(1.26-1.42 mmol/l; 5.04-5.68 mg/100 ml).⁵ Those workers used a trometamol (TRIS) buffer which is highly temperature dependent, whereas this is not the case for the imidazole buffer that we employed. Their buffer was made up to pH 7.4 at 37°C but used at room temperature. This would produce a rise in pH to roughly 7.7 at 25°C, which would be associated with a fall of approximately 0.15 mmol/l (0.6 mg/100 ml) in the measurement of CaD. As the mean of their reference range was 0.125 mmol/l (0.5 mg/100 ml) less than ours this technical difference may account for the differences in the normal ranges. This underlines the critical importance of the pH of the sample at which CaD is measured.

Another problem is that currently there is no internationally recognised reference standard for CaD. Our approach has been to calibrate the assay system against a solution of calcium carbonate, but this is open to criticism as it does not allow for the Donnan effect of protein. Nevertheless, in view of the fact that large changes in serum albumin concentration induced by venous stasis had no effect on CaD, the Donnan effect would appear to have no practical relevance. Although the problem of standardisation may be relevant to interpretation of results among different laboratories, once a standard has been developed within a particular laboratory the results will be internally consistent. Furthermore, if this method is widely accepted it should be possible to develop an acceptable international standard.

In favour of the use of CaD rather than other methods of calcium estimation is that the method is a simple modification of a continuous flow total calcium technique using equipment that has been tried and tested over many years and is readily available. It correlates well with CaI but has none of the practical problems associated with this technique. Although under many circumstances CaA gives a clinically equivalent result, calculation

requires the measurement of both CaT and albumin. Now that it is 50 years since the physiological irrelevance of protein associated calcium was first commented on it is time to move away from measuring CaT. In busy biochemical laboratories CaD estimation is an efficient and cost effective answer.

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References

- McLean FC, Hastings AB. A biological method for the estimation of calcium ion concentration. *J Biol Chem* 1934;**107**:337-50.
- Toffaletti J, Savory J, Gitelman HJ. Continuous-flow determination of dialysable calcium in serum. *Clin Chem* 1977;**23**:1258-63.
- Moore EW. Ionized calcium in normal serum, ultrafiltrates and whole blood determined by ion exchange electrodes. *J Clin Invest* 1970;**49**:318-34.
- Toffaletti J, Bowers GN. Improvement in and clinical utility of a continuous-flow method for routine measurement of dialysable (ultrafiltrable) calcium. *Clin Chem* 1979;**25**:1939-43.
- Toffaletti J, Kirvan K. Spectrophotometric micromethod for measurement of dialysable calcium by use of cresolphthalein complexone and continuous-flow analysis. *Clin Chem* 1980;**26**:1562-5.
- Anonymous. Correcting the calcium. [Editorial.] *Br Med J* 1977;**i**:598.
- Drop LJ, Laver MB. Low plasma ionized calcium and response to calcium therapy in critically ill man. *Anesthesiology* 1975;**43**:300-6.
- Payne RB. Clinically significant effect of protein concentration on ion-selective electrode measurements of ionized calcium. *Ann Clin Biochem* 1982;**19**:233-7.
- Conceicao SC, Weightman D, Smith PA, Luno J, Ward MK, Kerr DNS. Serum ionized calcium concentration: measurement versus calculation. *Br Med J* 1978;**i**:1103-5.
- Toffaletti J. Total, ultrafiltrable, dialysable and ionised calcium measurements in randomly selected samples compared with clinical diagnoses. *Ann Clin Biochem* 1983;**20**:349-52.
- Ladenson JH, Lewis JW, Boyd JC. Failure of total calcium corrected for protein, albumin and pH to correctly assess free calcium status. *J Clin Endocrinol Metab* 1978;**46**:986-93.

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The role of endogenous opiates in thermal regulation of the body during exercise

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Abstract

Naloxone abolished the rise in body temperature seen after bicycle ergometer tests performed by 10 healthy men. This suggests that endogenous opiates play a part in thermal regulation during muscular exercise.

Introduction

The effect of opiates on core temperature has been the subject of many studies. Evidence shows that endogenous opiates, in

particular endorphins, have a role in the central control of body temperature.¹⁻⁴ Because body temperature rises during dynamic muscular exercise we were intrigued to know whether endogenous opiates play a part in this phenomenon. We therefore studied the effect of naloxone (an opiate antagonist) on the changes in body temperature induced by exercise.

Subjects, methods, and results

Ten healthy men aged 22-28 years, who regularly participated in various sporting activities, underwent a graded ergometer test on three occasions. They exercised on a bicycle ergometer with electric brakes (Ergometric systems 380B, Siemens Elema, Sweden), and on each occasion an identical protocol was used. The tests were performed in the same laboratory at constant room temperature and humidity, at the same time of day, and with an interval of at least four days between each test for each subject.

The first test served as a control. In the second and third tests either placebo (5 ml saline) or 2 mg naloxone (naloxone hydrochloride 0.4 g/l, Dupont de Nemours, Belgium) was administered intravenously five minutes before exercising. The order of the placebo and naloxone tests was randomised and double blind; five subjects received placebo first and five received naloxone first to minimise carryover effects. The temperature in the laboratory was kept constant at 20°C and the humidity at 60%. Sublingual temperature was

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Mean (SD) sublingual temperatures ($^{\circ}\text{C}$) before, immediately after, and one hour after exercise in 10 healthy men given placebo or naloxone five minutes before exercise

	Control	Placebo	Naloxone
Before exercise	36.85 (0.24)	36.64 (0.28)	36.58 (0.29)
Immediately after	37.33 (0.26)	37.16 (0.27)	36.40 (0.54)*
An hour after	36.83 (0.24)	36.67 (0.34)	36.34 (0.41)*

* $p < 0.001$ compared with control and placebo studies.

measured with a simple thermometer before exercise, immediately afterwards, and one hour later. Statistical study of results was performed by two way analysis of variance.

There were no significant differences in maximal heart rate, maximal workload, or duration of exercise until exhaustion when the control, placebo, and naloxone trials were compared. As expected, a rise of body temperature of about 0.5°C was seen immediately after exercise in the control and placebo tests: mean sublingual temperature was 36.85 (SD 0.24) $^{\circ}\text{C}$ before and 37.33 (SD 0.26) $^{\circ}\text{C}$ after exercise in the control test and 36.64 (SD 0.28) $^{\circ}\text{C}$ before and 37.16 (SD 0.27) $^{\circ}\text{C}$ after exercise in the placebo test. Administration of naloxone completely abolished this rise in temperature: before exercise the sublingual

temperature in the naloxone test was 36.58 (SD 0.29) $^{\circ}\text{C}$ and after 36.40 (SD 0.54) $^{\circ}\text{C}$. The difference between the placebo and control tests was significant ($p < 0.001$) (table).

Discussion

Our finding that the rise in body temperature induced by exercise is antagonised by naloxone suggests that endogenous opiates play a part in thermal regulation during muscular exercise.

References

- 1 Yehuda S, Kastin A. Peptides and thermoregulation. *Neuroscience and Bio-behavioral Reviews* 1979;4:459-71.
- 2 Holaday JW, Loh HH, Li CH. Unique behavioural effects of beta endorphin and their relationship to thermoregulation and hypothalamic function. *Life Sci* 1978;22:1525-35.
- 3 Holaday JW, Wei E, Loh HH, Li CH. Endorphins may function in heat adaptation. *Proc Natl Acad Sci USA* 1978;75:2923-7.
- 4 Clark WG. Changes in body temperature after administration of amino acids, peptides, dopamine, neuroleptics, and related agents. *Neurosci Biobehav Rev* 1980;3:179-231.

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Dihydrocodeine in renal failure: further evidence for an important role of the kidney in the handling of opioid drugs

J N BARNES, A J WILLIAMS, M J F TOMSON, P A TOSELAND, F J GOODWIN

Abstract

The pharmacokinetics of a single oral dose of dihydrocodeine were studied in nine patients with chronic renal failure treated by haemodialysis and nine subjects with normal renal function. In the patients the mean peak plasma dihydrocodeine concentration occurred later and the area under the curve was greater than in the normal subjects. Furthermore, the drug was still detectable after 24 hours in all the patients but only three of the normal subjects.

These data, together with those obtained from previously published clinical case reports, contradict the traditional view that the body's ability to cope with opioid drugs is not altered in renal failure.

Introduction

The kidney is the main site for the elimination of many drugs and their metabolites from the body, and renal disease can consequently have important effects on the pharmacokinetics of such drugs. In addition, the pharmacokinetics of these drugs may be altered in uraemia by changes in plasma protein binding and the rates at which they are metabolised.^{1,2}

Although opioid drugs and their metabolites are excreted by the kidney^{3,4} and some have decreased plasma protein binding in uraemia,⁵ it is generally considered to be safe to prescribe them at the normal therapeutic dosage to patients with impaired renal function.⁶⁻⁸

This view must now be challenged. There have been several reports of serious narcosis in patients with renal failure treated with opioid drug,^{9,10} and evidence that the kidney has an important role in the elimination of opioid narcotics is accumulating.^{11,12}

The present study was performed to investigate the effect of end stage renal failure on the pharmacokinetics of a single oral dose of dihydrocodeine, a drug that has hitherto been considered to be safe at the conventional dosage in patients with chronic renal failure receiving maintenance haemodialysis.¹³

Subjects and methods

We studied nine subjects (five men), mean (SD) age 34.2 (4.2) years, with normal renal function and nine patients (seven men), age 40.8 (5.2) years, receiving maintenance haemodialysis. All the subjects attended after an overnight fast, and the patients attended on days when they were not receiving dialysis. A 19 G Butterfly cannula was inserted into a forearm vein in the normal subjects or into a vein on the back of the hand in the patients. Blood samples were taken immediately and without tourniquet for estimating plasma

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