Inotropic Activities of Imidazopyridines

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A series of 2-substituted 1H-imidazo[4,5-b]pyridines and the isomeric 1H-imidazo[4,5-c]pyridine derivatives has been prepared and evaluated as inotropic agents. The 1H-imidazo-[4,5-b] derivatives were found to be consistently more potent than their isomers in the [4,5-c] series in isolated guinea pig papillary muscle preparations. Structure-activity relationships and the species-dependence of inotropic potencies are discussed.

Inotrope Aktivität von Imidazopyridinen

Es wurde eine Reihe von 2-substituierten 1H-Imidazo[4,5-b]-pyridinen und der isomeren 1H-Imidazo[4,5-c]pyridin-Derivate hergestellt und als inotrope Agentien evaluiert. Die 1H-Imidazo-[4,5-b]-Derivate erweisen sich als deutlich wirksamer als ihre Isomere in der [4,5-c] Reihe am isolierten Papillarmuskel des Meerschweinchens. Es werden die Verhältnisse zwischen Struktur und Aktivität sowie die Artenabhängigkeit der inotropen Wirksamkeit diskutiert.

The pioneering work of the K. Thomae group led to the development of the inotropic drug sulmazole **1**[1]. Clinical evaluation of sulmazole has been terminated, however, because of undesirable toxicological effects and substantial metabolism[2]. Potent inotropic agents which lack the drawbacks associated with sulmazole or digoxin are of much current interest for the treatment of congestive heart failure[3]. Our initial approach to obtaining such an agent was to synthesise and determine the pharmacological profile of BWA746C ** **2**[4]. This 1H-imidazo[4,5-c]pyridine derivative was also discovered independently by workers at E. Lilly and E. Merck and was found to be a more potent inotropic agent than sulmazole itself[5,6].

As part of an exercise to develop some understanding of the structure-activity relationships for sulmazole we have undertaken a comparative study of the inotropic activity of 50 'C' ring modified 1H-imidazo[4,5-b]- and 1H-imidazo[4,5-c]pyridines.

The isolated guinea pig papillary muscle preparation was employed to assay the *in vitro* inotropism of these sulmazole analogues.

We now report the synthesis and pharmacological evaluation of a representative set of 28 of these analogues.

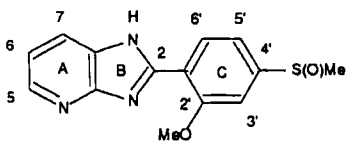
Chemistry

The imidazopyridines **3-15, 19-24, 27**, and **28** were readily prepared by condensation of either 2,3- or 3,4-diaminopyridine with the appropriate aromatic acid. Carboxamide **17** was obtained by hydrolysis of nitrile **15** using conc. H_2SO_4 at room temp. The 2-pyrazinyl analogue **29** was synthesised by base-catalysed condensation of 2,3-diaminopyridine with 2-cyanopyrazine. Synthesis of analogues **16, 18, 25, 26,** and **30** are described elsewhere as indicated in the experimental section. The reaction pathways are shown in the scheme.

Structure-Activity Relationships

The *in vitro* inotropic activities of the sulmazole analogues are given in Table 1. This set of 1H-imidazo[4,5-b]pyridines consistently displays more potent *in vitro* inotropism than the isomeric 1H-imidazo[4,5-c]pyridine derivatives. This finding, observed in guinea pig papillary muscle preparations, is in marked contrast to the results reported[5] by the E. Lilly group. These workers employed cat papillary muscle preparations to determine the *in vitro* inotropic potencies of a smaller set of 'C' ring modified 1H-imidazo[4,5-b]- and 1H-imidazo[4,5-c]pyridines. They observed that the 1H-imidazo[4,5-c]pyridine series was invariably more potent than the [4,5-b] series, e.g. **2>1, 4>3, 6>5**. The differences in the ranking order of potencies for a number of imidazopyridines in the [4,5-b] and [4,5-c] series appear to result from the different species employed.

Four other salient points emerged from the present study. Firstly, significant deviations from planarity lead to a marked reduction of activity. Thus the 2',6'-dimethoxy



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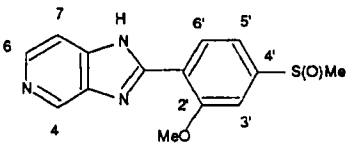

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** Also known as isomazole and subsequently referred to by this name.

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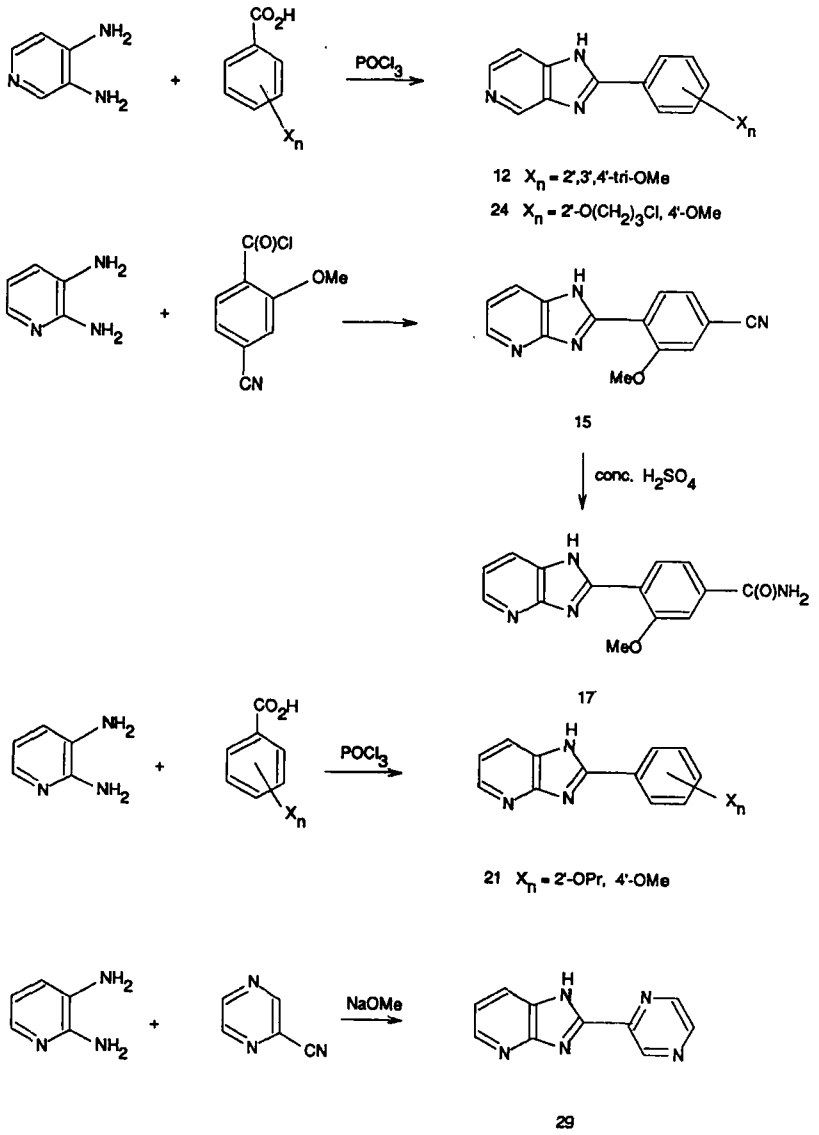
Scheme



Ar

(3)	Ph	(4)
(5)	2',4'-di-OMe.C_6H_4	(6)
(7)	2',6'-di-OMe.C_6H_4	(8)
(9)	3',4'-di-OMe.C_6H_4	(10)
(11)	2',3',4'-tri-OMe.C_6H_3	(12)
(13)	2'-OMe, 4'-Cl.C_6H_4	(14)
(15)	2'-OMe, 4'-CN.C_6H_4	(16)
(17)	2'-OMe, 4'-C(O)NH_2.C_6H_4	(18)
(19)	2'-OMe, 4'-$OCH_2C{\equiv}CH$.C_6H_4	(20)
(21)	2'-OPr, 4'-OMe.C_6H_4	(22)
(23)	2'-O$(CH_2)_3$Cl, 4'-OMe.C_6H_4	(24)
(25)	2'-O$(CH_2)_2$S(O)Me, 4'-OMe.C_6H_4	(26)
(27)	2-naphthyl	(28)
(29)	2-pyrazinyl	(30)

analogues **7** and **8** have been shown[6] by u.v. spectral and X-ray crystallographic methods to have an inter-planar angle between the phenyl and heterocyclic rings of ca. 40°. Analogue **7** displays only weak inotropic activity, and isomer **8** is inactive.

Secondly, the introduction of 3'-substituents usually gives rise to analogues with poorer activity. This was the case for **11** and **12** as can be seen by comparing their activities with those of **5** and **6**. One notable exception to this generalisation is provided by 3',4'-dimethoxy substitution. An analogue with this substitution pattern, the 1H-imidazo[4,5-c]pyridine **10**, is more potent than either the 2',4'-dimethoxy derivative **6** or the [4,5-b] isomer **9**.

Thirdly, the 4'-electron withdrawing substituents such as S(O)Me, $CONH_2$ or Cl tend to minimise the differences in potencies between the two series. Thus sulmazole and isomazole are approximately equipotent and the difference in $pA_{50\%}$ values between analogues **13** and **14** is only 0.3 and that for **17** and **18** only 0.2. Conversely, 4'-electron releasing substituents maintain or augment potency differences

Table 1: *In vitro* inotropic Activities of Isomazole Analogues

Compound	$pA_{50\%}$ [a]	Compound	$pA_{50\%}$
(1)	4.70±0.10(9)	(2)	4.64±0.15(17)
(3)	4.00±0.1	(4)	3.03±0.14
(5)	5.08±0.09(4)	(6)	4.01±0.09(9)
(7)	2.93±0.03	(8)	inactive
(9)	3.87±0.03	(10)	4.90±0.15
(11)	3.67±0.03	(12)	inactive
(13)	4.30±0.21	(14)	4.04±0.14(5)
(15)	insoluble	(16)	<6 [b]
(17)	4.26±0.06	(18)	4.43±0.08
(19)	5.49±0.04	(20)	3.91±0.35
(21)	6.00±0.06	(22)	inactive
(23)	6.10±0.02	(24)	inactive
(25)	5.80±0.1	(26)	2.70±0.15
(27)	4.60±0.15	(28)	inactive
(29)	3.00±0.06	(30)	2.73±0.15

a) Parameter of *in vitro* inotropic potency. Potent inotropic agents at low concentrations produce large increases in the force of contraction of isolated paced guinea-pig papillary muscles in organ bath experiments. $pA_{50\%}$ is the negative logarithm of the drug concentration required to give a 50% increase in basal contractile force for (n) experiments where n=3 unless otherwise stated. Compound **21**, for example, produces a 50% increase in the force of contraction of guinea-pig papillary muscles in such experiments at concentrations of 10^{-6} M. Its inotropic potency, $pA_{50\%}$, is therefore 6. Compounds which did not produce a 50% increase in the force of contraction were classified as inactive.

b) variable biphasic dose-response curve observed; accurate determination not possible.

Table 2: *In vivo* Activities of Isomazole Analogues

Compound	ED_{50}[a] (mg kg-1)
(1)	0.80
(2)	0.06
(5)	0.91
(6)	0.12
(10)	>10.0
(16)	0.014
(18)	0.014
(19)	0.28
(21)	2.3
(22)	>10.0

a) Parameter of *in vivo* inotropic potency. Potent inotropic agents produce a large increase in dP/dt when given as a single bolus (i.v.) injection to anaesthetised open chest dogs at low doses. P is left ventricular pressure and dP/dt is the first time differential of P. The ED_{50} value is the dose required to produce a maximum increase of 50% above the basal value of dP/dt.

between the two series, e.g. Δ $pA_{50\%}$ between **5** and **6** is 1.0. Δ $pA_{50\%}$ between **3** and **4** is 1.0.

Finally the inotropic activities of the 1H-imidazo[4,5-c]pyridines appear to be more sensitive to increased 'C' ring steric bulk than those of their [4,5-b] counterparts. Analogues **19, 21, 23, 25,** and **27**, for example, are far more potent than their respective isomers **20, 22, 24, 26,** and **28**. Particularly striking are the effects of bulky 2'-substituents. Replacement of the 2'-methoxy group by a 2'-propoxy or substitued propoxy group gives rise to increased potencies for the [4,5-b] analogues but renders inactive or weakly active the [4,5-c] isomers.

In conclusion it is worth noting that, in contrast to the guinea pig *in vitro* results, 1H-imidazo[4,5-c]pyridine analogues were generally observed to have more potent inotropic effects *in vivo* than their [4,5-b] analogues (Table 2). Relative potencies obtained in experiments using anaesthetised dogs are in agreement with those reported[5]. The index of potency employed to describe the inotropism in these experiments was the ED_{50}, the effective dose of drug required to increase ventricular dP/dt by 50% over its basal value. [dP/dt is the first time derivative of left ventricular pressure (P).] Typical ED_{50} values are those for hetero cycles **1** (0.8 mg kg^{-1}), **2** (0.06 mg kg^{-1}), **5** (0.91 mg kg^{-1}), and **6** (0.12 mg kg^{-1}). Thus it may be that for several classes of sulmazole analogues, *in vitro* assays employing cat, rather than guinea pig, papillary muscles give a better indication of relative *in vivo* potencies in dogs.

Other observations worth noting in regard to the *in vivo* studies are that (a) 2',4'-dimethoxy substitution is preferred to 3',4'-dimethoxy substitution in the [4,5-c] series, cf. ED_{50} values for **6** (0.12 mg kg^{-1}) and **10** (> 10 mg kg^{-1}); (b) replacement of a 4'-OMe substituent by a 4'-$OCH_2C{\equiv}CH$ group led to increased potency in the [4,5-b] series, cf. ED_{50} values for **19** (0.28 mg kg^{-1}) and **5** (0.91 mg kg^{-1}); (c) the most potent analogues possessed electron-withdrawing substituents in the 4'-position, i.e. ED_{50} values for **16** and **18** (0.014 mg kg^{-1} in both cases); (d) compound **21**, although one of the most potent inotropes *in vitro* ($pA_{50\%}$ 6.00), displayed relatively weak *in vivo* effects (ED_{50} value 2.3 mg kg^{-1}).

Detailed *in vivo* evaluations of several of these isomazole analogues are described in the following communication.

Experimental Part

Melting points: Koffler hot-stage instrument, uncorrected. - ^{1}H-NMR-spectra: Bruker HFX 90 (90 MHz), Bruker AM-200 (200 MHz) or WM-360 (360 MHz), TMS as internal standard. - Mass spectra: Kratos MS-25, 70 eV. - Org. extracts were dried over $MgSO_4$.

The syntheses of the following analogues are reported: 2-phenyl-1H-imidazo[4,5-b]pyridine (**3**)[7], 2-phenyl-1H-imidazo[4,5-c]pyridine (**4**)[8], 2-(2,4-dimethoxyphenyl)-1H-imidazo[4,5-b]pyridine (**5**)[7], 2-(2,4-dimethoxyphenyl)-1H-imidazo[4,5-c]pyridine (**6**)[5,9], 2-(2,6-dimethoxyphenyl)-1H-imidazo[4,5-b]pyridine (**7**)[10], [2-(2,6-dimethoxyphenyl)-1H-imidazo[4,5-c]pyridine (**8**)[5,9], 2-(3,4-dimethoxyphenyl)-1H-imidazo[4,5-b]pyridine (**9**)[5], 2-(3,4-dimethoxyphenyl)-1H-imidazo[4,5-c]pyridine (**10**)[5], 2-(2,3,4-trimethoxyphenyl)-1H-imidazo[4,5-b]pyridine (**11**)[7], 2-(4-chloro-2-methoxyphenyl)-1H-imidazo[4,5-b]pyridine (**13**)[7], 2-(4-chloro-2-methoxyphenyl)-1H-imidazo[4,5-c]pyridine (**14**)[5], 2-(4-cyano-2-methoxyphenyl)-1H-imidazo[4,5-c]pyridine (**16**)[11], 2-(4-carbamoyl-2-methoxyphenyl)-1H-imidazo[4,5-c]pyridine (**18**)[11], 2-[2-methoxy-4-(2-propynyl)oxyphenyl]-1H-imidazo[4,5-b]pyridine (**19**)[12], 2-[2-methoxy-4-(2-propynyloxy)phenyl]-1*H*-imidazo[4,5-c]pyridine (**20**)[12], 2-(4-methoxy-2-propoxyphenyl)-1H-imidazo[4,5-c]pyridine (**22**)[5], 2-[2-(3-chloropropyloxy)-4-

methoxyphenyl]-1H-imidazo[4,5-b]pyridine (**23**)[7], 2-[4-methoxy-2-(methylsulphinyl)ethoxyphenyl]-1H-imidazo[4,5-b]pyridine (**25**)[13], 2-[4-methoxy-2-(methylsulphinyl)-ethoxyphenyl]-1H-imidazo[4,5-c]pyridine (**26**)[5], 2-(2-naphthyl)-1H-imidazo[4,5-b]pyridine (**27**)[2], 2-(2-naphthyl)-1H-imidazo[4,5-c]pyridine (**28**)[2], 2-(2-pyrazinyl)-1H-imidazo[4,5-c]pyridine (**30**)[11].

2-(2,3,4-Trimethoxyphenyl)-1H-imidazo[4,5-c]pyridine dihydrochloride (**12**)

A finely powdered mixture of 3,4-diaminopyridine (2.2 g, 0.02 mol) and 2,3,4-trimethoxybenzoic acid (4.2 g, 0.02 mol) was added in portions to $POCl_3$ (40 ml) with stirring. This mixture was heated at reflux for 4 h with stirring and the excess $POCl_3$ removed *in vacuo*. Ice-water (100 ml) was added to the residue followed by aqueous ammonia until pH 11. The mixture was extracted with chloroform (3x), the combined extracts were dried and evaporated to dryness. The residual solid was dissolved in warm ethanol (100 ml), excess conc. HCl added, and the volatile material removed *in vacuo*. The crude product was recrystallised from ethanol - water (95:5) to give **12** as off-white crystals (2.9 g, 41%); m.p. 250-252°C (dec.). - $C_{15}H_{15}N_3O_3 \cdot 2HCl$ (358.2) Calcd. C 50.3 H 4.78 Cl 19.8 N 11.7 Found C 49.9 H 4.73 Cl 19.8 N 11.7. - ^{1}H-NMR (90 MHz, dmso-d_6): δ (ppm) = 3.81 (3H, s, OMe), 3.89 (3H, s, OMe), 4.04 (3H, s, OMe), 7.01 (1H, d, J = 8.8 Hz, H-5'), 8.03 (1H, d, J = 8.8 Hz, H-6'), 8.18 and 8.52 (2H, AB system, J = 6.4 Hz, H-6, H-7), 9.30 (1H, s, H-4), 12.3 (2H, v.br.s, 2xNH, exchangeable).

2-(4-Cyano-2-methoxyphenyl)-1H-imidazo[4,5-b]pyridine hydrochloride (**15**)

A solution of 4-cyano-2-methoxybenzoyl chloride[14]. (1.78 g, 9.0 mmol) in dry toluene (15 ml) was added over 20 min to a stirred mixture of 2,3-diaminopyridine (0.99 g, 9.0 mmol), triethylamine (9 ml) and dry pyridine (27 ml). The mixture was stirred at room temp. for 60 h, poured onto ice, and the solid removed by filtration. After washing with water and drying at 65°C, 1.80 g (75%) of 2-amino-3-(4-cyano-2-methoxybenzoyl)-aminopyridine, m.p. 180-182°C, m/z 268 (M^+), was obtained as a tan solid.

This amide (1.70 g), ethanediol (20 ml) and a few drops of conc. HCl were heated at 195°C for 1.5 h. The cooled mixture was poured onto ice-water containing a little Na_2CO_3 and the resulting solid collected by filtration. This material, after drying, was purified by column chromatography on silica. Elution with CH_2Cl_2-MeOH (99:1) gave a pale yellow solid. Treatment with excess saturated HCl-MeOH gave 0.52 g (29%) of **15** as pale yellow crystals, m.p. 260-265°C. $C_{14}H_{10}N_4O \cdot 0.95HCl$ (284.8) Calcd. C 59.0 H 3.85 Cl 11.9 N 19.7 Found C 59.4 H 3.89 Cl 12.2 N 19.7. - ^{1}H-NMR (200 MHz, dmso-d_6): δ (ppm) 3.90 (2H, v.br. peak, 2x NH, exchangeable), 4.12 (3H, s, OMe), 7.54 (1H, m, H-6), 7.63 (1H, dd, J = 8 Hz, 1 Hz, H-5'), 7.82 (1H, d, J = 1 Hz, H-3'), 8.35 (1H, br. d, J = 8 Hz, H-7), 8.48 (1H, d, J = 8 Hz, H-6'), 8.57 (1H, d, J = 5 Hz, H-5).

2-(4-Carbamoyl-2-methoxyphenyl)-1H-imidazo[4,5-b]pyridine hydrochloride (**17**)

Nitrile **15** (0.50 g) was added to conc. H_2SO_4 (8 ml) and stirred for 1 h at room temp. The resulting solution was allowed to stand at room temperature for 48 h and then poured onto ice. The mixture was neutralised by addition of 10 M NaOH, with cooling. The resulting solid was collected, washed with water and dried. Treatment with excess methanolic HCl and crystallisation from methanol-ether gave 0.35 g (55%) of **17**, m.p. 327-330°C. $C_{14}H_{12}N_4O_2 \cdot 1.1HCl$. 0.75 H_2O (321.7) Calcd. C 52.2 H 4.53 Cl 12.1 N 17.4 Found C 52.1 H 4.51 Cl 12.8 N 17.3. - ^{1}H-NMR (200 MHz, dmso-d_6): δ (ppm) 4.18 (3H, s, OMe), 4.0-5.0 (2H, v.br.peak, NH_2, exchangeable), 7.55-7.72 (3H, m, H-6, H-5', NH, 1H exchangeable), 7.80 (1H, br.s, H-3'), 8.27 (1H, br.s, NH, exchangeable), 8.42 (1H, d, J = 8 Hz, H-6'), 8.53 (1H, d, J = 8 Hz, H-7), 8.62 (1H, d, J = 5 Hz, H-5).

2-(4-Methoxy-2-propyloxyphenyl)-1H-imidazo[4,5-b]pyridine hydrochloride (**21**)

2,3-Diaminopyridine, 4-methoxy-2-propyloxybenzoic acid[5] and $POCl_3$ gave, as described for **12**, a 32% yield of **21**, m.p. 195-197°C. $C_{16}H_{17}N_3O_2 \cdot HCl$ (319.5) Calcd. C 60.1 H 5.63 N 13.2 Found C 60.3 H 5.71 N 13.1.

2-[2-(3-Chloropropoxy)-4-methoxy]phenyl-1H-imidazo[4,5-c]pyridine dihydrochloride (**24**)

3,4-Diaminopyridine, 2-(3-chloropropoxy)-4-methoxybenzoic acid[7] and $POCl_3$ gave, as described for **12**, a 38% yield of **24**, m.p. 187-188°C (dec.). $C_{16}H_{16}ClN_3O_2 \cdot 2HCl \cdot H_2O$ (408.8) Calcd. C 47.1 H 4.90 N 10.3 Found C 47.4 H 4.90 N 10.1. MS: m/z 317 (M^+, free base).

2-(2-Pyrazinyl)-1H-imidazo[4,5-b]pyridine dihydrochloride (**29**)

To a freshly prepared solution of sodium (230 mg, 0.01 mol), in dry methanol (30 ml) was added 2-cyanopyrazine[15] (1.05 g, 0.01 mol). The resulting solution was stirred at room temp. for 5 h and the methanol evaporated to give the crude imidate as an oil. This oil, 2-methoxyethanol (30 ml) and 2,3-diaminopyridine·HCl (1.45 g, 0.01 mol) were stirred and heated at reflux for 6 h. The cooled mixture was evaporated to dryness, water was added and the aqueous phase extracted with $CHCl_3$ (3x). The org. extracts were dried, the solvent was removed *in vacuo*, and the residue triturated with ether. The resulting solid was collected and treated with methanolic HCl to yield 0.29 g (10%) of **29**, m.p. 247-250°C. $C_{10}H_7N_5 \cdot 2HCl \cdot 0.75H_2O$ (283.5) Calcd. C 42.3 H 3.70 N 24.7 Found C 42.3 H 3.76 N 24.8. - ^{1}H-NMR (200 MHz, dmso-d_6): δ (ppm) 5.8-6.2 (3H, v.br. peak, 3x NH, exchangeable), 7.61 (1H, dd, J = 8.1, 5.1 Hz, H-6), 8.41 (1H, dd, J = 8.1, 1.4 Hz, H-7), 8.64 (1H, dd, J = 5.1, 1.4 Hz, H-5), 8.91 (2H, m, NCHCHN), 9.59 (1H, br.s, CHN).

Pharmacological Methods

Paced Guinea-pig Papillary Muscles

Male guinea-pigs (Halls 275-325 g) allowed free access to food and water were killed by a blow to the head. The heart was rapidly excised and washed in *Krebs-Henseleit* solution containing 2.5 mM Ca^{2+}, gassed with 95% O_2: 5% CO_2 at 30°C. The heart was transferred to a Petri dish containing the same buffer kept at approximately 34°C throughout the dissection. Fresh buffer was used for each dissection and washings were discarded after use.

A single right ventricular papillary muscle was employed from each heart, the tendinous end ligated to a stainless steel hook and the lower end ligated and cut away from the ventricle wall and attached to a Perspex clamp such that the tissue was in contact with a platinum punctate electrode. The stainless steel hook was suspended from a Grass FT.03 transducer which recorded isometric tension. The preparation was placed in a 20 ml Pyrex organ bath containing buffer gassed with 95% O_2: 5% CO_2 and maintained at 34°C. 500 mg loading tension was applied to the preparation. Stimulation was effected by rectangular pulses of 1 msec duration at 1.5 Hz at 20% above the threshold voltage (1-5 volts) by a SRI stimulator. The transducer inputs were coupled to a potentiometric recording device by a 6-channel Grass transducer coupler. A group of organ baths enabled up to six preparations to be utilised during one experimental run. After 60 min, preparations unable to sustain uniform contractions beyond this point were rejected. The total volume of sample solutions added generally amounted to less than 400 µl (2% change in total bath volume). Compound additions

were made to the baths in a cumulative fashion in multiples of the initial dose (1, 3, 10, 30-100 etc.), the individual responses being allowed to attain a plateau before the next addition was made. The inotropic potency is expressed as the negative logarithm of the concentration required to increase basal contractility by 50% ($pA_{50\%}$).

In vivo experiments utilising anaesthetised dogs

Details of the methods used in these experiments, and terminology relating to them, are as described[16].

Beagle dogs (of either sex) weighing between 8.5-13 kg were initially anaesthetised by an *i.v.* injection of thiopentone sodium (30 mg kg^{-1}) into a cephalic vein. Anaesthesia was subsequently maintained by intravenous injection of α-chloralose (15 mg kg^{-1}) and pentobarbitone sodium (6 mg kg^{-1}) through a cannula placed in the right femoral vein. The trachea was then cannulated and the animal artificially ventilated with room air using a Palmer pump (stroke volume 200-250 ml and respiration rate 20 min^{-1}). Arterial blood samples were removed before beginning the experiment and analysed (Radiometer Blood Gas Analyser) to ensure that the pump ventilation maintained blood gases within acceptable limits. The chest was opened along the length of the sternum and the pericardium opened to expose the heart. The root of the ascending aorta was located and cleared of fat and an electromagnetic flow probe (10-12 mm internal diameter) placed around the ascending aorta and connected to a Statham flowmeter to measure aortic blood flow. Extra corporeal pressure transducers were used to measure blood and left ventricular pressure. Left ventricular pressure (P) and its first derivative, dP/dt, were measured by the insertion of a short cannula (containing heparinised-saline and connected to a Statham pressure transducer) into the left ventricular chamber via the apex of the heart. This cannula was secured in place by a purse-string suture.

Arterial blood pressure was measured by means of a catheter (containing heparinised-saline and connected to a Statham pressure transducer) inserted into the right femoral artery and a Lead II electrocardiogram was obtained by use of subdermal needle electrodes. Heart Rate was derived by use of a tachograph triggered either by the arterial pulse or the ECG-QRS complex. Body temp. was maintained at 37-38°C by a heated under blanket. All recordings were made by use of a Grass Model 7D Polygraph, or a Gould 2800S Recorder. In some animals the stability of this preparation was assessed by administering no drugs, and minitoring the measured cardiovascular parameters for up to 300 min. In all animals the reponse to a bolus injection of vehicle only was observed.

References

1 B. Unterhalt, Drugs Future, *6*, 421 (1981).
2 D.W. Robertson, J.H. Krushinski, E.E. Beedle, V. Wyss, G. Don. Pollock, and J.S. Hayes, Eur. J. Med. Chem. *21*, 223 (1986) and references therein.
3 P.W. Erhardt, J. Med. Chem. *30*, 231 (1987).
4 D.W. Robertson and J.S. Hayes, Drugs Future *10*, 295 (1985).
5 D.W. Robertson, E.E. Beedle, J.H. Krushinski, G.Don. Pollock, H. Wilson, V.L. Wyss, and J.S. Hayes, J. Med. Chem. *28*, 717 (1985).
6 P. Barraclough, D. Firmin, R. Iyer, W.R. King, J.C. Lindon, M.S. Nobbs, S. Smith, C.J. Wharton, and J.M. Williams, J. Chem. Soc. Perkin Trans. 2, 1839 (1988).
7 E. Kutter and V. Austel, Ger. Patent 2,305,339; C.A. *82*, 4251y (1975).
8 R.W. Middleton and D.G. Wibberley, J. Heterocycl. Chem *17*, 1757 (1980).
9 P. Barraclough, R.M. Beams, J.W. Black, D. Cambridge, D. Collard, D.A. Demaine, D. Firmin, V.P. Gerskowitch, R.C. Glen, H. Giles, A.P. Hill, R.A.D. Hull, R. Iyer, W.R. King, D.J. Livingstone, M.S. Nobbs, P. Randall, G.P. Shah, S.J. Vine, and M.V. Whiting ,Eur. J. Med. Chem., in press (1990).
10 E. Kutter, V. Austel and W. Diederen, Ger. Patent 2,361,757; C.A. *83*, 206270k (1975).
11 P. Barraclough, J.W. Black, D. Cambridge, D.A. Demaine, V.P. Gerskowitch, H. Giles, A.P. Hill, R.A.D. Hull, R.Iyer, W.R. King, D.J. Livingstone, M.S. Nobbs, P. Randall, G.P. Shah, and M.V. Whiting, following communication.
12 R. Jonas, K. Minck, H.J. Enenkel, and H.J. Schliep, Eur. Patent 72,926; C.A. *99*, 139940u (1983).
13 D.W. Robertson and J.H. Krushinski, Org. Prep. Proc. Int. *20*, 311 (1988).
14 P. Barraclough, R. Iyer, J.C. Lindon, and J.M. Williams, Tetrahedron Lett. *27*, 5997 (1986).
15 H. Foks and J. Sawlewicz, Acta. Pol. Pharm., *25*,137 (1968); C.A. *69*, 77229p (1968).
16 G. Allan, D. Cambridge, M.J. Follenfant, D. Stone, and M.V. Whiting, Br.J. Pharmacol. *93*, 387 (1988).

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