injection and dissection being 24 h. All synganglia, after removal of the sheath forming the periganglionic sinus and neurilemma, were smeared very thinly on slides to obtain isolated neurones well separated from surrounding tissue. Preparations were air dried on slides, exposed for 1 h to formaldehyde vapour generated from paraformaldehyde equilibrated to 65±2% rel. humidity and coverslip mounted in paraffin oil. The large postero-dorsal neurones were clearly distinguishable by size from other groups of smaller neurones[5].

Excitation spectra were obtained using a microspectrofluorometer which was similar to other instruments[9] based on the Leitz MPV and equipped with an incident light illuminator and cooled photomultiplier. The spectral half band width was 8.5 nm for the excitation and emission monochromators. Glass optics were adequate since the definitive wavelength shifts occurred above 360 nm. After the initial recording of the spectral curve each preparation was exposed successively to HCl vapour in a Coplin jar for 2, 20 and 200 sec and spectra re-recorded precisely at the same location on each slide. The high voltage to the photomultiplier was kept constant for each preparation[7].

Isolated fluorescent neurones (figure 1) from synganglion smears untreated with catecholamines had emission spectra which were characteristic of either DA or NE fluorophores produced in protein or tissue models; the emission maximum was 480 nm which clearly excluded the visually distinguishable 5-hydroxy tryptamine which has an emission peak of 530 nm. However, after HCl treatment, there was a clear differentiation between DA and NE in protein and tissue models as the DA excitation maximum (408 nm) shifted to a lower wavelength (394 nm) where it remained while the NE maximum after a transitory shift to 395–400 nm moved back to 408 nm. In tissue models in which isolated cells were examined, this move by the NE maximum back to the higher wavelength was accompanied by an increase in intensity after 200 sec exposure. Neurones exposed to HCl vapour in a similar manner to the models had excitation spectra which closely resembled those of NE-treated protein and tissue models; the latter spectra provide the more valid basis of comparison and are therefore included for comparison (figure 2).

These observations demonstrate that NE is present in the larger postero-dorsal neurones of the cortex[5] and is almost certainly the principal catecholamine[10]. Both NE and DA have been extracted from synganglion tissue of *B. microplus*[2,3] and either or both occur in varicose fibres of the neuropile[5]. However, measurement of fluorescence intensity in the small terminals and isolation of these terminals and varicose fibres from surrounding tissue must await even more sensitive techniques.

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10 Formaldehyde vapour-treated protein models incorporating a 1:1 mixture of DA and NE had excitation spectra closely resembling that of DA so that DA, if present in the isolated neurones, would almost certainly be much lower in concentration than NE.

Seasonal changes in the electrical parameters of the small intestine, colon and bladder mucosa of land tortoises (*Testudo hermanni hermanni* Gmelin)[1]

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Summary. During hibernation of tortoises, a marked decrease in the short-circuit current together with an increase in the electrical resistance are observed across the small intestine, colon and bladder mucosa and D-aldosterone (which is decreased in the plasma) has no effect on these parameters.

It has been pointed out previously that permeability characteristics to inorganic ions of small intestine and colon epithelia are altered during hibernation[2]. Changes in the inorganic ion content of the small intestine, colon and bladder mucosa, as well as in muscle, were also detected[3]. The inexchangeable fraction of intracellular Na and K has also been shown to vary during the winter in the small intestine and colon[4] as well as in the bladder[5]. These changes should be reflected in the electrical parameters of these epithelia.

Each epithelium is cleared of muscular and connective layers by dissection, and set in between plexiglas chambers

Table 1. Potential difference (PD), short-circuit current (SCC) and resistance (R) across small intestine, colon and bladder mucosa of active and torpid tortoises

	PD (mV) Active	Torpid	SCC (μA/cm²) Active	Torpid	R (Ω × cm²) Active	Torpid
Small intestine	1.5± 0.9 (11)	2.6±0.9 (9) S	12.8± 3.3 (9)	4.5± 2.9 (8) HS	117	578
Colon	29.7±13.3 (22)	22.3±11.9 (10) NS	58.6±31.3 (11)	16.0±8.2 (13) HS	507	1394
Bladder	64.8±27.0 (14)	15.3± 4.4 (18) HS	205.3±82.9 (6)	25.2±19.8 (18) HS	316	607

Results are means±SD. Number of individuals between brackets. The probability to the equivalence of the means is given as follows (t-test of Student): S, the change is significant (level 5%); HS, highly significant (level 1%); NS, not significant.

as previously described[6]. Electrical potential difference (PD) is measured using a DC Differential Voltmeter (John Fluke MFG Co., Inc., Model 881A), connected to both sides of the epithelium through a pair of calomel electrodes and agar bridges. The short-circuit current (SCC) is measured using a device which is similar to the one described by Ussing and Zerahn[7]. The electrical resistance (R) is calculated by applying the Ohm law.

As shown in table 1 in all 3 isolated epithelia, the short-circuit current is much lower in torpid tortoises than in active ones. The potential difference increases during hibernation at the level of the small intestine but remains very low. In the colon no significant change of PD is observed. In contrast in the bladder there is an important drop in the values of PD recorded during the winter. In all 3 epithelia, these changes of PD and/or SCC bring about an increase of the transepithelial resistance.

This increase of the resistance is probably related to decreased passive permeabilities. Indeed the serosal border of small intestine and colon epithelia has been shown to be less permeable to inorganic ions during hibernation[2]. As to the decreased SCC values recorded during hibernation, one can assume it reflects a lower activity of the Na pump. This Na pump should be controlled by aldosterone. In a closely related species of land tortoise (*Testudo graeca*), it was shown that in in vitro experiments D-aldosterone was effective at the level of the bladder and perhaps of the colon, an increased SCC being recorded in active animals[8].

By radioimmuno-assay the aldosterone plasma level was studied in active and torpid tortoises (table 2). Despite tremendous individual variations, the aldosterone level is significantly lower in the torpid animals. But when the influence of aldosterone on the SCC of the 3 epithelia is tested, no effect is observed even after 7 h of incubation. Therefore it is tempting to conclude that lower SCC values recorded during hibernation are not the consequence of the lower aldosterone plasma level measured.

In the bladder, it is interesting to note that the water net flux is much lower in torpid animals than in active ones, and that it is controlled by antidiuretic hormones in torpid animals and not in active ones[9]. As to the SCC, it is also much lower in torpid animals, but it is controlled by D-aldosterone only in active animals and not in torpid ones.

Table 2. Aldosterone present in the blood plasma of active and torpid tortoises

	Activity	Hibernation
Aldosterone (pg/ml)	156.1 ± 198.8 (18) S	57.9 ± 55.8 (33)

Results are expressed as means ± SD. S, the change is significant at the 5% level.

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The sequential uptake of (^{14}C) deoxyglucose in brain after embolic stroke

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Summary. An i.v. bolus of (^{14}C) deoxyglucose was injected into 4 groups of rats which simultaneously were embolized through the internal carotid artery. 15 and 30 min post embolization there was a massive decrease in (^{14}C) deoxyglucose uptake in both deep and cortical structures. At 4 h a small zone of increased (^{14}C) deoxyglucose uptake persisted around each embolized microsphere.

Recent experiments have demonstrated that the technique of (^{14}C) 2-deoxyglucose (2-DG) autoradiography permits a pictorial and semi-quantitative representation of the relative rates of glucose uptake and/or utilization in functionally active neurons[1]. The technique is based upon the assumption that 2-DG is transported across neuronal cell membranes by the same carrier system utilized by glucose[2]. Once across the neuronal membrane it is phosphorylated to 2-DG-6-P but is not metabolized. The amount of glucose utilized by a particular neuronal population correlates with its activity and can be calculated from that population's 2-DG concentration, if: 1. adequate time has elapsed for

(^{14}C) 2-deoxyglucose in the embolized rat brain

	Embolized hemisphere				Non-embolized hemisphere			
	Cortex		Basal ganglia-thalamus		Cortex		Basal ganglia-thalamus	
	Hot	Cold	Hot	Cold	Hot	Cold	Hot	Cold
A (15′)	0.34 ± 0.02	0.04 ± 0.01	0.32 ± 0.02	0.13 ± 0.01	0.30 ± 0.02	0.10 ± 0.01	0.18 ± 0.01	0.11 ± 0.01
B (30′)	0.38 ± 0.02	0.13 ± 0.01	0.27 ± 0.02	0.05 ± 0.01	0.20 ± 0.01	0.18 ± 0.02	0.12 ± 0.01	0.09 ± 0.01
C (30′)	0.29 ± 0.01	0.09 ± 0.02	0.28 ± 0.03	0.07 ± 0.02	0.25 ± 0.03	0.18 ± 0.03	0.16 ± 0.02	0.08 ± 0.02
D (60′)	0.20 ± 0.02	0.05 ± 0.01	0.18 ± 0.02	0.06 ± 0.01	0.18 ± 0.02	0.16 ± 0.02	0.16 ± 0.01	0.05 ± 0.01
E (240′)	0.18 ± 0.02	0.09 ± 0.01	0.15 ± 0.01	0.05 ± 0.02	0.16 ± 0.01	0.04 ± 0.01	0.09 ± 0.01	0.03 ± 0.01

2 DG utilization in the embolized and non-embolized hemisphere. Hot areas represent areas of increased glucose uptake whereas cold areas represent decreased uptake. The values recorded are the average of 5 OD readings taken from each representative area of increased or decreased glucose consumption in the embolized and non-embolized hemisphere. 95% confidence intervals were calculated for these values using the normal distribution.