# Cardiotonic Agents. 1. Novel 8-Aryl-Substituted Imidazo[1,2-*a*]- and -[1,5-*a*]pyridines and Imidazo[1,5-*a*]pyridinones as Potential Positive Inotropic Agents

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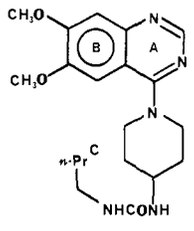
Several 8-arylimidazo[1,2-*a*]pyridines, 8-arylimidazo[1,5-*a*]pyridines, and 8-arylimidazo[1,5-*a*]pyridinones were prepared and tested in vitro for potential cardiac inotropic and electrophysiological activity. Selected analogues were further tested in vivo in canine hemodynamic and cardiac electrophysiology models. Compounds having an imidazole substituent consistently showed activity. A pharmacophoric relationship between heterocycle–phenyl–imidazole and positive inotropic activity was noted. The significance of this relationship is discussed.

During exploration of synthetic methodology that could provide novel heterocycles,[1] and as part of a program intended to produce cardiotonic agents,[2] we became interested in certain imidazo[1,2-*a*]- and -[1,5-*a*]pyridines. We felt that these heterocycles, as shown in Table I, bear some resemblance (regions A, B, C) to buquineran (1)[3] and papaverine (2),[4] which are known to inhibit cAMP phosphodiesterase (PDE). Compound 1 has been studied



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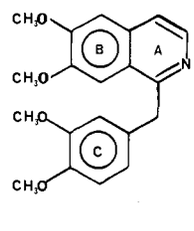

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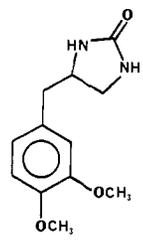

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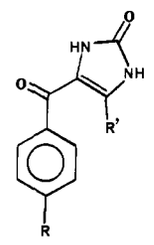
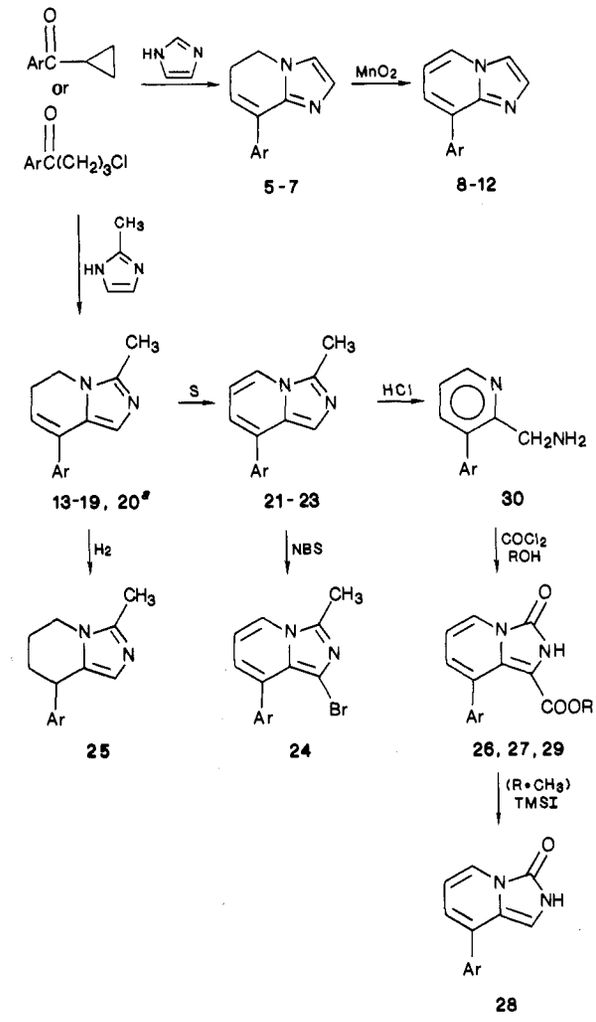


4a R=R'=H
b R=SCH$_3$, R'=CH$_3$

clinically as a positive inotropic agent although its electrophysiological effects complicate its use as a cardiotonic.[5] As our synthetic studies progressed, we also obtained the related imidazo[1,5-*a*]pyridinones shown in Table II. These heterocycles closely resemble the positive inotropic agent Ro7-2956 (3)[6] and the related aroylimidazolones 4.[7] Derivative **4b**, an inhibitor of cAMP PDE, recently has become of clinical interest as a potential cardiotonic.[8] Finally, simple imidazole[9] and benzimidazole[10] derivatives

**Scheme I**



[a] Ethylimidazole rather than methylimidazole.

have been reported to have positive inotropic activity, and their use as templates for the design of potential cardiotonic agents has been discussed previously.[8,11] We describe herein the synthesis and biological characterization of the

(1) Davey, D. *J. Org. Chem.*, in press.
(2) Erhardt, P. *J. Med. Chem.* **1987**, *30*, 231.
(3) Wetzel, B.; Hauel, N. *Annu. Rep. Med. Chem.* **1984**, *19*, 75.
(4) Preiniger in *The Alkaloids*; Manske, R., Ed.; Academic: New York, 1975; Vol. 15, p 209.
(5) Hillis, W. S.; Been, M. *Eur. Heart J.* **1982**, *3* (Suppl D), 97.
(6) Osborne, M. W.; Wenger, J. J.; Moe, R. A. *J. Pharmacol. Exp. Ther.* **1971**, *176*, 174.
(7) Duschinsky, R.; Dolan, L. *J. Am. Chem. Soc.* **1946**, *68*, 2350.
(8) Schnettler, R.; Dage, R.; Grisar, J. *J. Med. Chem.* **1982**, *25*, 1477.
(9) (a) Knope, R.; Moe, G. K.; Saunders, J.; Tuttle, R. *J. Pharmacol. Exp. Ther.* **1973**, *185*, 29. (b) Tuttle, R. S.; Garcia-Minor, C.; Simon, M. *J. Pharmacol. Exp. Ther.* **1975**, *194*, 624.
(10) Diederen, W.; Kadatz, R. *Arzneim.-Forsch.* **1981**, *31*, 141.
(11) Kutter, E.; Austel, V. *Arzneim.-Forsch.* **1981**, *31*, 135.
(12) Our results from testing several standard inotropic agents and cAMP phosphodiesterase inhibitors in cat, guinea pig, and ferret papillary tissues reveal that the ferret, in general, represents the most sensitive tissue for determining inotropic effects. Detailed results of our species comparison will be published elsewhere.

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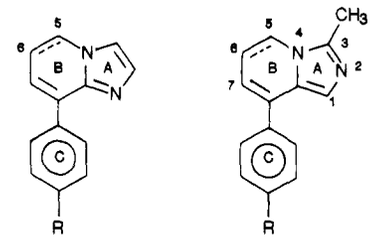
**Table I.** Imidazo[1,2-*a*]- and -[1,5-*a*]pyridines



5-12 13-25

| no. | 5,6[a] | R | yield, % | mp, °C | formula[b] | inotropic activity:[c] CF $C_{20}$, μM | electrophysiology:[d] FRP $C_{20}$, μM |
|---|---|---|---|---|---|---|---|
| **5** | s | H | 79 | 65–67 | $C_{13}H_{12}N_2$ | NR (−15) | NR (8) |
| **6** | s | $NH_2$ | 21 | 139–141 | $C_{13}H_{13}N_3$ | NR (−5) | NR (10) |
| **7** | s | Im[e] | 43 | 139–141 | $C_{16}H_{14}N_4$ | 29 | 100 |
| **8** | u | H | 78 | 69–70 | $C_{13}H_{10}N_2$ | 1[f] | NR (10) |
| **9** | u | OH | 17 | 199–201 | $C_{13}H_{10}N_2O$ | NR (15) | NR (−18) |
| **10** | u | $NH_2$ | 63 | 136–138 | $C_{13}H_{11}N_3$ | NR (15) | NR (10) |
| **11** | u | Im | 75 | 136–137 | $C_{16}H_{12}N_4$ | 30 | NR (18) |
| **12** | u | $NHSO_2CH_3$ | 51 | 237–239 | $C_{14}H_{13}N_3O_2S$ | 10 | 50 |
| **13** | s | H | 41 | 83–84 | $C_{14}H_{14}N_2$ | NR (10) | NR (5) |
| **14** | s | $OC_2H_5$ | 45 | 137–138 | $C_{16}H_{18}N_2O$ | 20[f] | 30 |
| **15** | s | OH | 36 | 270–272 | $C_{14}H_{14}N_2O$ | 30 | 100 |
| **16** | s | Im | 57 | 195–197 | $C_{17}H_{16}N_4$ | 9 | 14 |
| **17** | s | Me-Im[g] | 44 | 221–223 | $C_{18}H_{18}N_4$ | 5 | 5 |
| **18** | s | $NHCOCH_3$ | 13 | 258–260 | $C_{16}H_{17}N_3O$ | 10 | 40 |
| **19** | s | $NHSO_2CH_3$ | 17 | 235–237 | $C_{15}H_{17}N_3O_2S$ | 2 | 10 |
| **20**[h] | s | Et-Im[i] | 28 | 130–131 | $C_{20}H_{22}N_4$ | 0.8 | 9 |
| **21** | u | H | 69 | 70–71 | $C_{14}H_{12}N_2$ | −20 (F) | NR (18) |
| **22** | u | $OC_2H_5$ | 21 | 97–98 | $C_{16}H_{16}N_2O$ | −100 | NR (15) |
| **23** | u | Me-Im | 58 | 194–195 | $C_{18}H_{16}N_4$ | 13 | 80 |
| **24**[j] | u | Me-Im | 89 | 177–178 | $C_{18}H_{15}BrN_4$ | 1 (F) | 17 |
| **25**[k] | s | $NHSO_2CH_3$ | 92 | 206–207 | $C_{15}H_{19}N_3O_2S$ | 0.8 (F) | 80 |
| 1[l] | - | - | - | - | - | 7 (F) | NR (3)[m] |

[a] s indicates that the double bond is saturated and u indicates that a double bond is present. [b] For all compounds, C, H, and N analyses agree within ±0.4% of theory and NMR spectral data support the indicated structures. [c] Drug concentration (μM) causing a 20% positive or negative (−) change in contractile force (CF $C_{20}$) of papillary muscle from cat or ferret (F).[12] In general, data represent the mean from at least two determinations where the ranges are within ±10 μM. NR indicates that a 20% change was not reached at 100 μM (for these cases the percent change at 100 μM is reported in parentheses). [d] Drug concentration (μM) causing a 20% increase in the functional refractory period (FRP $C_{20}$) of canine ventricular muscle. In general, data represent the mean from at least two determinations where the ranges are within ±20 μM. NR indicates that a 20% change was not reached at 100 μM (for these cases the percent change at 100 μM is reported in parentheses). [e] Im indicates an imidazol-1-yl substituent. [f] Despite the initial activity, increases in CF greater than 25–30% could not be achieved with increasing doses up to 100 μM. [g] Me-Im indicates a 2-methylimidazol-1-yl substituent. [h] The substituent at position 3 is ethyl rather than methyl. [i] Et-Im indicates a 2-ethylimidazol-1-yl substituent. [j] The structure also contains a bromine at position 1. [k] In this compound the 7,8 positions are also saturated. [l] Buquineran. [m] A $C_{10}$ could be obtained at ca. 8 μM.

cardiac properties associated with our novel heterocycles and a limited series of analogues.

## Chemistry

Both the 8-arylimidazo[1,2-*a*]pyridines and the 8-arylimidazo[1,5-*a*]pyridines are prepared by a novel process[1,13] that involves reacting a phenyl cyclopropyl ketone or a 4-halobutyrophenone with an excess of an imidazole at 175–225 °C (Scheme I). When imidazole itself is employed in this process, the unsubstituted 5,6-dihydro 1,2-*a* ring system is formed. Similarly, when a 2-alkylimidazole is used, the 3-alkyl 5,6-dihydro 1,5-*a* ring system is produced. For example, the imidazole compounds **7** and **17** were prepared in one step from 4-chloro-4′-fluorobutyrophenone with imidazole and 2-methylimidazole, respectively. Whereas the 5,6-dihydro 1,2-*a* compounds are readily dehydrogenated with activated $MnO_2$ in methylene chloride at room temperature, the 5,6-dihydro 1,5-*a* compounds required either sulfur in refluxing dichlorobenzene or palladium on carbon in nitrobenzene at 200 °C to give the fully unsaturated 1,5-*a* ring system. Treatment of the 8-arylimidazo[1,5-*a*]pyridines with 6 N HCl[14] provided a useful synthesis of the 2-(aminomethyl)-3-arylpyridines (e.g., **30**). The latter were converted[15] with either phosgene or trichloromethyl chloroformate followed by alcohols to the 8-aryl-2,3-dihydro-3-oxoimidazo[1,5-*a*]pyridine-1-carboxylic acid esters. Compound **27** proved to be resistant to hydrolysis by aqueous hydroxide, but was cleanly dealkylated and decarboxylated by iodotrimethylsilane in methylene chloride to give compound **28** in good yield. As shown, catalytic hydrogenation of **19** and bromination of **23** with NBS provided the fully saturated analogue **25** and the bromo-substituted analogue **24**, respectively.

## Biology

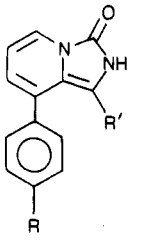
Compounds **5–25** were first tested in in vitro screens to determine their cardiac inotropic and electrophysiological properties. These data are listed in Table I. In the first series, **5–7**, which has a 5,6-dihydro 1,2-*a* ring system, para substitution on the phenyl ring with an imidazole (**7**) results in both positive inotropic activity and weak class III

(13) Davey, D. U.S. Patent 4 596 872, 1986; *Chem. Abstr.* **1986**, 133888*d*.

(14) Palazzo, G.; Picconi, G. *Farmaco, Ed. Sci.* **1975**, *30*, 3, 197.

(15) Dunham, D. Ph.D Dissertation, 1967, Ohio University, Athens, OH.

**Table II.** Imidazo[1,5-*a*]pyridin-3-ones



**26-29**

| no. | R | R′ | yield, % | mp, °C | formula[a] | inotropic activity:[b] CF $C_{20}$, μM | PDE inhibn:[c] $IC_{50}$, μM |
|---|---|---|---|---|---|---|---|
| **26** | H | $CO_2CH_2CH_3$ | 68[d] | 153–154 | $C_{16}H_{14}N_2O_3$ | ~10 | 74 |
| **27** | $CH_3$ | $CO_2CH_3$ | 72[d] | 197–198 | $C_{16}H_{14}N_2O_3$ | ~40 | 68 |
| **28** | $CH_3$ | H | 64 | 194–196 | $C_{14}H_{12}N_2O$ | 6 | 100 |
| **29** | Me-Im[e] | $CO_2CH_3$ | 36[f] | 228–230 | $C_{19}H_{16}N_4O_3$ | 30[g] | 73 |
| **1**[h] | - | - | - | - | - | 7[g] | 5 |
| **4b**[i] | - | - | - | - | - | 3 | 5 |

[a] For all compounds, C, H, and N analyses agree within ±0.4% of theory and NMR spectral data support the indicated structures. [b] Details are the same as for Table I footnote *c* except that all studies were done in ferret tissue. [c] Drug concentration (μM) causing 50% inhibition ($IC_{50}$) of canine cardiac cAMP phosphodiesterase. In general, data represent the mean from at least three determinations where the ranges are within ±10 μM. [d] Phosgene in toluene was used as reagent. [e] Me-Im indicates a 2-methylimidazol-1-yl substituent. [f] Trichloromethyl chloroformate was used as reagent. [g] Dose–response curves were biphasic with lower concentrations showing negative inotropy. [h] Buquineran. [i] Enoximone.

electrophysiological effects. The importance of the imidazole substituent[16] is clear since the parent ring system **5** is essentially inactive in these screens and because substitution with a simpler base (**6**) did not show significant activity. This pattern is repeated in series **8–12**, which has a fully unsaturated 1,2-*a* ring system. For this series, both an imidazole substituent (**11**) and substitution with a methanesulfonamide (**12**) resulted in significant activity. In series **13–20**, which has a 5,6-dihydro 1,5-*a* ring system, consistent potent inotropic and electrophysiological effects are again obtained by introduction of imidazole substituents (**16**, **17**, **20**) and in one case a methanesulfonamide substituent **19**. In the final series of compounds, **21–25**, which has a fully unsaturated 1,5-*a* ring system, significant activity is again found in the relationship heterocycle–phenyl–imidazole (**23**, **24**) and with a methanesulfonamide (**25**) substituent.

To determine the mechanism responsible for the inotropy exhibited by the imidazole and methanesulfonamide analogues, representative compounds were tested in the presence of 100 μM nadolol. While some attenuation of action was noted, in general the presence of nadolol did not significantly decrease the inotropy obtained from the imidazole and methanesulfonamide analogues. In contrast, the weak inotropic action elicited by the hydroxy analogue **15** was completely abolished in the presence of nadolol.[17] These studies suggest that while there may be some limited involvement of the β-adrenergic pathway, to a large extent the inotropy observed in vitro for the imidazole and methanesulfonamide analogues probably does not involve such mechanisms. Representatives were also tested for their ability to inhibit canine cardiac cAMP phosphodiesterase (partially purified fraction III PDE). In these tests the imidazole analogues were found to be weak ($IC_{50}$ for **7** was 550 μM) to moderate ($IC_{50}$ for **24** was 30 μM) inhibitors while the methanesulfonamide analogues were essentially inactive ($IC_{50}$ for **12** > 1000 μM). In addition, it was noted that the unsaturated systems (e.g., **11** $IC_{50}$ = 220 μM and **23** $IC_{50}$ = 65 μM) are more potent inhibitors than their corresponding 5,6-dihydro systems (e.g., **7**, and **17** $IC_{50}$ = 150 μM) and that substitution at position 1 of the 1,5-*a* ring system appears to enhance inhibitory potency (**24** compared to **23**).

To further clarify their electrophysiological activity, representatives were tested in canine Purkinje fibers where their ability to lengthen the action potential duration ($APD_{95}$) was determined. All of the representatives exhibited at least weak activity in this screen. Three compounds, **17**, **19**, and **20**, showed significant activity increasing the APD by 20% at 12, 2, and 10 μM concentrations, respectively.

Because they were potent in both front-line screens, compound **17** as an imidazole representative and compound **19** as a methanesulfonamide representative were chosen for in vivo characterization. In hemodynamic studies conducted in the anesthetized dog, infusion of **17** at 1 mg $kg^{-1}$ $min^{-1}$ consistently produced a 20% increase in $dp/dt$ (positive inotropic activity) with little effect on either heart rate or mean blood pressure. Higher infusion rates were accompanied by further increases in inotropy (up to a ca. 50%) and a modest reduction in heart rate (ca. 10–20%). However, the actions observed for **17** subsided within a few minutes after drug infusions were halted, suggesting that the biological half-life for **17** is very short. In addition, the inotropy observed for **17** in vivo was blocked by the administration of either nadolol or propanolol. This strongly suggests that, in contrast to its in vitro action, the in vivo inotropic activity observed for **17** largely results from interaction with the β-adrenergic system. In in vivo electrophysiology studies, compound **17** exhibited only limited activity on cardiac functional refractory period (increase of ca. 10%) with infusions up to 20 mg $kg^{-1}$ $min^{-1}$ and, again, the actions quickly diminished to control levels when drug infusions were terminated. When compound **19** was similarly studied in

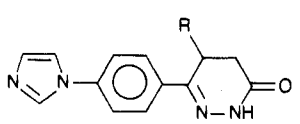
(16) We have also studied the inotropic properties of imidazole and its simple alkyl and aryl derivatives and found that such compounds exhibit comparatively weaker activity which does not appear to be associated with phosphodiesterase inhibition (e.g., imidazole is essentially inactive up to concentrations as high as 10 mM).

(17) This is not completely unexpected since the *p*-hydroxy compound bears some resemblance to β-adrenergic agents.

vivo, significant dose-related sinus arrhythmia was observed beginning at 3 mg/kg i.v. While some positive inotropy (ca. 20% increases in $dp/dt$) and bradycardic activity (ca. 10% reduction in HR) could be measured at lower doses, the effects were transient and their measurement became complicated by the arrhythmias that occurred as the dose was increased.

For the imidazopyridinone series **26–29** that more closely resembles known PDE inhibitors that do not have electrophysiological complications, front-line screening included measurement of cardiac cAMP PDE inhibition rather than in vitro assessment of electrophysiological properties. These data are listed in Table II. It is clear that while the parent nucleus, **28**, exhibits reasonably potent inotropic activity, it is a weak inhibitor of cAMP PDE. Substitution of the R′ position with ester functionality tends to increase inhibition of cAMP PDE. However, without an imidazole at R (**26**, **27**), this does not translate into an increase in positive inotropic action and, in fact, results in negative inotropy. Alternatively, for the imidazole case, and although biphasic in its overall dose–response relationship (lower dose being slightly negative), at least a modest positive inotropic action is obtained.

Taken together, the studies show that within the three structural series, the relationship heterocycle–phenyl–imidazole leads to compounds that exhibit positive inotropic activity accompanied by varying degrees of cAMP PDE inhibitory activity. For several compounds it appears that the inotropic activity derives from multiple mechanisms such as interaction with the β-adrenergic pathway as well as PDE inhibition. The importance of the imidazole substituent is particularly demonstrated by cases where the parent ring system is only weakly active in one or more of the screens. This relationship, where imidazole serves as a key substituent, is different from previous considerations[8,11] where the imidazole has been equated with the heterocyclic portion of structures such as **4b**. It should be noted that during the course of our studies, another group of investigators has independently revealed[18] a series of substituted pyridazinones, **31**, where the relationship




**31**

heterocycle–phenyl–imidazole also appears to be of critical importance for positive inotropic activity. In our next paper we further explore the potential generality of this relationship through examination of less rigid versions of the imidazole substituted imidazopyridinones.[19]

## Experimental Section

Melting points were taken on a Fisher-Johns melting point apparatus and are uncorrected. IR spectra were obtained on either a Sargent/Welch 3-300 spectrophotometer or a Beckman Aculab 2 spectrophotometer. $^1$H NMR spectra were recorded on a Varian XL-300 spectrometer. Elemental analyses were performed by the Berlex Analytical Department of by Galbraith Laboratories, Inc. Column chromatography was carried out on Merck silica gel 60, 230–400 mesh. Thin-layer chromatography (TLC) was performed with Merck silica gel 60 F254 plates.

**5,6-Dihydro-3-methyl-8-[4-(2-methyl-1*H*-imidazol-1-yl)phenyl]imidazo[1,5-*a*]pyridine (17). General Method for the Preparation of 8-Aryl-5,6-dihydroimidazopyridines.** 4-Chloro-4′-fluorobutyrophenone (50 g, 0.25 mol) and 2-methylimidazole (130 g, 1.58 mol) were combined and heated at 200 °C under nitrogen for 18 h. The mixture was cooled, dissolved in 1 L of $CH_2Cl_2$, and washed with water to remove the excess of 2-methylimidazole. The $CH_2Cl_2$ portion was dried over $MgSO_4$ and charcoal treated, and the solvent was removed under vacuum. The residue was crystallized with EtOAc to provide 51 g of crude product, which was recrystallized from MeCN to give 32 g of **17** as a light yellow solid.

**3-Methyl-8-[4-(2-methyl-1*H*-imidazol-1-yl)phenyl]imidazo[1,5-*a*]pyridine (23). General Method for the Dehydrogenation of 8-Aryl-5,6-dihydroimidazo[1,5-*a*]pyridines.** Sulfur (10 g, 0.31 mol) and **17** (24 g, 82.7 mmol) were combined in 500 mL of 1,2-dichlorobenzene, and the mixture was heated to reflux under nitrogen for 13 h. After cooling to room temperature, the miiture was extracted with 10% HCl. The aqueous extract was washed with $CH_2Cl_2$ and EtOAc, made basic with NaOH, and extracted with $CH_2Cl_2$. The $CH_2Cl_2$ extract was dried over $MgSO_4$ and chromatographed on silica gel with 2% $CH_3OH/CH_2Cl_2$ to provide 14 g of **23** as a yellow solid.

**8-[4-(1*H*-Imidazol-1-yl)phenyl]imidazo[1,2-*a*]pyridine (11). General Method for the Dehydrogenation of 8-Aryl-5,6-dihydroimidazo[1,2-*a*]pyridines.** Compound **7** (4 g, 15.2 mmol) was dissolved in 200 mL of $CH_2Cl_2$, and 10 g of activated $MnO_2$ was added. After the mixture was stirred at room temperature for 24 h, the solids were removed by filtration, and the solvent was removed under vacuum. The residue was crystallized with ether to provide 3 g of product as a white crystalline solid.

**1-Bromo-3-methyl-8-[4-(2-methyl-1*H*-imidazol-1-yl)phenyl]imidazo[1,5-*a*]pyridine (24).** Compound **23** (2.2 g, 7.6 mmol) was combined with *N*-bromosuccinimide (1.4 g, 7.9 mmol) in 50 mL of $CH_2Cl_2$, and the mixture was stirred at room temperature for 1 h. The mixture was washed with 10% $K_2CO_3$, dried over $MgSO_4$, and charcoal treated, and the solvent was removed under vacuum. The residue was crystallized from ether to provide 2.5 g of a light green solid.

**2-(Aminomethyl)-3-[4-(2-methyl-1*H*-imidazol-1-yl)phenyl]pyridine (30). General Method for the Preparation of 8-Arylimidazo[1,5-*a*]pyridin-3-ones.** Compound **23** (7 g, 24.3 mmol) was dissolved in 300 mL of 6 N HCl and heated to 90–100 °C for 20 h. The mixture was made basic with $K_2CO_3$ and extracted with $CH_2Cl_2$. The $CH_2Cl_2$ extracts were dried over $MgSO_4$ and charcoal treated, and the solvent was removed under vacuum. The residue was crystallized from ether to provide 5.5 g (86%) of a white solid: mp 115–117 °C; IR ($CH_2Cl_2$) 3040, 1560, 1510, and 1250 cm$^{-1}$; NMR ($Me_2SO$) δ 2.35 (s, 3), 2.50 (s, 2), 3.76 (s, 2), 6.94 (d, 1 ), 7.36 (d, 1), 7.38 (m, 1), 7.56 (d, 2), 7.59 (d, 2), 7.61 (d, 1), and 8.60 (d, 1). Anal. Calcd for $C_{16}H_{16}N_4$: C, 72.70; H, 6.10; N, 21.20. Found: C, 72.86; H, 6.25; N, 21.03.

**2,3-Dihydro-8-[4-(2-methyl-1*H*-imidazol-1-yl)phenyl]-3-oximidazo[1,5-*a*]pyridine-1-carboxylic Acid Methyl Ester (29).** Compound **30** (5 g, 18.9 mmol) was dissolved in a solution of 10 mL of *N*,*N*-diethylaniline and 100 mL of $CH_2Cl_2$, and the mixture was added dropwise to a solution of trichloromethyl chloroformate (7 g, 35.4 mmol) in 250 mL of $CH_2Cl_2$ at 0 °C. The mixture was allowed to warm to room temperature and then poured into 300 mL of $CH_3OH$. The solvents were removed under vacuum, and the residue was dissolved in $CH_2Cl_2$, washed with $KHCO_3$, and extracted with 1 N KOH. The KOH extract was neutralized with $NH_4Cl$, extracted with methylene chloride, dried over $MgSO_4$, and charcoal treated, and the solvent was removed under vacuum. The residue was crystallized with EtOAc to provide 2.4 g of a yellow solid.

**8-(4-Methylphenyl)imidazo[1,5-*a*]pyridin-3(2*H*)-one (28).** Compound **27** (18 g, 64 mmol) was dissolved in a solution of 500 mL of $CH_2Cl_2$ and 19 mL (130 mmol) of iodotrimethylsilane. The mixture was refluxed for 24 h, cooled to room temperature, and washed with 10% $K_2CO_3$. The $CH_2Cl_2$ portion was dried over $MgSO_4$ and charcoal treated, and the solvent was removed under vacuum. The residue was crystallized with ether to provide 9 g of a yellow solid.

**Papillary Muscle Studies.** Cats (2–4 kg, unselected for sex) were anesthetized with pentobarbital (30 mg/kg). The heart was

(18) Sircar, I.; Duell, B. L.; Bobowski, G.; Bristol, J. A.; Evans, D. B. *J. Med. Chem.* **1985**, *28*, 1405.
(19) Hagedorn, A. A., III; Erhardt, P. W.; Lumma, W. C., Jr.; Wohl, R. A.; Cantor, E.; Chou, Y-L.; Ingebretsen, W. R.; Lampe, J.; Pang, D.; Pease, C. A.; Wiggins, J. *J. Med. Chem.*, following paper in this issue.

removed and rinsed in a physiological salt solution (in millimoles/liter): NaCl, 123; $NaHCO_3$, 25; KCl, 4; $NaH_2PO_4$, 1.40; glucose, 5.5; $MgCl_2$, 0.50; $CaCl_2$, 2.5. The solution was aerated with 95% $O_2$/5% $CO_2$ to give a pH of 7.3–7.5. All solutions were at room temperature, and a papillary muscle of less than 1 mm in diameter and at least 3 mm in length was removed, attached at its distal end to a gold clamp, and placed in a tissue bath (20-mL volume) for study under isometric conditions at 35 °C with physiological salt solution. The muscle was stimulated through platinum point electrodes imbedded in the bottom muscle clamp, using constant current pulses of 1 ms in duration and approximately 10–20% above threshold at a frequency of 0.2 Hz, and then stretched to a resting tension of 250 mg. The muscle was then gradually stretched to the peak of its length–tension curve and allowed to equilibrate for at least 30 min before the study began. Muscles that failed to develop at least 500 mg of force/square millimeter of muscle cross-sectional area were discarded. During the stretching procedures, fresh physiological saline solution was perfused through the system using a flow rate of 1 mL/min. After the end of the equilibration period, the infusion was stopped for the injection of test compound into the bath. The tissue bath was washed for 15 min between dosing. All compounds were dissolved in deionized water and injected directly into the tissue bath (1:1000 dilution) to achieve the concentration desired. Following administration of the last concentration of test compound, norephinephrine (10 μM) was added to the tissue bath (in the presence of the test compound) as an internal standard. In some studies, autonomic blocking agents were used to assess mechanism of action of the positive inotropic effect of the test compounds. For these studies, the blocking agent (1 μM propranolol) was included in the physiological saline and allowed to act for at least 45 min before the test compound was added. All data were digitized and analyzed by a laboratory computer. Sample rate was 200 Hz. Six contractions were averaged to provide one data point, and one data point was stored every minute. The computer calculated developed force, maximum rate of force development, time to peak force, maximum rate of relaxation, and relative rate of relaxation (maximum rate of relaxation divided by developed force). At the end of the experiment, muscle cross-sectional area was calculated by dividing muscle weight by muscle length; all force measurements were then corrected for muscle size by dividing force and maximum rate of force development by muscle cross-sectional area.

**Electrophysiology Studies.** The in vitro Purkinje fiber studies were conducted in a manner analogous to published methods as were the in vivo studies except that the route of administration was iv rather than id.[20] The in vitro canine ventricular muscle studies were conducted in the following manner. Strips taken from the right ventricle papillary muscle near the base (5–6 mm long × 1–2 mm wide) were mounted on a silicone washer and placed in a 3.5-mL tissue bath. The preparation was superfused with warmed (36 ± 0.5 °C), physiological saline equilibrated with a gas mixture of 95% oxygen/5% carbon dioxide. The preparation was stimulated through bipolar, stainless steel Teflon-coated electrodes (0.005 in.) impaled into the muscle. A bipolar electrogram was recorded at the opposite end of the muscle strip with the same type of bipolar electrodes. The output of the electrical signal was amplified and displayed on an oscilloscope. The diastolic threshold (DT) of the muscle preparation was determined with bipolar stimuli 2 ms in duration at a cycle length of 4000 ms. The muscle tissue was then stimulated at 4 times the diastolic threshold for an initial equilibration period of 60 min. The functional refractory period (FRP) was determined at a cycle length of 1000 ms by applying a premature stimulus (S2) of the same duration and strength after every tenth basic stimulus (S1) at decreasing S1–S2 intervals until refractoriness occurred. Conduction time (CT), measured as the time interval from the stimulus to the peak of the electrogram, was determined for each S1–S2 interval that produced a propagated response. The relationship between the degree of prematurity (S1–S2 interval) and conduction time of the premature stimulus (S2) was constructed as a conduction–interval curve. The tissue was then superfused with various concentrations of test compound. Only one compound was tested per ventricular muscle strip preparation.

**Hemodynamic Studies.** Normal mongrel dogs (10–20 kg, unselected for sex) were anesthetized with sodium pentobartital (35 mg/kg, iv) via the cephalic vein. The animals were intubated and mechanically respired at 12 cycles/min (15 mL/kg/per cycle) with oxygen-enriched air. Lead II electrocardiogram was monitored with needle electrodes. The right femoral artery and vein were cannulated for blood pressure determination and for drug infusion, respectively. The heart was exposed through a left lateral thoracotomy (fourth or fifth intercostal space) and pericardial incision. Electromagnetic flow probes were placed around the ascending aorta and left ventricular circumflex artery (proximal to the first branch) and connected to a flow meter for measurement of mean and pulsatile flow. A 21 gauge needle, connected through a catheter to a pressure transducer, was inserted into the left ventricular chamber for pressure measurements. During surgery, approximately 150 mL of lactated Ringers solution was given for fluid replacement and maintenance. Blood gases were monitored and adjusted (altering mechanical respiration) whenever the preparation was unstable (normal values are pH 7.35–7.45, $pCO_2$ = 35–45 mmHg, and $pO_2$ = >80 mmHg). All wounds were covered with saline-soaked surgical gauze and sealed with Parafilm, and the preparation was allowed to equilibrate for at least 30 min or until hemodynamic parameters were stable. Pressure, flow, and ECG data were recorded continuously on an oscillographic recorder. Every 5 min experimental data were digitized and analyzed by computer. For this, the signals were sampled for 5 s, with sampling rates of 800 Hz for ECG, 400 Hz for pressure measurements, and 100 Hz for blood flows. The computer-processed data was used to evaluate the ongoing experiment. Parameters measured or calculated were (1) aortic and left circumflex blood flow, (2) systolic, diastolic, and mean arterial blood pressure, (3) left ventricular systolic and diastolic pressure as well as the rate of maximum pressure development ($dP/dt$) and relaxation ($-dP/dt$), (4) heart rate, (5) total peripheral resistance and rate pressure product, (6) stroke index, (7) lead II ECG, (8) central venous pressure and other parameters as needed. Only dogs demonstrating a basal heart rate between 120 and 170 beats per minute and a mean blood pressure between 80 and 135 mmHg were used for an experiment. Following equilibration and demonstration of stability of the preparation, the hemodynamic response of an iv bolus injection of 0.05 μg/kg of isoproterenol was determined as a positive control. If the response was abnormal (normal response is a mean arterial pressure decrease >15 mmHg, heart rate increase >25%, $dP/dt$ increase >100%, and circumflex blood flow increase >50%), the experimental preparation was adjusted and isoproterenol tried again; otherwise, such animals were not used. Following test compound evaluation, the isoproterenol challenge was repeated as an internal control and a check for any possible β-adrenergic blocking activity of the test compound. Each compound was tested with at least $N = 4$. Doses were taken as high as 100 mg $kg^{-1}$ $min^{-1}$ when no activity was obtained after 30-min observation periods between the dose increments.

**Biochemical Studies.** Crude cAMP phosphodiesterase (phosphodiesterase III) was prepared from homogenate of cardiac muscle tissue from mongrel dogs (10–12 kg) by differential centrifugation between 700*g* and 165000*g*. cAMP phosphodiesterase activities were determined in the presence of $10^{-6}$ M cAMP, $10^5$ cpm/mL [$^3$H]cAMP, 40 mM Tris, pH 8.0, 10 mM $MgCl_2$, 2.5 mM mercaptoethanol, 0.05 mg/mL bovine serum albumin, and 0.1 mg/mL enzyme at 37 °C.[21] The resultant product of tritiated adenosine monophosphate (AMP) and reacted labeled cAMP were separated by ion-exchange thin-layer chromatography with strips of polyethylenimine–cellulose.[22] AMP and cAMP spots on the chromatograms were located under UV light and quantified with liquid scintillation. Due to the presence of other nucleases, the

(20) Morgan, T. K., Jr.; Wohl, R. A.; Lumma, W. C., Jr.; Wan, C-N.; Davey, D. D.; Gomez, R. P.; Marisca, A. J.; Briggs, M.; Sullivan, M. E.; Wong, S. S. *J. Med. Chem.* **1986**, *29*, 1398.

(21) Thompson, W. J.; Terasaki, W. L.; Epstein, P. M.; Strada, S. *J. Adv. Cyclic Nucleotide Res.* **1979**, *10*, 69.

(22) Rangel-Aldao, R.; Schwartz, D.; Rubin, C. S. *Anal. Biochem.* **1978**, *87*, 367.

product, AMP, may also be degraded to adenosine, adenine, and inosine. To correct for further hydrolysis of the product, the radioactivity in the remaining portion of the chromatogram was also quantified and included in the calculation of the cAMP phosphodiesterase activity. Drugs were tested in triplicate up to concentrations as high as 100 $\mu$M.

**Acknowledgment.** We appreciate the technical assistance provided by A. Smart and H. Troy.

**Registry No.** **5**, 104271-32-1; **6**, 104271-38-7; **7**, 104271-43-4; **8**, 104271-33-2; **9**, 104271-34-3; **10**, 104271-50-3; **11**, 104271-44-5; **12**, 104271-51-4; **13**, 107454-13-7; **14**, 107454-22-8; **15**, 107454-17-1; **16**, 107454-18-2; **17**, 107454-18-2; **18**, 108060-51-1; **19**, 108060-52-2; **20**, 108060-53-3; **21**, 107454-23-9; **22**, 107454-26-2; **23**, 107454-24-0; **24**, 108060-54-4; **25**, 108060-55-5; **26**, 108060-56-6; **27**, 108060-57-7; **28**, 108060-58-8; **29**, 108060-59-9; **30**, 107454-29-5; $C_6H_5COnCH_2)_3Cl$, 939-52-6; $4\text{-}H_2NC_6H_4CO(CH_2)_3Cl$, 108060-60-2; $4\text{-}FC_6H_4CO(CH_2)_3Cl$, 3874-54-2; $4\text{-}HOC_6H_4CO(CH_2)_3Cl$, 7150-55-2; $4\text{-}CH_3SO_2NHC_6H_4CO(CH_2)_3Cl$, 108060-61-3; $4\text{-}C_2H_5OC_6H_4CO\text{-}(CH_2)_3Cl$, 75343-08-7; $4\text{-}CH_3CONHC_6H_4CO(CH_2)_3Cl$, 56924-11-9; $ClCOOCCl_3$, 503-38-8; $COCl_2$, 75-44-5; imidazole, 288-32-4; 2-methylimidazole, 693-98-1; 2-ethylimidazole, 1072-62-4.

# Cardiotonic Agents. 2.[1] (Imidazolyl)aroylimidazolones, Highly Potent and Selective Positive Inotropic Agents[2]

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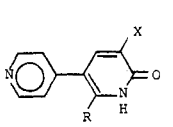
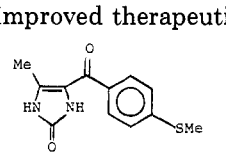
A series of 4-alkyl-1,3-dihydro-5-[(1*H*-imidazolyl)benzoyl]-2*H*-imidazol-2-ones **9** was synthesized and evaluated in vitro for positive inotropic and cyclic AMP phosphodiesterase inhibitory activity. A wide range of inotropic and enzyme-inhibitory potencies was observed, substitution on the imidazolyl moiety being the major determinant of activity. The 4-ethyl-5-[4-(1*H*-imidazol-1-yl)benzoyl] congener **9g** exhibited the highest potency in vitro. Incorporation of a methyl group at the imidazolyl 2-position gave **9h**, which was less potent but remarkably selective in vivo for positive inotropic effects over heart rate and hypotensive effects.

For some 200 years, digitalis and its constituent cardiac glycosides such as digoxin and digitoxin have been used as positive inotropic agents for the treatment of congestive heart failure (CHF). Although these drugs are selective in their inotropic action and exhibit no significant direct effects on the vasculature, their potential for producing cardiac arrhythmia leads to undesirably low therapeutic ratios. This problem and the increasingly high death rate from CHF in developed countries have spurred attempts to find an orally available "digitalis replacement".[3] The discovery[4] of amrinone (**1**) has led to the synthesis of a number of agents that show varying promise for CHF treatment. A few of these compounds are shown in structures **2–5** (**2**, milrinone;[5] **3**, enoximone;[6] **4**, pelrinone;[7] **5**, imazodan[8]). Although their mode of action has not been fully elucidated, it differs from that of digitalis and appears to result, in large part, from inhibition of the low-$K_m$ (cyclic AMP specific) phosphodiesterase (PDE).[9] As noted elsewhere,[3] these compounds also have substantial direct vasorelaxant activity and, therefore, are not strict digitalis replacements. Thus, there still exists a need for a selective inotropic agent having an improved therapeutic ratio.[10]

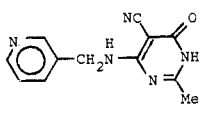


**1** R = H, X = $NH_2$

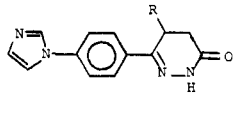
**2** R = Me, X = CN



**3**



**4**



**5a** R = H

**5b** R = Me

Recently, we discovered two series of cardioactive compounds, the imidazopyridines **6** and the related imidazopyridinones (e.g., **7**),[1] where the attachment of an imidazolyl substituent to an aromatic ring leads to improved activity. To further explore the generality of this relationship, we thought that it would be useful to examine less rigid versions of **7**. The aroylimidazolones **8**, first prepared by Duschinsky and Dolan[11] (**8a**) and more recently elaborated as cardiotonic agents by Schnettler et al.[6a,12] (**3**), provided a suitable framework on which to test

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(1) For paper 1, see: Davey, D.; Erhardt, P. W.; Lumma, W. C., Jr.; Wiggins, J.; Sullivan, M.; Pang, D.; Cantor, E. *J. Med. Chem.*, preceding paper in this issue.
(2) Presented in part at the 192nd American Chemical Society National Meeting, Anaheim, CA, September 1986 (MEDI 10), and at the IXth International Symposium on Medicinal Chemistry, Berlin (West), September 1986 (Abstract 110).
(3) Erhardt, P. W. *J. Med. Chem.* **1987**, *30*, 231.
(4) Alousi, A. A.; Farah, A. E.; Lesher, G. Y.; Opalka, C. J. *Circ. Res.* **1979**, *45*, 666.
(5) Braunwald, E.; Sonnenblick, E. J.; Chakrin, L. W.; Schwartz, R. P., Eds. *Milrinone: Investigation of New Inotropic Therapy for Congestive Heart Failure*; Raven: New York, 1984.
(6) (a) Schnettler, R. A.; Dage, R. C.; Grisar, J. M. *J. Med. Chem.* **1982**, *25*, 1477. (b) Petein, M.; Levine, T. B.; Weingarten, J.; Cohn, J. N. *Clin. Res.* **1984**, *32*, 198A.
(7) McQuillan, J.; Bagli, J.; Grimes, D.; Lee, D.; Metcalf, G. *Pharmacologist* **1984**, *26*, Abstr 204.
(8) (a) Bristol, J. A.; Sircar, I.; Moos, W. H.; Evans, D. B.; Weishaar, R. E. *J. Med. Chem.* **1984**, *27*, 1101. (b) Sircar, I.; Duell, B. L.; Bobowski, G.; Bristol, J. A.; Evans, D. B. *J. Med. Chem.* **1985**, *28*, 1405.
(9) Weishaar, R. E.; Cain, M. H.; Bristol, J. A. *J. Med. Chem.* **1985**, *28*, 537 and references therein.
(10) Like digitalis, such an agent could be used independently for its inotropic effect or, when desired, administered with a vascular drug that has been specifically tailored for use in treating CHF. In the latter case, complementary inotropic and vasodilatory effects could then be readily balanced through separate dosage adjustment in order to provide maximum combination therapy.
(11) (a) Duschinsky, R.; Dolan, L. A. *J. Am. Chem. Soc.* **1945**, *67*, 2079. (b) Duschinsky, R.; Dolan, L. A. *J. Am. Chem. Soc.* **1946**, *68*, 2350.

0022-2623/87/1830-1342$01.50/0 