Correlation of Blood Levels of 4-Hydroxybutyrate with State of Consciousness

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Blood levels were correlated with clinical observations following the intravenous administration of 4-hydroxybutyrate in the unpremedicated patient. The blood level rose rapidly immediately following injection and then fell in a biphasic fashion. The peak clinical effect lagged behind the peak blood levels suggesting activity by means of a metabolite rather than the parent compound.

There is evidence to suggest that the intercurrent administration of thiobarbiturate produces a rise in the blood level of 4-hydroxybutyrate.

Normal metabolic intermediates have very rarely been found to exert significant physiologic effects when administered parenterally. It is therefore of interest that 4-hydroxybutyrate, recently reported by Laborit and co-workers to have anesthetic properties, is actually a normal metabolite of brain. In 1956 Roberts and his co-workers and Awapara discovered gamma-aminobutyric acid in mammalian brain. This was found to arise through the specific enzymatic breakdown of glutamic acid present in large quantities in brain. Gamma-aminobutyric acid was considered to be an end product active in transmission of the inhibitory impulse: it has long been viewed as a repressor substance in brain. Despite statements to the contrary, gamma-aminobutyric acid has never been shown conclusively to enter the brain from the blood. Attempts to use it to suppress central nervous system activity have been unsuccessful. The finding that gamma-aminobutyric acid undergoes transamination to a chemically active compound in brain tissue opened up a series of investigations involving the function of pyridoxine and its active phosphorylated forms in relation to convulsive disorders. The pyridoxine coenzymes are required both for the formation and transamination of gamma-aminobutyric acid.

Early work on the transamination of gamma-aminobutyric acid gave evidence that another compound must be formed in brain. This compound has now been identified as 4-hydroxybutyric acid. It is of interest that this material is formed and degraded through a pathway leading to gamma-aminobutyric acid. It is possible that 4-hydroxybutyric acid is the active metabolite. As a result of the experimental work leading to the finding of 4-hydroxybutyrate in brain, a method for its determination was developed which permits the measurement of blood levels of 4-hydroxybutyrate in relation to the state of consciousness.

In view of the enthusiastic reports on this compound by Laborit and his co-workers and by Blumenfeld, Suntay and Harmel, it was deemed appropriate to study hypnotic effect simultaneously with blood levels of 4-hydroxybutyrate. In this way, it was hoped to correlate the state of consciousness with blood levels of the compound. It is apparent from the observations described herein that there is a close parallelism between the increment of 4-hydroxybutyrate in the blood and the mental state of the patient.

Method

Sixteen healthy adult patients were studied for periods varying from 20–300 minutes following the administration intravenously of 4-hydroxybutyrate. No premedication was used and no other drugs were administered during the period of study. Table 1 includes details

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of the patients, dose of the compound and condition for which patient was admitted.

A 15 gauge plastic needle was inserted into a forearm vein and connected via a 3-way stopcock to a bottle of heparinized saline. Venous blood samples were drawn into a fresh syringe prior to the injection of the drug and subsequently at 15 minute intervals. A second slow intravenous infusion of normal saline in the opposite arm was employed for 4-hydroxybutyrate administration to avoid contamination of the “sampling vein.”

Urine samples were collected over a 24 hour period during the first few studies, but were discontinued when it became evident that less than 2 per cent was being excreted by this route. Samples were analyzed using a modification of the colorimetric analysis of protein free filtrates reported by Fishbein and Bessman and recorded in micromoles/ml.

An effort was made to compare the effects of small, moderate and large doses both from the standpoint of clinical response and blood levels. There was further effort to compare the blood levels obtained and maintained by the single injection technique, intermittent injection and continuous drip following a priming dose. Blood pressure, pulse and respiratory rate were recorded every 5 minutes. Tidal volume was measured at appropriate intervals employing a Wright respirometer. However, periodic breathing which occurred in a large proportion of the patients made respiratory measurements difficult to interpret.

Immediately following the withdrawal of samples the following stimuli were applied and the response noted: voice, touch, pin prick, deep pressure, conjunctival reflex, oropharyngeal reflex, laryngeal reflex, and carinal reflex.

Three levels of sleep were recorded based on response to the above stimuli:

Deep Sleep. No response to any of the stimuli applied.

Moderate Sleep. Spontaneous blinking occurred and response occurred to deep pressure.

Light Sleep. Spontaneous movements occurred with occasional opening of eyes.

The drug acted as a general depressant agent with a sedative-hypnotic effect and was in no way a total anesthetic. Even at the deepest levels of sleep it was never possible
to apply a surgical stimulus without producing reflex movement.

Results

Using the smallest dosage, 50 mg./kg., the blood level did not rise above 1.75 micromoles/ml. With the largest dose 165 mg./kg., a peak level of over 4.0 micromoles/ml was obtained. Table 2 outlines the concentrations in relation to level of consciousness. Sample curves are illustrated in figures 1, 2, 3, and 4. Fourteen patients received doses of 100 mg./kg. with resulting peak blood levels ranging from 2.25 to 5.0 micromoles/ml, the mean being 2.95 micromoles/ml.

The typical blood level curve showed a high initial peak in the first 15 minutes followed by a fall which was biphasic in type. This fall was initially steep for a period of approximately 60 minutes, then less steep for the remainder of the study period.

The greatest clinical effect lagged behind the peak blood levels by about 15 minutes,

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Average

* Followed by 10 doses of 0.6 g. each.
† Followed by intravenous dose of 2.0 g./hour.
A—Awake; L—Light sleep; M—Medium sleep; D—Deep sleep.
A/L P = 0.01 L/M P = 0.01 M/D P = 0.05

![Fig. 1](http://example.com/fig1.png)

**Fig. 1.** Levels of 4-hydroxybutyrate in blood following the administration of 50 mg./kg. (total dose—3.6 g.) and 100 mg./kg. (total dose—6.5 g.).

![Fig. 2](http://example.com/fig2.png)

**Fig. 2.** Levels of 4-hydroxybutyrate in blood following the administration of 165 mg./kg. (total dose—6.0 g.).
sustaining that a metabolite of 4-hydroxybutyrate may be the active compound. This is likely because it has already been shown that the lactone form of 4-hydroxybutyrate is apparently the active form in hypnosis in rats. The evidence presented suggested that the lactone is formed in the liver and that the actual brain content of the acid form is not related to sleep.

Clinically, three merging levels of sleep could be identified and we were able to relate these to blood levels.

With a level of over 2.5 micromoles/ml a state of deep sleep was seen with no response to touch, pin-prick or deep pressure and no response to skin preparation or vaginal examination. Blinking ceased, the pupils being small, the eyes central and fixed. There was abolition of pharyngeal and laryngeal reflexes but there was no abolition of reflex response to surgical incision, stimulation of the carina or traction on the peritoneum.

With levels of 2.5 down to 1.5 micromoles/ml moderate sleep was present, spontaneous blinking occurred and responses occurred to deep pressure.

Blood levels of 1.5 down to 0.5 micromoles/ml were found to correspond with light sleep during which spontaneous movements with occasional opening of eyes were seen. With patients awake the blood levels were below 0.5 micromoles/ml.

It was noted in 3 cases that the administration of intravenous thiopental resulted in a further rise in the blood level of 4-hydroxybutyrate. While the number of observations is small, the relation suggested is important and is being studied further.

**Discussion**

As might be expected, the peak concentration of 4-hydroxybutyrate in blood is proportional to the size and rate of the dose administered. The fall in blood levels then
follows a biphasic pattern which would seem to indicate initially a general redistribution of the compound in the various body tissues, followed by a longer period of metabolic degradation. The negligible quantity (less than 2 per cent) recovered in the urine would suggest that the compound is almost completely metabolized.

The findings described by other investigators of a delay following administration of 4-hydroxybutyrate until the onset of full clinical effect was confirmed in our study where maximal clinical response followed the peak blood level by a period of 15 minutes. This is fair evidence that some metabolite rather than the parent compound is responsible for the effect. From reports with rats, it seems most likely that the active metabolite is the lactone. The data of Bessman and Skolnik also show that peak brain levels of 4-hydroxybutyrate parallel blood levels but do not reach more than 50 per cent of blood concentrations. This supports the concept of a diffusion gradient or blood-brain barrier effect. The lag in the human response might be related to this phenomenon, but in view of the data cited above, which shows the 4-hydroxybutyrate level in the brain to be at a maximum when the animals awaken, suggests that metabolic transformation is the more likely explanation. Further work in progress on the actual concentrations in human brain should clarify this point.

Summary

An attempt was made to correlate blood levels with clinical observations of narcosis following intravenous administration of 4-hydroxybutyrate in the unpremedicated patient. The blood level rose rapidly immediately following injection and then fell in a biphasic fashion.

The peak clinical effect lagged behind the peak blood levels suggesting activity of a metabolite rather than the parent compound.

There was a suggestion that the concurrent administration of thiobarbiturate produces a rise in the blood level of 4-hydroxybutyrate.

References


SUXAMETHONIUM MUSCLE PAIN Pain following suxamethonium was observed in 83 per cent of 300 patients and in 59 per cent was so severe as to be distressing to the patients. When suxamethonium was preceded by an intravenous injection of 3 mg. of tubocurarine only 20 per cent developed pain and this never was severe. A simple correlation between fasciculations and pain was not found. Development of pain was independent of the dose of suxamethonium. In a small group of patients who received suxamethonium twice within one week, only one developed pain after the second anesthesia. (Bennike, K., and Nielsen, E.: Muscle Pain Following Suxamethonium, Danish Med. Bull. 11: 122, 1964.)