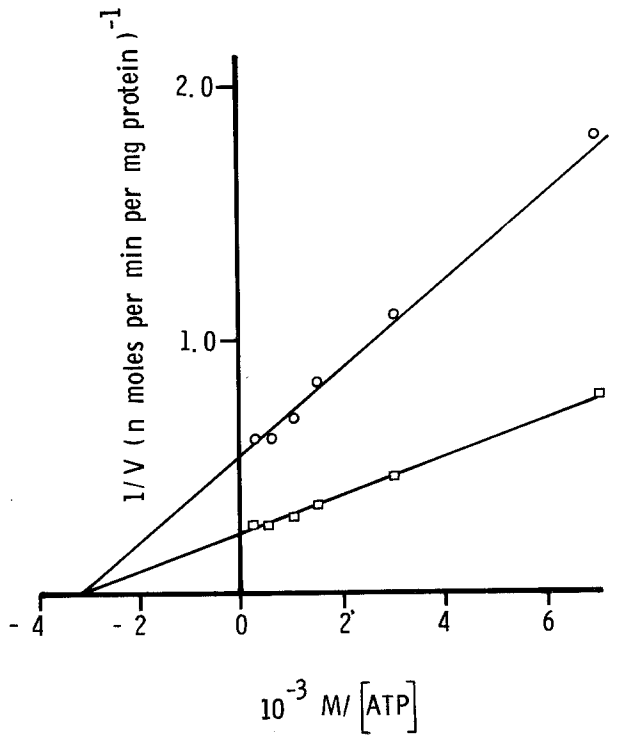


Fig. 3. Lineweaver-Burk plot for adenylate cyclase activity. cAMP synthesis as a function of ATP with (□) and wihtout (○) 6.0×10^{-3} *M* NaF. Caffeine concentration 5×10^{-2} *M*; protein concentration, 1.0 mg/ml. Each point represents a single reading.

preparations particularly where undergraduate teaching, in vitro pharmaceutical trials or kinetic studies are involved. Excess IO_4^- – consuming materials (sucrose or glycerol), excess anions forming insoluble lead salts ($> 2 \times 10^{-3}$ *M*) and cGMP interfere. Over broad concentration ranges many chemical effecters: catecholamines, F^-, Mg^{++}, Ca^{++}, Zn^{++}, Mn^{++}, Co^{++}, Fe^{++}, purines, pyrimidenes and xanthines do not affect the assay. Sulphydryl inhibitors which can interfere in other assays[12,13] were without effect.

Loss of linearity with high protein concentrations is likely due to excess diesterase and ATPase activities under these conditions. Caffeine at 5×10^{-2} *M* is optimal in minimizing interference by the former (Figure 2). Substantially lower caffeine concentrations were employed in earlier brain cyclase studies[3,8,14]. Similar to adipose adenylate cyclase[15], brain cyclase displays a V_{max} increase and no *Km* effect by the addition of NaF to the reaction medium (Figure 3)[16].

Zusammenfassung. Es wird eine direkte einfache chemische Methode zur Bestimmung von Gehirn Adenylatcyclase beschrieben, die auf der Neigung nichtzyklischer Nukleotiden zur Per-Jod-Oxydation im Gegensatz zur zyklischen AMP basiert. Empfindlichkeit: 10–100 nmol zyklischer AMP.

I. A. MACDONALD[17]

Department of Chemistry, University of Ottawa, Ottawa 2 (Ontario, Canada), 2 May 1974.

[12] W. B. WASTILA, J. T. STULL, S. E. MAYER and D. A. WALSH, J. biol. Chem. *246*, 1996 (1971).
[13] A. G. GILAMN, Proc. natn. Acad. Sci., USA *67*, 305 (1970).
[14] L. S. BRADHAM, D. A. HOLT and M. SIMS, Biochim. biophys. Acta *201*, 250 (1970).
[15] H. BAR and O. HECHTER, Analyt. Biochem. *29*, 476 (1969).
[16] I gratefully acknowledge Professor B. BELLEAU, Dept. of Chemistry McGill University for his help in developing this assay.
[17] Address: Dalhousie Clinical Research Centre, 5849 University Avenue, Halifax (Nova Scotia, Canada).

Surface Area and Effect of Drying Temperature Related to Dry Weight of Jejunal Tissue in Rat

Various workers have used different techniques to estimate the dry weight of jejunal tissue. Often, however, as is the convention in short-circuit work, ion movements are expressed per unit surface area. In the course of other experiments, the effect of drying above or below the boiling point of water was investigated and also the relationship of gut dry weight to wet surface area. Since results in the literature are often expressed per tissue weight incorporating different drying techniques, it was felt that a brief investigation of this and the elucidation of the regression of surface area on estimated dry weight might facilitate comparisons and transformations and be of general interest in transport studies.

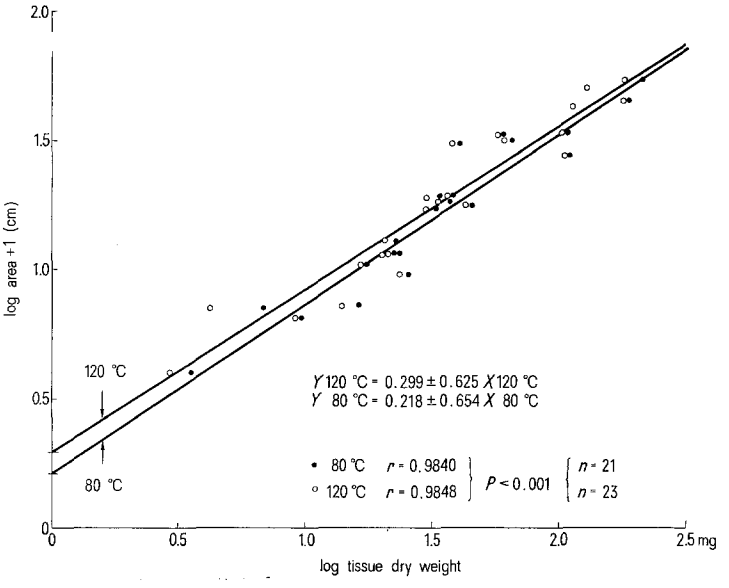
Methods. The proximal jejunum was removed from narcotized rats of about 200–250 g body weight and a section cut from sac I[1], 5 cm distal to the duodenal-

[1] B. A. BARRY, J. MATTHEWS and D. H. SMYTH, J. Physiol., Lond. *157*, 279 (1961).

Tissue dry weight as percentage wet weight under various drying conditions

	Dry weight (%)	Significance (p)
a) 120 °C for 4 h (not incubated)	17.45 ± 0.83 (16)	a) against b)
b) 80 °C for 4 h (not incubated)	19.35 ± 0.66 (16)	< 0.001
c) Freeze-dried (not incubated)	18.62 ± 0.22 (4)	c) against d)
d) Freeze-dried (incubated)	15.15 ± 0.40 (4)	< 0.001
e) 80 °C for 24 h (incubated)	17.25 ± 0.40 (12)	e) against f)
f) 80 °C for 48 h (incubated)	15.65 ± 0.41 (12)	< 0.02

Percentage given with standard error of mean and the number of animals in brackets.



Regression of log surface area on log tissue dry weight.

jejunal flexure. Tissue of varying surface area was consistently taken from this region, the surface area measured to the nearest millimetre and the flat section laid out on a microscope slide. Sections of jejunal tissue were dried in an oven for 4 h at 80 °C, weighed and then dired for a further 4 h at 120 °C with subsequent weighing. From these simple measurements, the dry weight percentage of the total wet weight was calculated at the 2 temperatures and from the assembled data, the regression of surface area on final dry weight examined. This data was compared with similar data for dry weight percentage of total wet weight from specimens dried to constant weight over a long period of time and with specimens that had been freeze-dried.

Results and discussion. The regression of surface area on tissue dry weight at both temperatures is best expressed by a double logarithmic relationship: –

1. $\log \text{area (in cm)}_{120\,^\circ C} + 1 = (299.0 \pm 4.6) \times 10^{-3} + (624.9 \pm 2.1) \times 10^{-3} \log \text{dry wt. (mg)}_{120\,^\circ C}$

2. $\log \text{area (cm)}_{(80\,^\circ C)} + 1 = (218.5 \pm 4.9) \times 10^{-3} + (653.8 \pm 2.6) \times 10^{-3} \log \text{dry wt. (mg)}_{(80\,^\circ C)}$

The reason for this logarithmic relationship (see Figure) is not immediately clear since rats were of the same age approximately and sacs sections were taken from the same region of the proximal jejunum. If a gradient of tissue dry weight were to exist increasing distally (which is unlikely since tissue wet weight and wall thickness decrease distally[2,3]), the regression seen might reflect the fact that larger samples would incorporate increasing amounts of tissue, so that per unit surface area, weight would increase with increasing surface area taken. Irrespective of the reason for this logarithmic relationship, surface area can be rapidly estimated accurately for sacs over the weight range shown. Estimations are best for small sacs of up to 100 mg dry weight (approximately 8 cm long) and are independent of drying temperature since the slopes of the regression lines do not significantly differ.

From the dry weight percentages (see Table) it can be seen that a significant difference does exist between drying at the 2 temperatures for a short period of time. The mean difference was 2.65 ± 0.29 mg ($n = 21$) (mean ± standard error of mean) which was significant ($p < 0.001$). This difference increased with increasing initial wet weight of tissue taken (difference = 1.87 + 0.0034 × tissue initial wet weight, $r = 0.5328$, $p < 0.02$). The dry weight percentage obtained after drying at 120 °C agrees with other estimates in the literature[4] for drying before without incubation. Tissue that had been freeze-dried with and without incubation gave similar percentages to those seen in the literature[4,5] and showed a similar loss of tissue dry weight on incubation. Both freeze-drying overnight and drying at 80 °C for not less than 48 h proved to be acceptable alternative procedures that led to standard values for drying above 100 °C for short periods of time.

Zusammenfassung. Es werden Gleichungen angeführt, die eine Abschätzung der Oberfläche des Rattenjejunums aus Trockengewichtsbestimmungen erlauben. Sollte sich eine Trocknung des Gewebes bei Temperaturen über 100 °C als unmöglich erweisen, ist anzuraten, eine Trocknung bei 80 °C für nicht weniger als 48 h oder Gefriertrocknung über Nacht auszuführen; hierbei werden gleiche Resultate erzielt.

M. L. LUCAS[6] and I. T. JOHNSON

Department of Physiology, Ludwig-Maximillian-University, D-8 München (German Federal Republic, BRD), 10 May 1974.

[2] T. H. WILSON and G. WISEMAN, J. Physiol., Lond. *123*, 126 (1954).
[3] H. NEWEY, P. A. SANFORD and D. H. SMYTH, J. Physiol., Lond. *208*, 705 (1970).
[4] F. DICKENS and H. WEIL-MALHERBE, Biochem. J. *35*, 7 (1941).
[5] R. D. BAKER and M. J. GEORGE, Biochim. biophys. Acta *225*, 315 (1971).
[6] Present address: Biological Sciences Department, Aston University, Birmingham 4 (England).