NMR) with *A*; and both could be oxidized by air in aqueous solution around neutrality with color changes as observed with extracts of *Mercurialis perennis*.

Compound *A* was oxidized by nitric acid to 4-methoxy-1-methylpyridine-2,3,6-(1*H*)-trione (**3**), m.p. 132 °C; $M^+$169, $C_7H_7NO_4$ requires 169; $\delta$ ($CDCl_3$) 3.35 (3H, s, N–Me), 3.9 (3H, s, O–Me), 6.25 (1H, s, –CH=); $\delta$ ($CF_3CO_2H$), 3.45 (3H, s, N–Me), 4.0 (3H, s, O–Me), 6.5 (1H, s, –CH=). When equimolecular amounts of this and *A* were mixed at pH 7.1 an immediate deep blue color was produced. Compounds *A* and (**3**) can be regarded as analogous to a quinol and a quinone, respectively, and the blue compound might then be a semiquinone-like radical-anion (**4**). ESR spectroscopic evidence consistent with the latter structure is presented in the following communication by A.R. Forrester[6].

When a solution of *A* in methanol was warmed in the presence of air rapid oxidation occurred with the formation of a very sparingly soluble, colorless, crystalline compound (*B*), 5,5′-dihydroxy-4,4′-dimethoxy-1,1′-dimethyl[3,3′-bipyridine]-2,6,2′,6′(1*H*,3*H*,1′*H*,3′*H*)-tetrone (**5**), which on acetylation afforded a crystalline product formulated as either 5,6,5′,6′-tetra-acetoxy-4,4′-dimethoxy-1,1′-dimethyl [3,3′-bipyridine]-2,2′-dione (**8**) or 2,5,2′,5′-tetraacetoxy-4,4′dimethoxy-1,1′-dimethyl[3,3′-bipyridine]-6,6′-dione, and which had m.p. 174–175 °C (decomp.); $M^+$508.1313, $C_{22}H_{24}N_2O_{12}$ requires 508.1329; $\delta$ ($CDCl_3$) 2.15 (6H, s, 2OCO$CH_3$), 2.3 (6H, s, 2OCO$CH_3$), 3.35 (6H, s, 2 N–Me), 3.8 (6H, s, 2OMe). In $CF_3CO_2H$ *B* disproportionates to give *A* and (**3**). The symmetrical homolysis of *B* on heating was demonstrated by trapping the resulting radical with 2,2′-azobis(2-methylpropionitrile) to yield 3-[2′-(2′-cyanopropyl)]-5-hydroxy-4-methoxy-1-methylpyridine-2,6-(1*H*,3*H*)-dione (**9**), m.p. 152 °C, $M^+$238.0953, $C_{11}H_{14}N_2O_4$ requires 238.0953.

When a solution of either *A* or *B* in pyridine was exposed to air it afforded a yellow compound 4,4′-dimethoxy-1,1′-dimethyl[3,3′-bipyridine]-2,2′,-5,5′,6,6′(1*H*,1′*H*)-hexone (**7**); *m/e* 336, *321*; $\delta$ ($CDCl_3$) 3.35 (N–Me) and 4.15 (O–Me) equal in area.

When *B* was oxidized with ceric ammonium nitrate in acetic acid it yielded an orange-yellow product, apparently a quinhydrone-like compound formed from equimolecular amounts of (**6**) and (**7**), *m/e* 338, 336, *321*, 169. From n.m.r. spectra it appeared that in either $CDCl_3$ or in $CF_3CO_2H$ this decomposed to give a mixture of 1 mol of (**7**) and 2 mol of (**3**). Extraction with chloroform of an aqueous extract of *Mercurialis perennis* after it had been oxidized by air afforded the same material; $M^+$338.0746, $C_{14}H_{14}N_2O_8$ requires 338.0750; $M^+$336.0604, $C_{14}H_{12}N_2O_8$ requires 336.0594.

Potentiometric titration of *A* at pH 4 gave a curve with only 1 step after the addition of 2 equivalents of $K_3Fe(CN)_6$ per mol of *A*, corresponding to oxidation to (**3**). However at pH 7.6 the first step [oxidation to (**3**)] occurred after the addition of rather less than 2 equivalents, but was followed by a second step circa 0.5 equivalent later, representing the oxidation of *B*, formed by reaction of *A* with (**3**). The curve obtained by titrating the *Mercurialis perennis* extract at pH 7.6 resembled the latter curve[5].

To summarize, an extract of *Mercurialis perennis* prepared as described by Haas and Hill contains *A* (hermidin), which on oxidation by air or $K_3Fe(CN)_6$ gives first a radical-anion (cyanohermidin) (**4**), then (**3**) which by reaction with *A* and further oxidation gives a complex of compounds (**6**) and (**7**) (chrysohermidin).

Compound *A* was also obtained by homogenizing the fresh plant in cold sodium acetate-sodium dithionite solution for 1 min, followed immediately by extraction with chloroform.

1 Acknowledgment. I thank Professor R.H. Thomson for his interest.
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## Autoxidation of hermidin: an ESR study

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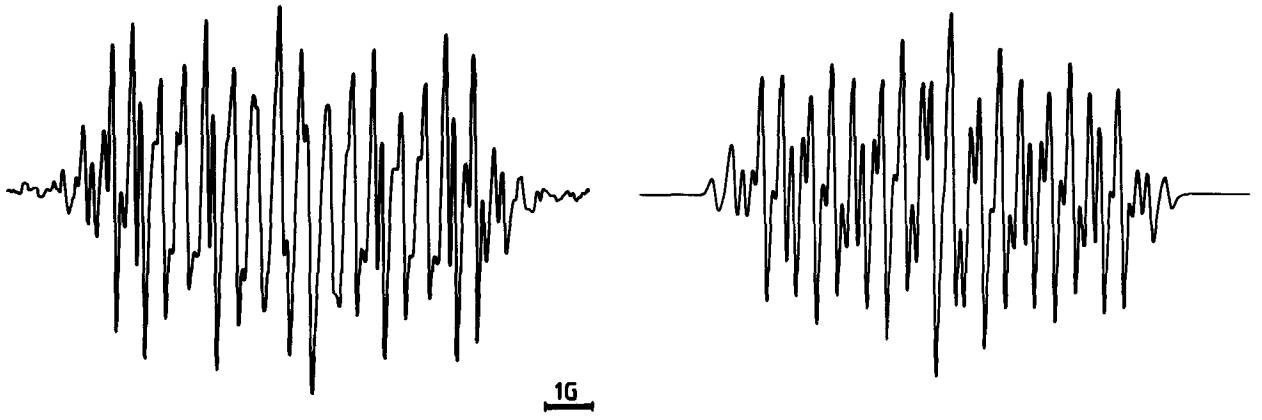
*Summary.* Aqueous solutions of hermidin readily give rise to a blue transient radical-anion on exposure to air, the identity of which has been established by ESR spectroscopy.

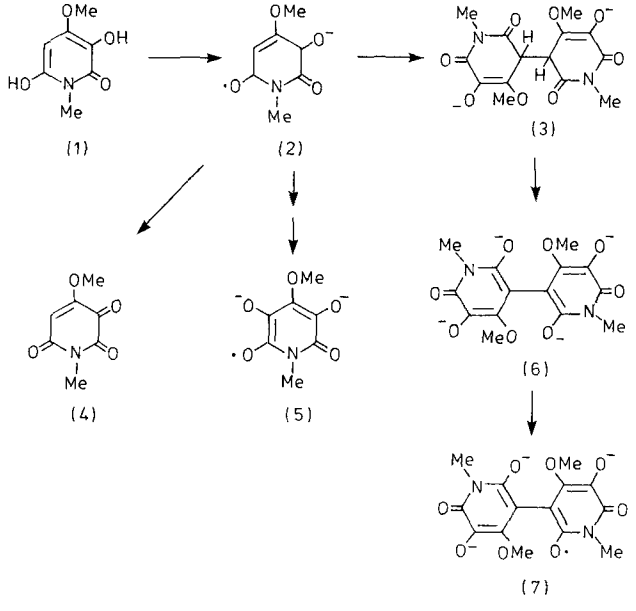
The preceding paper by Swan[2] describes the chemical changes which occur when the colorless chromogen hermidin (**1**), isolated from the plant *Mercurialis perennis* L. is oxidized. Aqueous solutions of hermidin (**1**) on autoxidation rapidly turn blue, then green and finally yellow-brown which suggests that radicals may be implicated. This has now been confirmed by ESR spectroscopy, the results providing supporting evidence for the previously proposed reaction scheme[2].

Hermidin (**1**) dissolved slowly in degassed aqueous buffer (pH 7.17) to give a pale blue solution which in an ESR cavity gave a complex and unsymmetrical spectrum. Over a period of a few hours the spectrum almost disappeared and was replaced by a secondary spectrum composed simply of a triplet of quarters [$a_N$ = 1.5 and $a_H$ = 0.5 G(3H)]. Lack of symmetry in the original spectrum made analysis difficult but repetition of the experiment using deuterated solvent ($D_2O$) provided a spectrum which was symmetrical and lasting. Analysis gave $a_H$ = 0.4(3H), $a_H$ = 0.6(3H), $a_N$ = 1.5 and $a_D$ = 0.5 G. Simulation using $a_H$ = 3.45 ($a_H = a_D \times 6.5$), $a_H$ = 0.6(3H), $a_H$ = 0.4(3H) and $a_N$ = 1.45 G produced a spectrum which matched well these parts of the original spectrum which were symmetrical. Hence this spectrum is attributed to the radical-anion (**2**) which exchanges H-5 for D-5 in deuterated solvent. Confirmation of this assignment was achieved by measuring a solution containing approximately equal amounts of hermidin (**1**) and the trione (**4**). This blue solution gave an unperturbed spectrum which was a good overall match for the simulated spectrum (fig.).

The secondary spectrum (triplet of quartets) had relatively broad lines (0.4 G) and did not show a large splitting due to a ring proton. Therefore, the radical giving rise to this spectrum must be substituted at C-5. The two most likely possibilities are (**5**) and (**7**) the latter arising from dimerization of (**2**) and subsequent oxidation and the former by hydroxylation of her-




Actual (left) and simulated (right) ESR spectra of radical-anion from hermidin.



midin (**1**) for which there is analogy[3]. Of the two, (**5**) seems the more likely since (**7**) would have been expected to show splittings from 2 nitrogens, 2 methoxyls, etc.[3] Further, when the water insoluble dimer (**3**) (written as the dianion) was dissolved in methanol-aqueous buffer the resulting solution did not give an ESR spectrum until alkali was added. Then, a weak spectrum of the radical-anion (**2**) was detected. This presumably arises from partial dissociation[2] of the dimer (**3**). The transient blue color produced on exposure of solutions of hermidin (**1**) (and possibly of bruised or cut stems of the plant from which it is derived) to air is due therefore to the formation of the radical-anion (**2**) "cyanohermidin".

1 Acknowledgment. I thank Professor R. H. Thomson for helpful discussion.

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## Viscero-somatic reflexes following distension of urinary bladder in cats: Role of supraspinal neuraxis[1]

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*Summary.* Viscero-somatic reflexes have been studied by recording monosynaptic reflexes following distension of the urinary bladder in intact, decerebrate and spinal animals. It was observed that the viscero-somatic responses following bladder distension are inhibitory in nature and this inhibition was highest in decerebrates and least in spinal animals. The site of viscero-somatic interaction probably lies in the bulbar area (supraspinal) and spinal cord.

It is well known that dynamic behavior of the somatic muscle is altered with the distension of hollow viscera like the intestine, urinary bladder, uterine horn etc. Downman and McSwiney[2] showed that pinching or gently squeezing the intestine, head of the pancreas or uterine horn in spinal and decerebrate cats, produced movement of hind limbs. Evans and McPherson[3,4] have also reported the effects of bladder distension on the monosynaptic reflexes in different experimental conditions and preparations of the animals. The aim of the present investigation is to study the spinal and supraspinal control on the viscero-somatic reflexes following distension of the bladder in cats.

*Materials and methods.* The experiments were carried out in cats of either sex, weighing 2–3 kg. Animals were anesthetized with sodium pentobarbitone (Nembutal, Abott) at a dose of 30 mg/kg b.wt i.p. and maintained with a dose of 10 mg/kg b.wt i.v. The urethra was exposed through a midline suprapubic incision. The bladder was cannulated with a polythene catheter via the urethra in order to change bladder volume and monitor intravesicular pressure. The catheter was connected to a