Eur. J. Med. Chem. (1990) 25, 187–190
© Elsevier, Paris

Short communication

Synthesis and cardiotonic activity of 2-indolinones

Aldo ANDREANI*[1], Mirella RAMBALDI[1], Alessandre LOCATELLI[1], Rosaria BOSSA[2], Iraklis GALATULAS[2] and Mauro NINCI[2]

[1]*Dipartimento di Scienze Farmaceutiche, University of Bologna, Via Belmero 6, 40126 Bologna; and*
[2]*Dipartimento di Farmacologia, CTM, University of Milan, Via Vanvitelli 32, 20129 Milan; Italy*

(Received Decembre 28, 1988; accepted April 27, 1989)

Summary – The paper reports the synthesis of 3-(2,5-dimethoxybenzylidene)- **2** and 3-(4-pyridylmethylene)-2-indolinones **3** which, as expected, proved to have significant positive inotropic activity. On the contrary, 3-(2,5-dimethoxybenzyl)- **4** and 3-(4-pyridylmethyl)-2-indolinones **5**, obtained by reduction of the corresponding compounds **2** and **3**, were devoid of cardiotonic activity.

Résumé – **Synthèse et activité cardiotonique d'indolin-2-ones.** *Les auteurs rapportent la synthèse de 3(2,5-diméthoxybenzylidène)- et 3-(4-pyridylméthylène)-indolin-2-ones, **2** et **3** présentant, comme attendu une activité inotrope positive significative. En revanche, les 3-(2,5-diméthoxybenzyl)- et 3-(4-pyridylméthyl)-indolin-2-ones, **4** et **5**, obtenues par réduction des composés correspondants **2** et **3** sont dépourvues d'activité cardiotonique.*

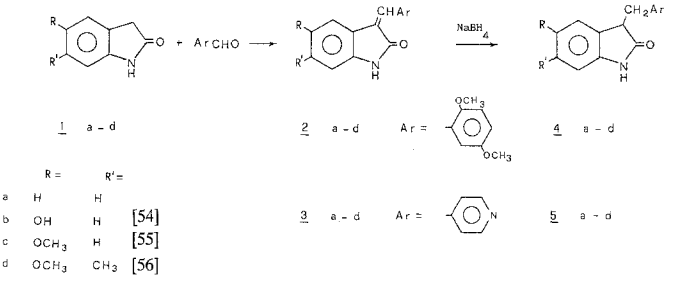
2-indolinone / cardiotonic activity

Introduction

In our previous papers on the synthesis of new cardiotonic agents [1–5], we reported a number of imidazo[2,1-*b*] thiazole derivatives and we stressed in particular 2 important pharmacophores: the 2,5-dimethoxyphenyl and the pyridyl group. All the molecules considered were devoid of a cyclic carboxamido group which is present in many of the most widely known positive inotropic agents such as amrinone, milrinone and other pyridinones [6–11], pimobendan [12, 13], imazodan and other pyridazinones-pyrazolones [14–24], piroximone and other imidazolones [25–31], OPC 8212 and other quinolones-quinazolones [32–42], adibendan and other indolinones [43–47], RS 82856 [48–50], SKF 94120 [51]; in compound LY 195115 (indolidan) [52, 53], the group is present twice in opposite parts of the molecule.

Chemistry

On the basis of these considerations we planned the synthesis of new compounds bearing a cyclic carboxamido group and one of the aforementioned pharmacophores which produced imidazo[2,1-*b*] thiazoles with significant cardiotonic activity [1–5]. The structure of choice was a 2-indolinone **1a–d** (see Scheme 1) which, reacted with the proper aldehyde, gave the desired 3-(2,5-dimethoxybenzylidene)- **2a–d** and 3-(4-pyridylmethylene)-2-indolinones **3a–d**. Compounds **2** and **3** were obtained in the presence of piperidine: it was already suggested for analogous compounds that, under basic conditions, the yield of the E-form is very high whereas the Z-isomer is present as a trace only or may even be absent [58]. Both the configurations were discussed with respect to IR and NMR data [58, 59]. For these reasons we believe that compounds **2** and **3** belong to the E-configuration. These derivatives, treated with sodium borohydride, could be reduced to 3-(2,5-dimethoxybenzyl)- **4a, c, d** and 3-(4-pyridylmethyl)-2-indolinones **5a, d.** The ^{1}H NMR spectra of compounds **4** and **5** show that the $CHCH_2$ group gives rise to a set of signals which is typical of diastereotopic protons. Compounds **2–5** are reported in Tables I, II. We did not complete the series with the synthesis of compounds **4b** and **5b, c** as all the other compounds **4** and **5**, compared with the corresponding **2** and **3**, showed a dramatic drop in activity or even a negative inotropic potential (see *Pharmacological results*).



*Author to whom correspondence should be addressed.

Table I. 2-Indolinones **2**–**5**.

Compound	R	R'	Ar	Formula (MW)	Anal.	Mp (°C)
2 a	H	H	2,5-dimethoxyphenyl	$C_{17}H_{15}NO_3$ (281.3)	C,H,N	193–195
2 b	OH	H	"	$C_{17}H_{15}NO_4$ (297.3)	"	270–273 dec.
2 c	OCH_3	H	"	$C_{18}H_{17}NO_4$ (311.3)	"	207–210
2 d	OCH_3	CH_3	"	$C_{19}H_{19}NO_4$ (325.3)	"	179–181
3 a	H	H	4-pyridyl	$C_{14}H_{10}N_2O$ (222.2)		228–230 [a]
3 b	OH	H	"	$C_{14}H_{10}N_2O_2$ (238.2)	"	>300
3 c	OCH_3	H	"	$C_{15}H_{12}N_2O_2$ (252.3)	"	211–214
3 d	OCH_3	CH_3	"	$C_{16}H_{14}N_2O_2$ (266.3)	"	205–207
4 a	H	H	2,5-dimethoxyphenyl	$C_{17}H_{17}NO_3$ (283.3)	"	148–152
4 c	OCH_3	H	"	$C_{18}H_{19}NO_4$ (313.3)	"	132–135
4 d	OCH_3	CH_3	"	$C_{19}H_{21}NO_4$ (327.3)	"	118–120
5 a	H	H	4-pyridyl	$C_{14}H_{12}N_2O$ (224.3)		195–198 [b]
5 d	OCH_3	CH_3	"	$C_{16}H_{16}N_2O_2$ (268.3)	"	197–198

[a]Lit. 229–231°C dec. [57].
[b]Lit. 199–201°C dec. [57].

Table II. IR and ^1H NMR of compounds **2**–**5** (py = pyridine).

Compound	$\nu_{max.}$ (cm^{-1})	δ (ppm), J (Hz)
2 a	1690, 1630, 1610, 1220	3.70(3H,s,OCH_3) 3.78(3H,s,OCH_3) 6.88(2H,m,ar.) 7.20(4H,m,ar.) 7.50(1H,d,ar.) 7.66(1H,s, CH) 10.60(1H,s,NH)
2 b	3400-3050, 1675, 1595, 1215	3.78(3H,s,OCH_3) 3.82(3H,s,OCH_3) 6.70(2H,s,ar.) 7.08(3H,s,ar.) 7.25(1H,s,ar.) 7.60(1H,s, CH) 9.03(1H,s,OH) 10.35(1H,s,NH)
2 c	1700, 1220, 1200, 1040	3.62(3H,s,OCH_3) 3.75(3H,s,OCH_3) 3.82(3H,s,OCH_3) 6.87(2H,m,ar.) 7.10(1H,m,ar.) 7.15(2H,m,ar.) 7.30(1H,m,ar.) 7.68(1H,s, CH) 10.48(1H,s,NH)
2 d	1715, 1270, 1215, 1080	2.20(3H,s,CH_3) 3.60(3H,s,OCH_3) 3.80(3H,s,OCH_3) 3.88(3H,s,OCH_3) 6.77(1H,s,ar.) 7.14(3H,m,ar.) 7.35(1H,m,ar.) 7.65(1H,s, CH) 10.40(1H,s,NH)
3 a	1725, 1710, 1610, 1595	6.90(2H,m,ar.) 7.35(2H,m,ar.) 7.60(1H,s, CH) 7.67(2H,m,py 3,5) 8.80(2H,m,py 2,6) 10.80(1H,s,NH)
3 b	3500-3100, 1700, 1600, 1210	6.72(3H,m,ar.) 7.55(1H,s, CH) 7.65(2H,m,py 3,5) 8.78(2H,m,py 2,6) 9.08(1H,s,OH) 10.50(1H,s,NH)
3 c	1710, 1590, 1480, 1440	3.60(3H,s,OCH_3) 6.88(3H,m,ar.) 7.60(1H,s, CH) 7.68(2H,m,py 3,5) 8.80(2H,m,py 2,6) 10.52(1H,s,NH)
3 d	1700, 1600, 1190, 1080	2.15(3H,s,CH_3) 3.55(3H,s,OCH_3) 6.71(1H,s,ar.) 6.95(1H,s,ar.) 7.50(1H,s, CH) 7.70(2H,m,py 3,5) 8.80(2H,m,py 2,6) 10.48(1H,s,NH)
4 a	1710, 1495, 1220, 1015	2.70-3.35-3.75(3H,m, CH-CH_2) 3.68(3H,s,OCH_3) 3.72(3H,s,OCH_3) 6.60(1H,m,ar.) 6.80(5H,m,ar.) 7.10(1H,m,ar.) 10.45(1H,s,NH)
4 c	1690, 1315, 1220, 1035	2.75-3.30-3.68(3H,m, CH-CH_2) 3.55(3H,s,OCH_3) 3.65(3H,s,OCH_3) 3.71(3H,s,OCH_3) 6.22(1H,s,ar.) 6.75(3H,m,ar.) 6.90(2H,m,ar.) 10.27(1H,s,NH)
4 d	1690, 1495, 1225, 1020	2.08(3H,s,CH_3) 2.62-3.30-3.65(3H,m, CH-CH_2) 3.48(3H,s,OCH_3) 3.68(3H,s,OCH_3) 3.72(3H,s,OCH_3) 6.15(1H,s,ar.) 6.62(1H,s,ar.) 6.75(1H,s,ar.) 6.90(2H,m,ar.) 10.20(1H,s,NH)
5 a	1710, 1600, 1230, 745	3.18-3.90(3H,m, CH-CH_2) 7.0(4H,m,ar.) 7.20(2H,m,py 3,5) 8.45(2H,m,py 2,6) 10.45(1H,s,NH)
5 d	1710, 1605, 1190, 1130	2.08(3H,s,CH_3) 3.10(3H,m, CH-CH_2) 3.66(3H,s,OCH_3) 6.51(1H,s,ar.) 6.72(1H,s,ar.) 7.03(2H,m,py 3,5) 8.38(2H,m,py 2,6) 10.0(1H,s,NH)

Pharmacological results

The positive inotropic activity of compounds **2–5** was tested on spontaneously beating guinea pig atria (see *Experimental protocols*). All the 3-(2,5-dimethoxybenzylidene)-2-indolinones **2a–d** produced a fleeting increase of the contractile force with the following rank of potency: **2a > 2d > 2c > 2b**. Unfortunately, due to their poor solubility in the test medium, it was not possible to complete the experiments and to draw quantitative data. On the other hand, all the reduced derivatives **(4a, 4c, 4d, 5a, 5d)** were inactive or produced a negative inotropic effect which, in some cases, completely stopped atrial beating. We did not consider further study of these compounds as potential calcium antagonists to be useful, because the block was irreversible. The positive inotropic activity of the 3-(4-pyridylmethylene)-2-indolinones **3** is reported in Table III in comparison to sulmazole used as a reference. The data collected to date show that our initial approach was correct and clearly outline the important role played by the double bond at the 3-position.

Experimental protocols

Chemistry

The melting points are uncorrected. Analyses indicated by the symbols of the element were within ±0.4% of the theoretical values. Bakerflex plates (silica gel IB2-F) were used for TLC and Kieselgel 60 (Merck) for column chromatography: the eluent was a mixture of petroleum ether/acetone in various proportions. The IR were recorded in Nujol on a Perkin–Elmer 298. The ^{1}H NMR were recorded in DMSO–d_6 on a Varian EM-390 (90 MHz) using tetramethylsilane as the internal standard. 2-Indolinone is commercially available (Aldrich); the other indolinones were prepared according to the literature (see Scheme 1).

*3-(2,5-Dimethoxybenzylidene)-2-indolinones **2a–d***
The appropriate 2-indolinone **1** (75 mmol) was dissolved in 100 ml of methanol and treated with 13.3 g (80 mmol) of 2,5-dimethoxybenzaldehyde and 3 ml of piperidine. The reaction mixture was refluxed for 3 h: after cooling, the resulting precipitate was collected anc crystallized from methanol with a yield of 85–95%. Only for compound **2b**, toluene (500 ml) was employed as the solvent and the crude precipitate was purified by column chromatography.

*3-(4-Pyridylmethylene)-2-indolinones **3a–d***
The appropriate 2-indolinone **1** (75 mmol) was dissolved in 50 ml of methanol and treated with 7.7 ml (8.6 g, 80 mmol) of 4-pyridinecarboxaldehyde and 3 ml of piperidine. The reaction mixture was refluxed for 3 h and, after working up as above described, 80–90% of the expected compounds was obtained.

*3-(2,5-Dimethoxybenzyl)-2-indolinones **4a, c, d** and 3-(4-pyridylmethyl)-2-indolinones **5a, d***
The appropriate 2-indolinone **2–3** (4 mmol) was dissolved in 100 ml of methanol and treated portionwise with 1.5 g (40 mmol) of sodium borohydride. The mixture was stirred at room temperature for 12 h and the solvent was then evaporated under reduced pressure. The precipitate formed by adding water was pure on TLC. With this method compound **4a** was obtained in almost quantitative yield. The other compounds, **4** and **5**, were prepared with yields ranging from 50 to 60% and the crude precipitates were crystallized from methanol (compound **5d** was first purified by column chromatography; compound **5a** was crystallized from petroleum ether).

Table III. Positive inotropic activity of the indolinones **3.**

Compound	EC_{50} (μg/ml)	MAXIMUM INCREASE IN CONTRACTILE FORCE Changes in % from baseline value = 100 [a]	Concentration (μg/ml)
3 a	14.4	166 ± 22.8	50
3 b	50.2	171 ± 7.7	300
3 c	6.9	146 ± 8.2	45
3 d	13.6	159 ± 5.6	40
Sulmazole	4.2	163 ± 9.5	100

[a]Initial contractile force = 0.54 ± 0.17 g.

Pharmacology

The experiments were carried out on spontaneously beating isolated atria of guinea pig (350–500 g body weight). The preparations were suspended, at 37°C, in a 20 ml bath of Tyrode solution (composition in g/l: NaCl: 8.0; $NaHCO_3$: 1.0; KCl: 0.2; NaH_2PO_4: 0.005; $MgCl_2$: 0.1; $CaCl_2$: 0.2; glucose: 1.0). An initial tension of 1 g was applied to the preparation. Isometric contractions were recorded by a strain gauge transducer connected to a recording microdynamometer. After the basal responses were taken, the test compounds were administered at 5–400 μg/ml on a cumulative basis and responses were recorded. The contact time for each dose was 5 min. Concentrations producing 50% of the maximal effect (EC_{50}) were calculated from concentration-response curves (mean values of 3–5 atria).

References

1 Andreani A., Rambaldi M., Bonazzi D., Lelli G., Bossa R. & Galatulas I. (1984) *Eur. J. Med. Chem.* 19, 219
2 Andreani A., Rambaldi M., Andreani F., Bossa R. & Galatulas I. (1985) *Eur. J. Med. Chem.* 20, 93
3 Andreani A., Rambaldi M., Bonazzi D., Bossa R. & Galatulas I. (1985) *Arch. Pharm.* 318, 1003
4 Andreani A., Rambaldi M., Andreani F., Bossa R. & Galatulas I. (1986) *Eur. J. Med. Chem.* 21, 55
5 Andreani A., Rambaldi M., Mascellani G., Bossa R. & Galatulas I. (1986) *Eur. J. Med. Chem.* 21, 451
6 (1979) *Drugs Future* 4, 245
7 (1982) *Drugs Future* 7, 757
8 (1985) *Annual Drug Data Rep.* 7, 572
9 (1987) *Annual Drug Data Rep.* 9, 217
10 (1988) *Drugs Future* 13, 514
11 Sircar I., Duell B.L., Bristol J.A., Weishaar R.E. & Evans D.B. (1987) *J. Med. Chem.* 30, 1023
12 (1985) *Drugs Future* 10, 570
13 Wetzel B. (1988) *Actual. Chim. Thér.* 15, 83
14 (1984) *Drugs Future* 9, 256
15 Bristol J.A., Sircar I., Moos W.H., Evans D.B. & Weishaar R.E. (1984) *J. Med. Chem.* 27, 1099
16 Sircar I., Duell B.L., Bobowski G., Bristol J.A. & Evans D.B. (1985) *J. Med. Chem.* 28, 1405
17 Sircar I., Bobowski G., Bristol J.A., Weishaar R.E. & Evans D.B. (1986) *J. Med. Chem.* 29, 261
18 Sircar I., Duell B.L., Cain M.H., Burke S.E. & Bristol J.A. (1986) *J. Med. Chem.* 29, 2142
19 Sircar I., Morrison G.C., Burke S.E., Skeean R. & Weishaar R.E. (1987) *J. Med. Chem.* 30, 1724
20 Sircar I., Weishaar R.E., Kobylarz D., Moos W.H. & Bristol J.A. (1987) *J. Med. Chem.* 30, 1955

21 Sircar I., Steffen R.P., Bobowski G., Burke S.E., Newton R.S., Weishaar R.E., Bristol J.A. & Evans D.B. (1989) *J. Med. Chem.* 32, 342
22 (1987) *Drugs Future* 12, 856
23 Okushima H., Narimatsu A., Kobayashi M., Furuya R., Tsuda K. & Kitada Y. (1987) *J. Med. Chem.* 30, 1157
24 Robertson D.W., Krushinski J.H., Don Pollock G. & Scott Hayes J. (1988) *J. Med. Chem.* 31, 461
25 (1984) *Drugs Future* 9, 905
26 (1983) *Drugs Future* 8, 343
27 Schnettler R.A., Dage R.C. & Grisar J.M. (1982) *J. Med. Chem.* 25, 1477
28 Dage R.C., Roebel L.E., Hsieh C.P., Weiner D.L. & Woodward J.K. (1982) *J. Cardiovasc. Pharmacol.* 4, 500
29 Dage R.C., Roebel L.E., Hsieh C.P. & Woodward J.K. (1984) *J. Cardiovasc. Pharmacol.* 6, 35
30 Schnettler R.A., Dage R.C. & Palopoli F.P. (1986) *J. Med. Chem.* 29, 860
31 Hagedorn III A.A., Erhardt P.W., Lumma Jr. W.C., Wohl R.A., Cantor E., Chou Y., Ingebretsen W.R., Lampe J.W., Pang D., Pease C.A. & Wiggins J. (1987) *J. Med. Chem.* 30, 1342
32 (1985) *Drugs Future* 10, 28
33 Shimizu T., Osumi T., Niimi K. & Nakagawa K. (1984) *Arzneim. Forsch.* 34, 334
34 Tominaga M., Yo E., Ogawa H., Yamashita S., Yabuuchi Y. & Nakagawa K. (1986) *Chem. Pharm. Bull.* 34, 682
35 Ogawa K., Tamada S., Fujioka T., Teramoto S., Kondo K., Yamashita S., Yabuuchi Y., Tominaga M. & Nakagawa K. (1988) *Chem. Pharm. Bull.* 36, 2253
36 Ogawa H., Tamada S., Fujioka T., Teramoto S., Kondo K., Yamashita S., YabuuchiY., Tominaga M. & Nakagawa K. (1988) *Chem. Pharm. Bull.* 36, 2401
37 Alabaster C.T., Bell A.S., Campbell S.F., Ellis P., Henderson C.G., Roberts D.A., Ruddock K.S., Samuels G.M.R. & Stefaniak M.H. (1988) *J. Med. Chem.* 31, 2048
38 Alabaster C.T., Bell A.S., Campbell S.F., Ellis P., Henderson C.G., Morris D.S., Roberts D.A., Ruddock K.S., Samuels G.M.R. & Stefaniak M.H. (1989) *J. Med. Chem.* 32, 575
39 Leclerc G., Marciniak G., Decker N. & Schwartz J. (1986) *J. Med. Chem.* 29, 2427
40 Decker N., Grima M., Velly J., Marciniak G., Leclerc G. & Schwartz J. (1987) *Arzneim. Forsch.* 37, 1108 and 1233
41 Bandurco V.T., Schwender C.F., Bell S.C., Combs D.W., Kanojia R.M., Levine S.D., Mulvey D.M., Appollina M.A., Reeds M.S., Malloy E.A., Falotico R., Moore J.B. & Tobia A.J. (1987) *J. Med. Chem.* 30, 1421
42 Kaiho T., San-nohe K., Kajiya S., Suzuki T., Otsuka K., Ito T., Kamiya J. & Maruyama M. (1989) *J. Med. Chem.* 32, 351
43 (1988) *Drugs Future* 13, 102
44 Mertens A., Müller-Beckmann B., Kampe W., Hölck J.P. & von der Saal W. (1987) *J. Med. Chem.* 30, 1279
45 Robertson D.W., Jones N.D., Krushinski J.H., Don Pollock G., Swartzendruber J.K. & Scott Hayes J. (1987) *J. Med. Chem.* 30, 623
46 Robertson D.W., Krushinski J.H., Don Pollock G., Wilson H., Kauffman R.F. & Scott Hayes J. (1987) *J. Med. Chem.* 30, 824
47 (1987) *Annual Drug Data Rep.* 9, 930
48 (1984) *Annual Drug Data Rep.* 6, 326
49 Jones G.H., Venuti M.C., Alvarez R., Bruno J.J., Berks A.H. & Prince A. (1987) *J. Med. Chem.* 30, 295
50 Venuti M.C., Jones G.H., Alvarez R. & Bruno J.J. (1987) *J. Med. Chem.* 30, 303
51 (1986) *Drugs Future* 11, 129
52 (1986) *Drugs Future* 11, 377
53 Robertson D.W., Krushinski J.H., Beedle E.E., Wyss V., Don Pollock G., Wilson H., Kauffman R.F. & Scott Hayes J. (1986) *J. Med. Chem.* 29, 1832
54 Beer R.J.S., Davenport H.F. & Robertson A. (1953) *J. Chem. Soc. (Lond.)* 1262
55 Koelsch C.F. (1944) *J. Am. Chem. Soc.* 66, 2019
56 Andreani A., Rambaldi M., Bonazzi D., Greci L. & Andreani F. (1979) *Farm. Ed. Sci.* 34, 132
57 Walker G.N., Smith R.T. & Weaver B.N. (1965) *J. Med. Chem.* 8, 626
58 Tacconi G. and Marinone F. (1968) *Ric. Sci.* 38, 1239
59 Tacconi G., Dacrema Maggi L., Righetti P., Desimoni G., Azzolina O. & Ghislandi V. (1976) *J. Chem. Soc. Perkin Trans. II* 12, 150