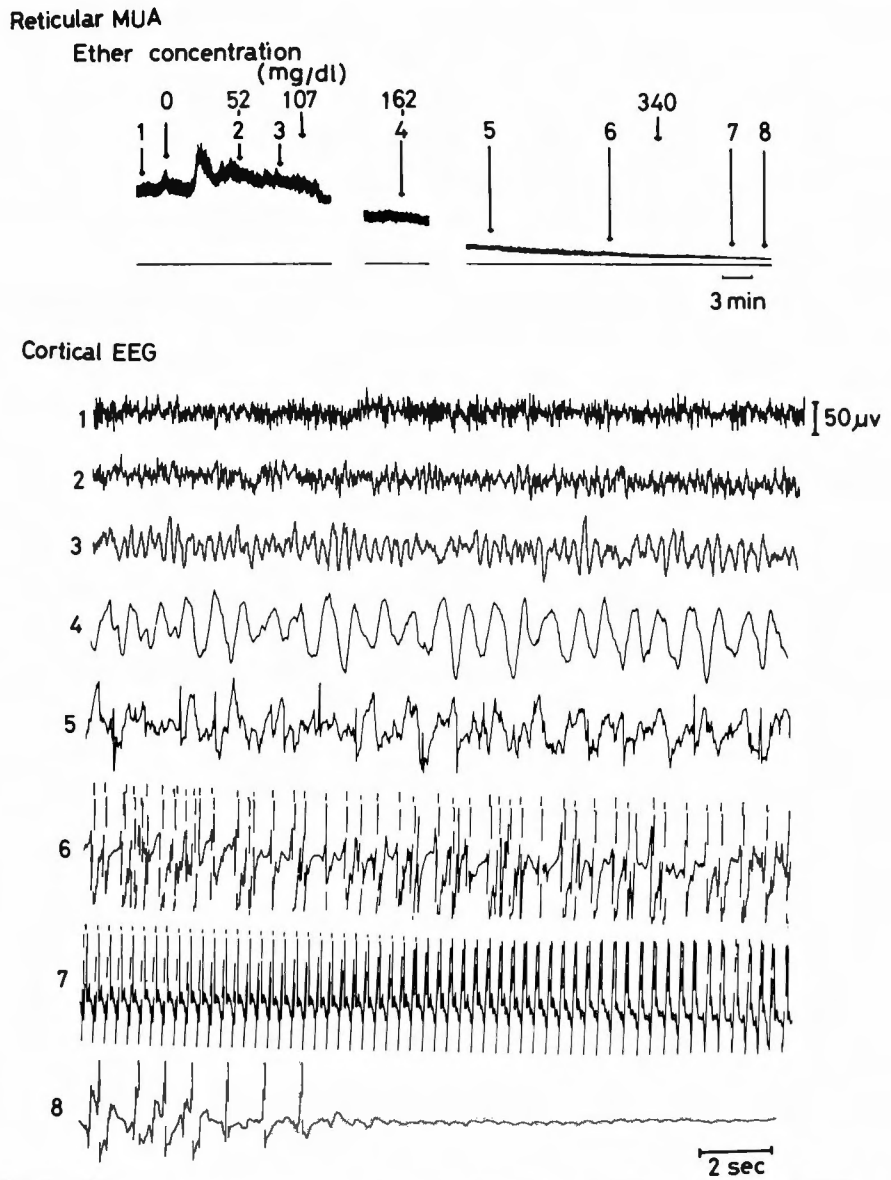

Fig. 1. [9]Effects of ether on the reticular multi-unit activity (R-MUA) and the cortical EEG in a dog. The upper figure shows the R-MUA and the lower figure the EEG. The arabic numbers, 1–8, in the upper figure correspond to those of the lower figure; 1 represents control and the succeeding numbers represent those during ether administration. The arabic numbers, 0–340, represent the arterial blood ether concentrations in mg/dl. The R-MUA is illustrated in amplitude demodulated signal: the upward deflection indicates an increase of firing of neurons, and the downward deflection shows its decrease. The line beneath the R-MUA record represents the level of absence of neuronal firing, which was obtained after discontinuation of ventilation and the death of animal. Ether induced high amplitude rhythmic slow waves (3, 4) with clinical blood concentrations, and high amplitude epileptiform activity (6, 7) with massive concentrations and which was followed by post-ictal depression. The R-MUA increased initially (2, 3), and then decreased in the stage of seizure (6, 7).

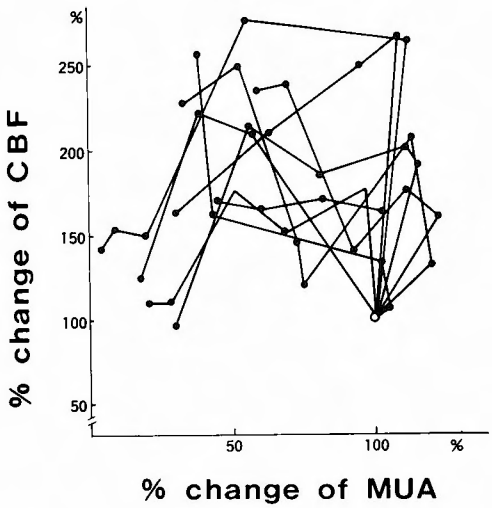

Fig. 2. Individual CBF values plotted against reticular multi-unit activity (R-MUA). The abscissa represents the percent change of R-MUA and the ordinate that of the CBF. There was no correlation between the level of neuronal activity (R-MUA) and the CBF.

seizure appeared in the cortical records, the R-MUA was markedly depressed, and there appeared to be no further significant changes.

There was no correlation between the level of R-MUA and CBF. The level initially increased to 150–180% of control value, then decreased with a fall in the arterial blood pressure. However, administration of methoxamine iv restored the CBF to the control level or above (Fig. 2).

Following administration of ether, when the R-MUA increased, the $CMRo_2$ increased in 5 dogs and decreased in 3, the average being an increase by $14.4 \pm 4.5\%$ of control ($p<.02$).

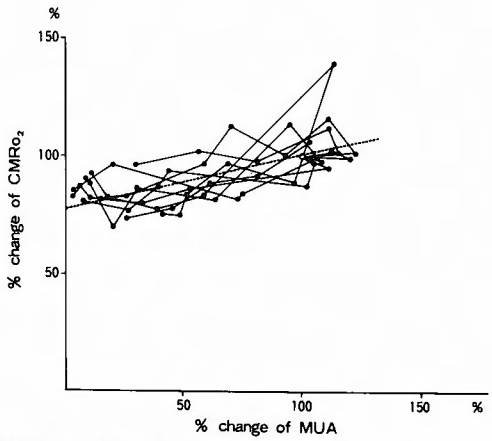

Fig. 3. Individual $CMRo_2$ values (percent of control) plotted against reticular multi-unit activity (R-MUA). The abscissa represents the percent change of R-MUA, and the ordinate that of $CMRo_2$. The broken line represents regression line: $Y=0.21X+78.6$. In each dog, an initial increase in $CMRo_2$ was followed by a progressive decrease.

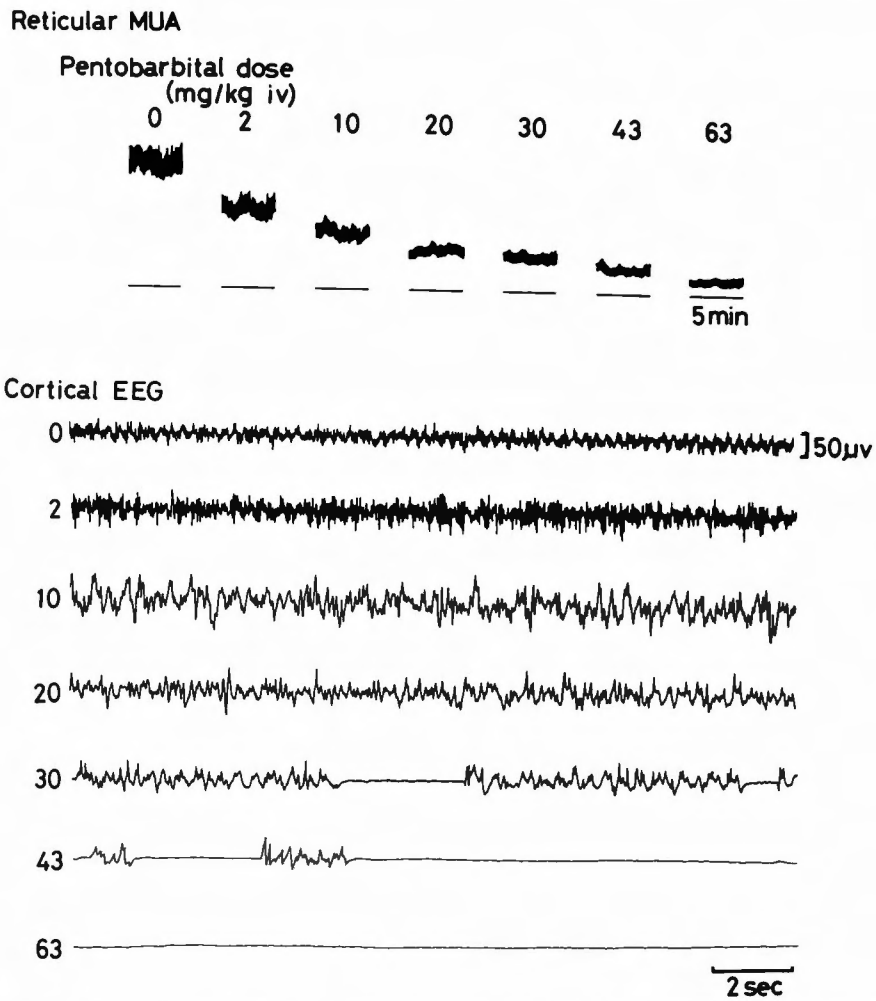

Fig. 4. Effects of pentobarbital on the reticular multi-unit activity (R-MUA) and the cortical EEG in a dog. The upper figure shows the R-MUA, and the lower figure the EEG. The arabic numbers in both figures represent the incremental dose of pentobarbital iv. Otherwise descriptions are the same as in figure 1. Pentobarbital initially induced high amplitude slow waves in the EEG and which were followed by a gradual flattening, and finally a total electrical silence. These EEG changes were associated with a gradual decrease in the R-MUA.

The succeeding decrease in R-MUA was associated with a linear decrease in the $CMRo_2$ (Fig. 3). The general trend of decrease in R-MUA was associated with a decrease in $CMRo_2$: the slope of regression line of $CMRo_2$ against R-MUA was 0.21 ($r=0.63$; 95% confidence interval: 0.16–0.26), and the intersect at the zero line of $CMRo_2$ was 78.6% of control (95% confidence interval: 75.4–81.9) (Fig. 3).

Pentobarbital

Administration of incremental doses of pentobarbital iv induced a definite slowing of EEG

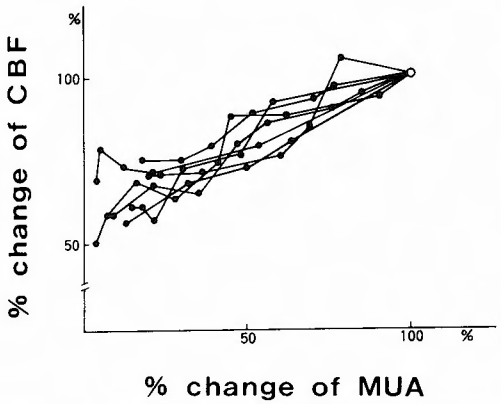

Fig. 5. Individual CBF values plotted against R-MUA. The abscissa represents the percent change of R-MUA, and the ordinate that of the CBF. A linear decrease in the CBF was noted in relation to the decrease in the R-MUA.

and which was accompanied initially by an increase in the amplitude, then a decrease, and finally an iso-electric pattern appeared (Fig. 4).

Increments in the pentobarbital iv induced a dose-related decrease in the CBF and $CMRo_2$ (Fig's 5 and 6). The slope of the regression line of $CMRo_2$ against the R-MUA was 0.45 ($r=0.77$; 95% confidence range of 0.38–0.52), and its intersect at the zero line of R-MUA was 58.5% of control (95% confidence range of 55.4–61.6).

Comparison of the regression lines

The significance of difference of regression lines between ether and pentobarbital was studied statistically. The analysis of covariance showed a significant difference between these two lines at ${}_{95}^{1}F=12.18$ ($p<.001$).

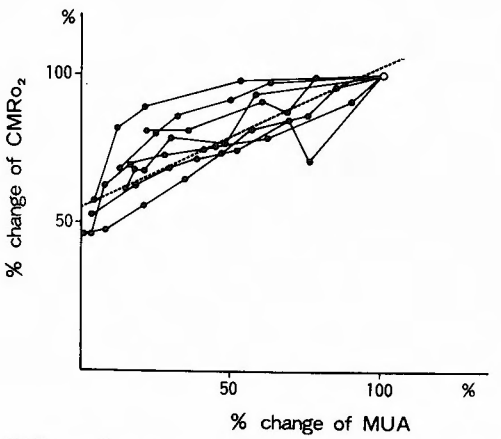

Fig. 6. Individual $CMRo_2$ values (percent of control) plotted against R-MUA (percent of control) following pentobarbital iv. The abscissa represents the percent change of R-MUA, and the ordinate that of $CMRo_2$. The broken line represents regression line: $Y=0.45X+58.5$. In each dog the decrease in R-MUA was associated with a linear decrease in $CMRo_2$.

Discussion

The present study on dogs, confirmed our previous findings in cats regarding the actions of ether and pentobarbital on the CNS electrical activities: ether induced rhythmic (hypersynchronous) slow waves associated with relatively unchanged or even slightly increased levels of reticular neuronal firing, with a clinical depth of anesthesia, and high amplitude epileptiform EEG associated with marked depression of reticular neuronal firing, in the profoundly deep plane. Incremental doses of pentobarbital induced a gradual EEG slowing and then a flattening associated with a linear decrease of reticular neuronal firing[15-17].

Although statistically significant changes were induced in the values of systemic acid-base balance, these changes did not significantly alter the values of $CMRo_2$: changes in $Paco_2$ induce alteration in CBF but not in $CMRo_2$[6]; changes in the arterial blocd pH do not alter either CBF or $CMRo_2$[5].

The $CMRo_2$ measured by the method used in the present study represents the sum of the values derived from the fronto-parietal cortices[10]. Instead of recording the neuronal firing in the reticular formation, an ideal approach would be the measurement of mean of the neuronal firing, in various cortical areas. However, it is also known that electric stimulation of a given small area of the midbrain reticular formation induces a diffuse generalized EEG activation over the entire cerebral hemisphere[18], and the level of neuronal activity of a population of units in the reticular formation can be taken to represent an average of the activity in the cerebral cortex.

The present study provided definite evidence to support our working hypothesis, that is, with pentobarbital, the cerebral metabolic requirement is depressed in parallel with the depression of reticular neuronal activity. In contrast, although a similar and highly reliable ($p<.01$) regression line was also obtained with ether, the slope differed from that related to pentobarbital. If the level of neuronal activity is the determinant for metabolic requirements of the brain, the slope of regression lines should be identical with these two anesthetics. Nevertheless, the present study did not necessarily invalidate of our hypothesis because we noted that when spikes appeared on the isoelectric EEG, phasic enhancement appeared only in the cortical multi-unit activities and which were otherwise markedly suppressed, while neither EEG spikes nor phasic enhancement in the multi-unit activity appeared in the reticular records[15,16]. The lesser depression of $CMRo_2$ per a given degree of depression in R-MUA and a greater residual $CMRo_2$ at the zero level of R-MUA in the case of ether should be due to the metabolic processes related to the cortical EEG spikes.

Of particular interest in the present study was the presence of a significant level of residual $CMRo_2$ when the neuronal firing was completely blocked. This indicates that the oxygen consumption of the brain probably consists of at least two components, i.e., the activity-dependent (electrical) and activity-independent (non-electrical) metabolic processes. This view supports the postulate of Michenfelder[9] who showed a residual $CMRo_2$ when the cortical EEG showed an iso-electric pattern. The activity-independent oxygen consumption may result from metabolic processes in glial cells and/or that which are necessary to maintain the resting state of neurons.

The relative resistance of the non-electric process of $CMRo_2$ to the anesthetics discussed by Michenfelder[9] was not given attention in present study.

Another point of interest was the dissociation of EEG-$CMRo_2$ coupling during seizure: the high amplitude EEG seizure was associated with minimum increases in oxygen requirements, thereby indicating that the EEG is not the determinant of cerebral metabolic requirements. Using thiopental, Michenfelder[11] proposed a similar postulate with regard to EEG criteria. However, since EEG activity was not related directly to the neuronal firing[20], and could not be measured quantitatively, he showed the data as a function of drug dosage and discussed it with the presence or absence of cortical EEG activities. One example of the incompatibility of applying EEG to such a study was given by Matsumoto[8], who showed only the phase of $CMRo_2$ elevation in association with the EEG slow waves by ether and did not provide evidence for the deeper planes of anesthesia, as was done in the present study. The drawback of the present study was that ether did not induce a total cortical electrical silence, albeit an inherent nature of ether. Here, data on either ketamine or cyclopropane could be considered: at the stage of EEG slow waves, the depression of $CMRo_2$ by cyclopropane is much less than thiopental[9], and ketamine enhances it[22]. However, neither agent can be used for a reference since cyclopropane does not produce significant depression in the R-MUA at the stage of cortical EEG silence[13,16], and ketamine induces electrographic seizure at the deepest stage[14,16].

In summary, the present study shows that while conventional neurophysiological techniques do not provide conclusive evidence they do lend considerable support to the hypothesis that the determinant of metabolic requirement altered by anesthetics is secondary to the altered neuronal activity.

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和文抄録

エーテル及びペントバルビタール麻酔中の犬脳酸素消費量と脳電気活動の相関性

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播　磨　晃　宏

犬を用い，ペントバルビタールとエーテルの脳電気活動と脳酸素消費量に及ぼす影響を検討した．これら麻酔薬による中脳網様体の多ニューロン活動の変化は，脳酸素消費量の変化とよい相関を示した．即ち，エーテルによるニューロン活動の増加，及びエーテル或はペントバルビタールによるニューロン活動の減少は，それぞれ脳酸素消費量の増加及び減少を伴っていた．然し，深麻酔によって，中脳網様体多ニューロン活動が完全に抑制された時点においても，脳酸素消費量は相当量が残存していることが明らかとなり，このことから，脳酸素消費量が二つの要素，即ち，脳の電気現象，或はニューロン活動と関連するもの，及びそれらと無関係なものから成ることが示唆された．