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Synthesis and Evaluation of 1,1′-Hydrocarbylenebis(indazol-3-ols) as Potential Antimalarial Drugs

Miriam Martins Alho,[a] Rory N. García-Sánchez,[b] Juan José Nogal-Ruiz,[b] José Antonio Escario,[b] Alicia Gómez-Barrio,[b] Antonio R. Martínez-Fernández,*[b] and Vicente J. Arán*[c]

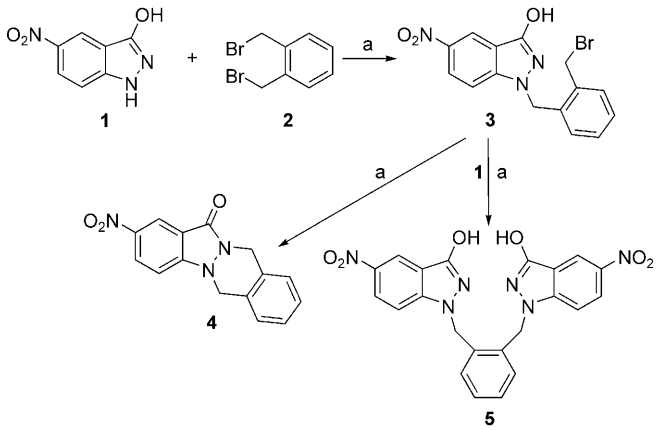
This paper is dedicated to our dear colleagues Drs. Vicente Gómez Parra and Salvador Vega Noverola (IQM, CSIC, Madrid) for their outstanding contribution to Medicinal Chemistry, on the occasion of their retirement.

*Bis(indazol-3-ol) derivatives (**5**, **30–38**) were prepared by alkylation of 3-alkoxyindazoles with α,ω-dibromides, followed by removal of the O-protecting groups. These compounds were subsequently evaluated as inhibitors of biocrystallization of ferriprotoporphyrin IX (heme) to hemozoin, a* Plasmodium *detoxification specific process. Most bis(5-nitroindazol-3-ols) were good inhibitors, however, a denitro analogue (**38**), the intermediate bis(3-alkoxyindazoles) (**15–29**) as well as bis(indazolin-3-ones) (**39–42**) were not active, showing the importance of the NO_2 and OH groups in the inhibition process.*

Introduction

In recent years, we have developed different methods of the synthesis of indazoles,[1–4] indoles,[5] cinnolines[6] and quinoxalines.[7] Some of these compounds have shown interesting properties as cytostatic drugs[2–4] and/or antiparasitic agents against *Trypanosoma cruzi*[4,8] and *Trichomonas vaginalis*.[4,9]

In silico investigations followed by the synthesis and screening of the predicted active compounds as inhibitors of biomineralization (or more appropriately, biocrystallization) of ferriprotoporphyrin IX (heme) to hemozoin,[10] led to the discovery of indazole **5**, and its potential as an antimalarial drug.[11] 1,1′-(*o*-Xylylene)bis(indazol-3-ol) (**5**) had previously been obtained in low yield (24%) as a by-product in the preparation of the cytostatic agent **4** starting from 5-nitroindazolol (**1**) and α,α′-dibromo-*o*-xylene (**2**) (Scheme 1).[2]



Scheme 1. Reported synthesis of compound **5**. *Reagents and conditions*: a) K_2CO_3, acetone, reflux, 5 h.

Malaria, mainly caused by the protozoan *Plasmodium falciparum*, is one of the most deadly diseases in the world, affecting 400–500 million people and causing 1–2 million deaths each year. *Plasmodium* ingest and digest large amounts of hemoglobin within their digestive vacuole at the intraerythrocytic stage, in order to obtain the amino acids needed to synthesize their own proteins. After cleavage of the peptidic chain, the remaining ferriprotoporphyrin IX is toxic and it is converted through a *Plasmodium* specific biocrystallization process into the inert pigment called hemozoin (β-hematin). Chloroquine, a known antimalarial agent hypothesized to act by inhibiting the detoxification of heme,[12,13] has been the mainstay of antimalarial treatments for decades but, the emergence of resistant parasites in the early 1960 s has seriously limited its efficacy. It is generally assumed that this same mechanism, i.e., the prevention of hemozoin formation leading to enhanced heme toxicity, also mediates the antimalarial activities of some chloroquine analogues, antifungal imidazoles, xanthones, porphyr-

[a] *Dr. M. Martins Alho*
CIHIDECAR (CONICET), Departamento de Química Orgánica
Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires
1428 Buenos Aires (Argentina)

[b] *R. N. García-Sánchez, Dr. J. J. Nogal-Ruiz, Dr. J. A. Escario, Dr. A. Gómez-Barrio, Dr. A. R. Martínez-Fernández*
Departamento de Parasitología, Facultad de Farmacia
Universidad Complutense, 28040 Madrid (Spain)
Fax: (+34) 91-394-1815
E-mail: arm@farm.ucm.es

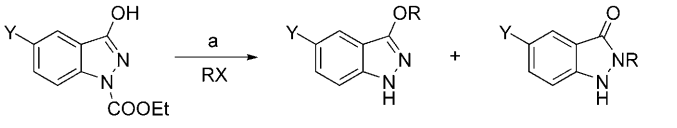
[c] *Dr. V. J. Arán*
Instituto de Química Médica (IQM), CSIC
c/Juan de la Cierva 3, 28006 Madrid (Spain)
Fax: (+34) 91-564-4853
E-mail: vjaran@iqm.csic.es

ins, acridines, etc., as well as that of methylene blue and some related phenothiazines.[13] Much of the current efforts against malaria are directed towards the identification of new targets and drugs, but the therapeutic void produced by the development of resistance to chloroquine, and related analogues, has not yet been filled satisfactorily.[14]

Results and Discussion

Owing to the therapeutic potential of compound **5**, we first attempted to improve the efficacy of the synthesis shown in Scheme 1. We carried out modifications in reaction temperature, solvents, molecular ratios of reagents, etc., however, no improvement in yield was achieved; in all cases, the intramolecular cyclization of the 1-substituted indazolol intermediate **3** to the fused derivative **4** is preferred over reaction with a second molecule of compound **1** to yield *o*-xylylenebis(indazol-3-ol) (**5**).

This led us to develop a different method for the preparation of compound **5** and several analogues carrying different linker moieties between the two indazole systems. The synthesis was based on the alkylation of 3-alkoxyindazole derivatives **8–10**, a reaction that is known to afford 3-alkoxy-1-alkylindazoles exclusively.[15,16] The 3-alkoxy analogues of compound **3**, such as the expected intermediates **14** are not able to undergo intramolecular cyclization (Scheme 3). 3-Alkoxyindazoles **8–10** were prepared, together with the corresponding 2-alkylindazolinones **11–13**, respectively, by treatment of 1-(ethoxycarbonyl)indazolols **6** and **7** with the corresponding alkyl halides, followed by removal of 1-protecting group as previously described (Scheme 2).[3,15,17]

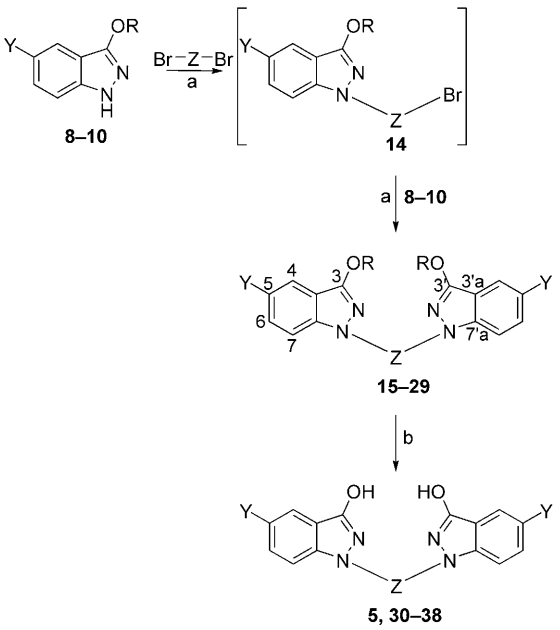


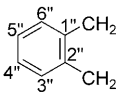
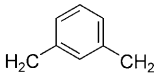
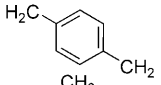
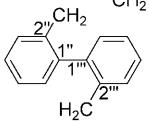
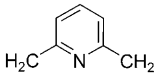
6 Y = NO_2
7 Y = H

8 R = Bn, Y = NO_2
9 R = Me, Y = NO_2
10 R = Me, Y = H

11 R = Bn, Y = NO_2
12 R = Me, Y = NO_2
13 R = Me, Y = H

Scheme 2. Synthesis of 3-alkoxyindazoles **8–10** and the corresponding 2-alkylindazolin-3-ones **11–13**. *Reagents and conditions*: a) K_2CO_3 or Cs_2CO_3, acetone, reflux, 24 h, and then KOH, EtOH, RT, 1 h.



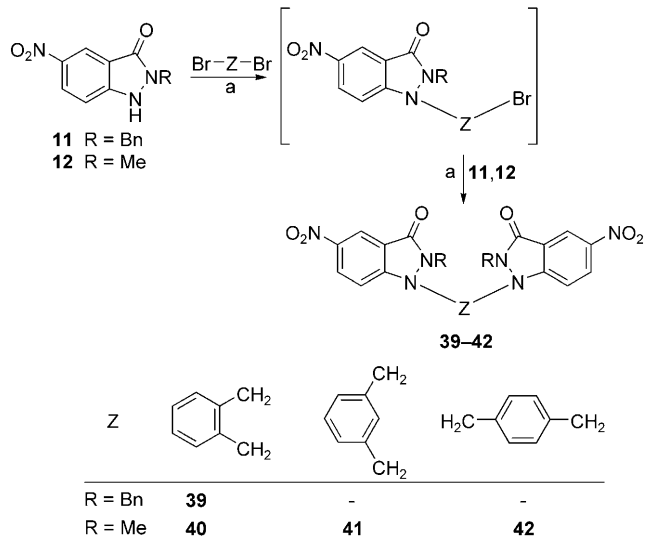
Z	R = Bn Y = NO_2	R = Me Y = NO_2	R = Me Y = H	Y = NO_2	Y = H
	**15**	**20**	**29**	**5**	**38**
	**16**	**21**	-	**30**	-
	**17**	**22**	-	**31**	-
	**18**	**23**	-	**32**	-
	**19**	**24**	-	**33**	-
$[CH_2]_2$	-	**25**	-	**34**	-
$[CH_2]_4$	-	**26**	-	**35**	-
$[CH_2]_5$	-	**27**	-	**36**	-
$[CH_2]_6$	-	**28**	-	**37**	-

Scheme 3. Synthesis of 1,1′-hydrocarbylenebisindazole derivatives **5**, **15–29** and **30–38**. *Reagents and conditions*: a) K_2CO_3, acetone, reflux, 3–24 h; b) 48% aq HBr, reflux, 1–3 h.

Alkylation of compounds **8–10** with the required α,ω-dibromides afforded bis(3-alkoxyindazole) derivatives **15–29** in good yields (Scheme 3). In the synthesis of 1,1′-ethylenebis(3-methoxyindazole) **25**, the 1-(2-bromoethyl)indazole intermediate **14** (R = CH_3, Y = NO_2, Z = $[CH_2]_2$) underwent partial hydrogen bromide elimination to afford the corresponding 3-methoxy-1-vinyl derivative as a by-product; consequently, some starting material **9** remained unreacted in this process.

We have previously described the sensitivity of 3-(benzyloxy)-indazoles to acids;[3,4] treatment of compounds **15–19** with hydrobromic acid afforded the corresponding indazol-3-ols **5**, **30–33**. Analogously, we have found that 3-methoxy substituted indazoles, like the corresponding methoxy-substituted pyrazoles,[18] can also be cleaved with hydrobromic acid. Following this method, we have prepared compounds **5**, **30–33**, polymethylenebis(indazol-3-ols) **34–37**, and *o*-xylylene derivative **38**, a denitro analogue of the lead compound **5**. Additionally, starting from 2-substituted indazolinones **11** and **12** and the corresponding α,ω-dibromides, we also prepared bis(indazolin-3-one) derivatives **39–42** (Scheme 4). According to the literature data, alkylation of the 2-substituted indazolin-3-ones affords only the corresponding 1,2-disubstituted derivatives. [3,15,16]

The prepared indazoles were tested as inhibitors of ferriprotoporphyrin IX biocrystallization;[11] hydrocarbylenebis(indazol-



Scheme 4. Synthesis of 1,1′-hydrocarbylenebis(indazolin-3-ones) **39**–**42**. *Reagents and conditions*: a) K_2CO_3, acetone, reflux, 10–20 h.

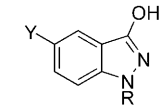
3-ols) (**30**–**33**, **36**, **37**) were, in most cases, more active than the control (chloroquine diphosphate, $IC_{50} = 28$ μM), but similar in activity to that of the lead compound **5** (Table 1). Only ethylene derivative **34** and *o*-xylylene derivative **38** were inactive, while tetramethylenebis(indazol-3-ol) (**35**) could not be evaluated owing to low solubility. The 3-benzyloxyindazoles (**15**, **16** and **19**), 3-methoxyindazoles (**20**–**24**, **27** and **28**) and bis(indazolin-3-one) **42** were all inactive, indicating the importance of the acidic (ionizable) 3-OH group of the indazolol moiety to biological activity. In fact, both OH groups, i. e. the bis(indazol-3-ol) structure, play a crucial role in the activity of the molecule, as simple 1-substituted 5-nitroindazol-3-ols show only very weak inhibitory activity [e. g. the chloro analogue of intermediate **3** ($IC_{50} = 1.65$ mM), related to bisindazolol **5** or 1-(4-chlorobutyl)indazolol **43** ($IC_{50} = 1.91$ mM), related to bisindazolol **35**] or are inactive [e. g. the "half-analogue" of bisindazolol **33**, 1-(2-picolyl)indazolol **44** ($IC_{50} > 5$ mM)].[10]

Table 1. Activity of hydrocarbylenebis(indazol-3-ols) **5** and **30**–**38** on the ferriprotoporphyrin IX biocrystallization inhibition test (FBIT).[a,b]

Compound	IC_{50} (μM)	Compound	IC_{50} (μM)
5	11 ± 4	**34**	> 1000
30	21 ± 6	**35**	–[c]
31	20 ± 5	**36**	22 ± 4
32	12 ± 3	**37**	14 ± 5
33	21 ± 4	**38**	> 1000

[a] Chloroquine difosfate: $IC_{50} = 28 \pm 6$ μM. [b] The assayed bis(3-alkoxyindazoles) (**15**, **16**, **19**, **20**–**24**, **27** and **28**) and bis(indazolin-3-ones) (**42**) are not active ($IC_{50} >$ 1000 μM). [c] Compound **35** could not be tested due to solubility issues.

The biological activity of these compounds may be derived from their interaction with ferriprotoporphyrin IX, preventing biocrystallization to hemozoin. It is possible that the interaction of bisindazololate bidentate ligands with the porphyrin iron hinders the interactions necessary for biocrystallization, i.e. the reciprocal bonds of Fe^{3+} with the propionate of an adjacent heme to yield dimers, which in turn form chains linked by hydrogen bonds in the crystal structure;[19] π-π interactions between indazole and porphyrin rings, similar to those postulated for chloroquine–heme complexes, must also be taken into consideration.[13,20]



1 R = H, Y = NO_2
43 R = $[CH_2]_4Cl$, Y = NO_2
44 R = 2-picolyl, Y = NO_2
45 R = H, Y = H
46 R = Me, Y = NO_2
47 R = Me, Y = H
48 R = Bn, Y = NO_2
49 R = Bn, Y = H

It is probably that derivative **34** is not an appropriate ligand since the ethylene linker chain is too short to provide an adequate distance between the two indazololate moieties. Additionally, it seems that an appropriate pK_a value is also very important for interaction of indazol-3-ols with heme. In fact, we suspected that the 5-nitroindazol-3-ol moiety present in the active compounds **5**, **30**--**33**, **36** and **37** was more acidic than the corresponding indazol-3-ol unit present only in the inactive compound **38**.[21] In order to clarify this point, we undertook, as reported for related indazolols,[15,22] a potentiometric determination of pK_a values for compounds **5** and **38** as well as those of the simple analogues **1** and **45**–**49**; owing to the very low solubility of the nitroindazolols and the bisindazolols **5** and **38** in water, measurements in a dimethyl sulfoxide/water 2:1 (*v*/*v*) mixture were also carried out for comparative purposes. According to the results gathered in Table 2, pK_a values for 1-substituted 5-nitroindazol-3-ols in dimethyl sulfoxide/water are ~2 units lower than those of the corresponding 5-H analogues (pairs **5**/**38**, **46**/**47** and **48**/**49**, respectively). pK_a values of indazolols in water are ~1–2 units lower than those observed in the dimethyl sulfoxide/water mixture but, despite the incomplete results, it is evident that 5-NO_2 derivatives are

Table 2. pK_a values of compounds **1**, **5**, **38** and **45**–**49** measured by potentiometric titration at 25 °C.

Compound	pK_a (H_2O)	pK_a [$(CH_3)_2SO/H_2O$ 2:1 (*v*/*v*)]
1	6.07 ± 0.03	7.02 ± 0.02
5	–[a]	8.04 ± 0.03[b]
38	–[a]	10.03 ± 0.04[b]
45	8.21 ± 0.03[c]	10.27 ± 0.03
46	5.98 ± 0.05	7.98 ± 0.03
47	7.60 ± 0.03	9.85 ± 0.03
48	–[a]	7.85 ± 0.04
49	7.44 ± 0.03[d]	9.86 ± 0.02

[a] Compound insoluble in water, pK_a could not be measured by potentiometric titration in this solvent. [b] Only one inflection point observed in the titration curves, pK_a value given is the average of the expected and close pK_{a1} and pK_{a2} values. [c] Literature value; 8.04 ± 0.02,[15] 8.28. [22] [d] Literature values; 7.21.[22]

still more acidic than the corresponding 5-H analogues (ΔpK_a = -2.14 and -1.62 for the pairs **1**/**45** and **46**/**47**, respectively).

Theoretical and experimental studies, similar to those carried out on the complexes of antimalarial drugs or other ligands with porphyrins,[20,23] directed to better understanding of the interaction of the bisindazolols with ferriprotoporphyrin IX are planned for the near future.

Finally, compounds **5**, **32** and **37** were tested for in vivo antimalarial activity in mouse by the classical 4 day suppressive test against *Plasmodium berghei* (ANKA strain),[24] but none of them showed activity at the evaluated doses (100 and 50 mg kg^{-} day^{-}). The acidic properties ($pK_a \sim 6$) of the prepared bis(5-nitroindazol-3-ols), suggest that they are ionized at physiological pH, leading to poor pharmacokinetic properties in mice, and potentially preventing the passage of these molecules through the four membranes that lock up the "active site", i.e., the digestive vacuole of intraerythrocytic parasites. The synthesis of some 3-*O*,3′-*O*-diprotected 1,1′-hydrocarbylenebis(indazol-3-ols), neutral (nonionizable) and labile prodrugs of the active compounds, is being carried out in order to circumvent this problem.

Conclusions

According to a FBIT method, 1,1′-hydrocarbylenebis(indazol-3-ols) are good inhibitors of biocrystallization of heme to hemozoin. A 5-NO_2 group, imparting an appropriate pK_a value (~6) to the indazolol moieties, seems essential for activity. On the other hand, the activities of most 5-nitro derivatives are similar or higher than that of chloroquine, showing that a broad range of aliphatic or aromatic chains are effective as linkers of the two indazolol moieties; only ethylene chain seems to be too short to allow an adequate interaction of the compound with heme. However, further work is needed to understand the mechanism of heme biocrystallization inhibition, and to convert these products into useful antimalarial drugs.

Experimental Section

General methods: Melting points were determined using a Stuart Scientific melting point apparatus SMP3. ^{1}H (300, 400 or 500 MHz) and ^{13}C (75, 100 or 125 MHz) NMR spectra were recorded on a Varian Unity 300, Bruker Avance 300, Varian Inova 400 or Varian System 500 spectrometer, the latter was equipped with a HCN cold probe. The chemical shifts are reported relative to TMS (δ scale), and corrected against the residual solvent signal [$(CD_3)_2SO$: δ_H = 2.49 ppm, δ_C = 39.50 ppm; C_5D_5N ([D_5]pyridine): δ_H = 8.71 ppm (2- and 6-H), δ_C = 149.90 ppm (C-2 and -6)]. *J* values are given in Hz. The assignments have been performed by means of different 1D and 2D standard experiments (NOE, gCOSY, gHSQC and gHMBC). Most NMR spectra were recorded at room temperature (~20 °C), however, owing to solubility problems, some spectra were recorded at elevated temperatures (60–80 °C) as indicated. Numbering used in the description of NMR spectra is given in Scheme 3. Electron impact (EI) and electrospray (ES+) mass spectra were obtained at 70 eV on a Hewlett Packard 5973 MSD spectrometer or on a Hewlett Packard 1100 MSD spectrometer, respectively. DC-Alufolien silica gel 60 PF_{254} (Merck, layer thickness 0.2 mm) was used for TLC, and silica gel 60 (Merck, particle size 0.040–0.063 mm) for flash column chromatography. Microanalyses were performed by the Departamento de Análisis, Centro de Química Orgánica "Manuel Lora Tamayo", CSIC, Madrid (Spain). Potentiometric titrations were carried out with a Metrohm 654 pH meter. Ferriprotoporphyrin IX biocrystallization inhibition test (FBIT)[11] and in vivo activity against *Plasmodium berghei* (ANKA strain) in mice[24] were carried out according to the respective literature procedures. The animal experiments were carried out in accordance with the guidelines published by the European Council (86/609ED), and controlled in Spain by Royal Decree (223/1988, March 14), on the protection of animals used for experimental and other scientific purposes. ^{1}H and ^{13}C NMR spectra of the prepared 1-substituted indazol-3-ols (**5**, **30**–**38**), 3-alkoxy-1-alkylindazoles (**15**–**29**) and 1,2-disubstituted indazolin-3-ones (**39**–**42**) are in agreement with the data previously reported for related compounds.[1,3,4] Owing to atropisomerism, protons of NCH_2 groups of biphenyl derivatives **18**, **23** and **32** are diastereotopic; they have been distinguished in the description of spectra as H_A and H_B.

Preparation of 1-ethoxycarbonyl-1*H*-indazol-3-ol (7): Ethyl chloroformate (5.53 g, 51 mmol) was slowly added to a suspension of 1*H*-indazol-3-ol (or 3-indazolinone) (6.71 g, 50 mmol) in pyridine (35 mL), and the reaction mixture was stirred for 30 min at RT. After addition of further ethyl chloroformate (~1 mL), the reaction was stirred for 30 min and then poured into water (400 mL) and stored in the refrigerator overnight. The precipitate was collected by filtration, washed with water (200 mL) and acetone (3×50 mL), and then air dried to give the title compound, which was used without further purification. Yield: 8.87 g (86%); mp: 195–197 °C (AcOH) (lit. 193 °C;[16] lit. 198–201 °C[25]).

Preparation of 3-benzyloxy-5-nitroindazole (8) and 2-benzyl-5-nitroindazolinone (11): A mixture of 1-ethoxycarbonyl-5-nitroindazolol **6**[3] (7.54 g, 30 mmol), K_2CO_3 (4.42 g, 32 mmol) and BnBr (5.47 g, 32 mmol) in acetone (500 mL) was refluxed while stirring for 24 h and then evaporated to dryness. KOH in EtOH (0.4 N, 90 mL) was added to the residue and the solution was stirred for 1 h at RT and then evaporated to dryness. After addition of water (250 mL) to the residue, the insoluble material was collected by filtration, washed with water and air dried; the resultant solid was a mixture (84/12/4 molar ratio; 80/15/5 weight ratio) of the 3-benzyloxy derivative **8** and the corresponding 1,2-dibenzylindazolinone and 1-benzyl-3-benzyloxyindazole. We have previously reported the isolation of pure compound **8** from this mixture by preparative TLC;[3] when larger quantities of compound are involved, column chromatography (chloroform/acetone, 20:1) or recrystallization (AcOH) is more convenient. In fact, the mixture can be used directly in the following step since the by-products do not react and are easily removed during the workup. On the other hand, after collection of crude compound **8**, the basic filtrate was extracted with chloroform (3×50 mL) and then acidified (pH 4) with AcOH; the precipitate was collected by filtration, washed with water and air dried to give 2-benzylindazolinone **11** without the need for further purification.

3-Benzyloxy-5-nitro-1*H*-indazole (8): Yield: 2.42 g (30%; recrystallization, two crops); 3.39 g (42%, by column chromatography; 0.59 g of 1,2-dibenzyl-5-nitroindazolinone[3] and 0.16 g of 1-benzyl-3-benzyloxy-5-nitroindazole[3] were also obtained); R_f = 0.22 (compound **8**), 0.35 (1,2-dibenzyl derivative), 0.70 (1-benzyl-3-benzyloxy derivative) (chloroform/acetone, 20:1); mp: 148–150 °C (AcOH) (lit. 148–150 °C.[3]).

2-Benzyl-5-nitro-1,2-dihydro-3*H*-indazol-3-one (11): Yield: 3.80 g (47%); mp: 209–211 °C (1-propanol) (lit. 210–212 °C[3]).

Preparation of 3-methoxy-5-nitroindazole (9) and 2-methyl-5-nitroindazolinone (12): Prepared as described for analogues **8** and **11**, starting from 1-ethoxycarbonyl-5-nitroindazolol (**6**)[3] (7.54 g, 30 mmol), K_2CO_3 or Cs_2CO_3 (32 mmol) and excess MeI (6 mL) in acetone (500 mL). The corresponding 3-methoxy derivative **9** was isolated by filtration, washed with water, etc., and used without the need for further purification. The basic filtrate was treated as before and afforded 2-methylindazolinone **12** as pure material.

3-Methoxy-5-nitro-1*H*-indazole (9): Yield: 2.14 g (37%) (using K_2CO_3), 2.84 g (49%) (using Cs_2CO_3); mp: 223–225 °C (1-propanol). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 12.70$ (1H, br s, NH), 8.49 (1H, d, $J = 2.2$, 4-H), 8.14 (1H, dd, $J = 9.3$, $J = 2.2$, 6-H), 7.51 (1H, d, $J = 9.3$, 7-H), 4.03 ppm (3H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 158.28$ (C-3), 143.19 (C-7a), 140.27 (C-5), 121.98 (C-6), 117.33 (C-4), 110.84 (C-7), 110.22 (C-3a), 56.30 ppm (CH_3); MS (EI): *m/z* (%): 193 (M^+, 100), 178 (11), 163 (13), 147 (19), 119 (23), 92 (14). Anal. calcd. for $C_8H_7N_3O_3$ (193.16): C, 49.74; H, 3.65; N, 21.75. Found: C, 49.86; H, 3.71; N, 21.59.

2-Methyl-5-nitro-1,2-dihydro-3*H*-indazol-3-one (12): Yield: 3.24 g (56%) (using K_2CO_3), 2.67 g (46%) (using Cs_2CO_3); mp: 264–267 °C (decomp) (1-propanol). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 12.04$ (1H, br s, NH), 8.45 (1H, d, $J = 2.2$, 4-H), 8.25 (1H, dd, $J = 9.2$, $J = 2.2$, 6-H), 7.39 (1H, d, $J = 9.2$, 7-H), 3.46 ppm (3H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 158.35$ (C-3), 145.53 (C-7a), 140.35 (C-5), 125.72 (C-6), 120.30 (C-4), 114.47 (C-3a), 111.87 (C-7), 30.58 ppm (CH_3); MS (EI): *m/z* (%): 193 (M^+, 100), 165 (8), 147 (27), 119 (9), 104 (8). Anal. calcd. for $C_8H_7N_3O_3$ (193.16): C, 49.74; H, 3.65; N, 21.75. Found: C, 49.50; H, 3.67; N, 21.62.

Preparation of 3-methoxyindazole (10) and 2-methylindazolinone (13): Prepared as described for analogues **8** and **11**, starting from 1-(ethoxycarbonyl)indazolol (**7**) (6.18 g, 30 mmol), K_2CO_3 (4.42 g, 32 mmol) and excess MeI (6 mL) in acetone (500 mL). After treatment with KOH and evaporation to dryness as described, water (20 mL) was added; the insoluble material was collected by filtration, washed with water (3×10 mL) and air dried to give pure 3-methoxyindazole (**10**). The basic filtrate was concentrated to 10 mL, acidified with AcOH (pH 5) and stored in the refrigerator overnight. The precipitate was collected by filtration, washed with cold 5% aq AcOH (2×5 mL) and air dried to give pure indazol-3-one (**13**); concentration of the filtrate afforded a second crop of crystals.

3-Methoxy-1*H*-indazole (10): Yield: 1.29 g (29%); mp: 103–104 °C (2-propanol/water) (lit. 104–105 °C[15]).

2-Methyl-1,2-dihydro-3*H*-indazol-3-one (13): Yield: 3.02 g (68%); mp: 198–205 °C (decomp) (water) (lit. 192–200 °C (decomp);[15] lit. 207–214 °C[25]).

Preparation of bis(3-alkoxyindazole) derivatives 15–29: A mixture of the required 3-alkoxyindazole (**8**, **9** or **10**) (3.00 mmol), the corresponding dibromo derivative (1.52 mmol) and K_2CO_3 (0.80 g, excess) in acetone (40 mL) was refluxed for 3–4 h (for **15**–**24**) or 24 h (for **25**–**29**). The mixture was then evaporated to dryness and the residue triturated with EtOH (5 mL). In each case, the insoluble material was collected by filtration, washed with EtOH (2×5 mL) and water (3×50 mL) and air dried to give the desired derivative. In the preparation of compound **25**, the EtOH extract of the crude reaction material was evaporated to dryness, and the obtained mixture (153 mg) was separated by flash column chromatography; elution with chloroform afforded 3-methoxy-5-nitro-1-vinyl-1*H*-indazole (82 mg, 12%) and further elution with a chloroform/MeOH mixture (50:1) gave the unreacted starting 3-methoxyindazole **9** (56 mg, 10%).

1,1′-(*o*-Xylylene)bis(3-benzyloxy-5-nitro-1*H*-indazole) (15): Yield: 0.89 g (93%); mp: 176–177 °C (2-butanone). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.55$ (2H, d, $J = 2.1$, 4- and 4′-H), 8.20 (2H, dd, $J = 9.3$, $J = 2.1$, 6- and 6′-H), 7.74 (2H, d, $J = 9.3$, 7- and 7′-H), 7.48 (4H, m, Bn 2- and 6-H), 7.33 (6H, m, Bn 3-, 4- and 5-H), 7.18 (2H, m, 4″- and 5″-H), 6.84 (2H, m, 3″- and 6″-H), 5.81 (4H, s, NCH_2), 5.38 ppm (4H, s, OCH_2); ^{13}C NMR [125 MHz, $(CD_3)_2SO$]: $\delta = 156.92$ (C-3 and -3′), 142.83 (C-7a and -7′a), 140.71 (C-5 and -5′), 136.16 (Bn C-1), 134.86 (C-1″ and -2″), 128.43 (Bn C-3 and -5), 128.22 (two overlapped signals; Bn C-2, -4 and -6), 128.06 (C-4″ and -5″), 127.86 (C-3″ and 6″), 122.57 (C-6 and -6′), 117.76 (C-4 and -4′), 111.24 (C-3a and -3′a), 110.50 (C-7 and -7′), 70.65 (OCH_2), 49.48 ppm (NCH_2); MS (EI): *m/z* (%): 549 ([*M*−Bn]$^+$, 18), 370 (9), 280 (100), 264 (5), 234 (13), 91 (87). Anal. calcd. for $C_{36}H_{28}N_6O_6$ (640.64): C, 67.49; H, 4.41; N, 13.12. Found: C, 67.23; H, 4.59; N, 12.95.

1,1′-(*m*-Xylylene)bis(3-benzyloxy-5-nitro-1*H*-indazole) (16): Yield: 0.90 g (94%); mp: 140–142 °C (ethyl acetate). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.40$ (2H, d, $J = 2.1$, 4- and 4′-H), 8.07 (2H, dd, $J = 9.3$, $J = 2.1$, 6- and 6′-H), 7.61 (2H, d, $J = 9.3$, 7- and 7′-H), 7.47 (4H, m, Bn 2- and 6-H), 7.36 (6H, m, Bn 3-, 4- and 5-H), 7.30 (1H, t, $J = 7.1$, 5″-H), 7.18 (2H, d, $J = 7.1$, 4″- and 6″-H), 6.52 (1H, s, 2″-H), 5.50 (4H, s, NCH_2), 5.29 ppm (4H, s, OCH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.78$ (C-3 and -3′), 142.60 (C-7a and -7′a), 140.42 (C-5 and -5′), 137.43 (C-1″ and -3″), 136.04 (Bn C-1), 128.87 (C-5″), 128.36 (Bn C-3 and -5), 128.14 (Bn C-4), 128.04 (Bn C-2 and -6), 126.37 (C-4″ and -6″), 124.77 (C-2″), 122.16 (C-6 and -6′), 117.37 (C-4 and -4′), 110.95 (C-3a and -3′a), 110.14 (C-7 and -7′), 70.51 (OCH_2), 51.43 ppm (NCH_2); MS (ES+): *m/z* (%): 1303 ([2*M*+Na]$^+$, 8), 663 ([*M*+Na]$^+$, 75), 641 ([*M*+H]$^+$, 100). Anal. calcd. for $C_{36}H_{28}N_6O_6$ (640.64): C, 67.49; H, 4.41; N, 13.12. Found: C, 67.22; H, 4.17; N, 13.22.

1,1′-(*p*-Xylylene)bis(3-benzyloxy-5-nitro-1*H*-indazole) (17): Yield: 0.88 g (92%); mp: 211–213 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.53$ (2H, d, $J = 2.2$, 4- and 4′-H), 8.19 (2H, dd, $J = 9.5$, $J = 2.2$, 6- and 6′-H), 7.80 (2H, d, $J = 9.5$, 7- and 7′-H), 7.50 (4H, m, Bn 2- and 6-H), 7.32 (6H, m, Bn 3-, 4- and 5-H), 7.14 (4H, s, 2″-, 3″-, 5″- and 6″-H), 5.52 (4H, s, NCH_2), 5.40 ppm (4H, s, OCH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.83$ (C-3 and -3′), 142.58 (C-7a and -7′a), 140.63 (C-5 and -5′), 136.33 and 136.18 (C-1″ and -4″, and Bn C-1), 128.32 (Bn C-3 and -5), 128.17 (Bn C-2 and -6), 128.06 (Bn C-4), 127.56 (C-2″, -3″, -5″ and -6″), 122.40 (C-6 and -6′), 117.61 (C-4 and -4′), 111.26 (C-3a and -3′a), 110.42 (C-7 and -7′), 70.57 (OCH_2), 51.38 ppm (NCH_2); MS (ES+): *m/z* (%): 663 ([*M*+Na]$^+$, 25), 641 ([*M*H]$^+$, 75), 105 (100). Anal. calcd. for $C_{36}H_{28}N_6O_6$ (640.64): C, 67.49; H, 4.41; N, 13.12. Found: C, 67.57; H, 4.36; N, 13.29.

1,1′-[(2,2′-Biphenyldiyl)bismethylene]bis(3-benzyloxy-5-nitro-1*H*-indazole) (18): Yield: 0.95 g (88%); mp: 193–195 °C (2-butanone). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.45$ (2H, d, $J = 2.0$, 4- and 4′-H), 8.02 (2H, dd, $J = 9.3$, $J = 2.0$, 6- and 6′-H), 7.40 (4H, m, Bn 2- and 6-H), 7.28 (10H, m, 4″-, 5″-, 4‴- and 5‴-H, and Bn 3-, 4- and 5-H), 7.15 (2H, d, $J = 9.3$, 7- and 7′-H), 7.09 (4H, m, 3″-, 6″-, 3‴and 6‴-H), 5.28 (4H, s, OCH_2), 5.26 (2H, d, $J = -15.9$, NCH_A), 5.14 ppm (2H, d, $J = -15.9$, NCH_B); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.81$ (C-3 and -3′), 142.59 (C-7a and -7′a), 140.34 (C-5 and -5′), 138.80 (C-2″ and

-2‴), 136.04 (Bn C-1), 134.22 (C-1″ and -1‴), 129.89 and 128.66 (C-3″, -6″, -3‴ and -6‴), 128.32 (two overlapped signals; Bn C-2, -3, -5 and -6), 128.16 (Bn C-4), 128.03 and 127.82 (C-4″, -5″, -4‴ and -5‴), 122.09 (C-6 and -6′), 117.45 (C-4 and -4′), 110.85 (C-3a and -3′a), 109.80 (C-7 and -7′), 70.51 (OCH_2), 49.92 ppm (NCH_2); MS (ES+): *m/z* (%): 739 ([*M*+Na]$^+$, 34), 717 ([*M*+H]$^+$, 100). Anal. calcd. for $C_{42}H_{32}N_6O_6$ (716.74): C, 70.38; H, 4.50; N, 11.73. Found: C, 70.15; H, 4.67; N, 11.62.

1,1′-[(2,6-Pyridinediyl)bismethylene]bis(3-benzyloxy-5-nitro-1*H*-indazole) (19): Yield: 0.71 g (74%); mp: 142–144 °C (EtOH). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.32 (2H, d, J = 2.1, 4- and 4′-H), 7.94 (2H, dd, J = 9.3, J = 2.1, 6- and 6′-H), 7.77 (1H, t, J = 7.8, 4″-H), 7.47 (4H, m, Bn 2- and 6-H), 7.36 (8H, m, 7- and 7′-H, and Bn 3-, 4- and 5-H), 7.19 (2H, d, J = 7.8, 3″- and 5″-H), 5.54 (4H, s, NCH_2), 5.27 ppm (4H, s, OCH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: δ = 156.64 (C-3 and -3′), 155.75 (C-2″ and -6″), 143.17 (C-7a and -7′a), 140.15 (C-5 and -5′), 137.99 (C-4″), 136.14 (Bn C-1), 128.39 (Bn C-3 and -5), 128.15 (Bn C-4), 128.09 (Bn C-2 and -6), 121.77 (C-6 and -6′), 120.65 (C-3″ and -5″), 117.17 (C-4 and -4′), 110.71 (C-3a and -3′a), 110.32 (C-7 and -7′), 70.43 (OCH_2), 53.03 ppm (NCH_2); MS (ES+): *m/z* (%): 664 ([*M*+Na]$^+$, 20), 642 ([*M*+H]$^+$, 100). Anal. calcd. for $C_{35}H_{27}N_7O_6$ (641.63): C, 65.52; H, 4.24; N, 15.28. Found: C, 65.59; H, 4.11; N, 15.02.

1,1′-(*o*-Xylylene)bis(3-methoxy-5-nitro-1*H*-indazole) (20): Yield: 0.70 g (96%); mp: 224–225 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.51 (2H, d, J = 2.2, 4- and 4′-H), 8.20 (2H, dd, J = 9.4, J = 2.2, 6- and 6′-H), 7.72 (2H, d, J = 9.4, 7- and 7′-H), 7.18 (2H, m, 4″- and 5″-H), 6.80 (2H, m, 3″- and 6″-H), 5.82 (4H, s, CH_2), 4.03 ppm (6H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: δ = 157.78 (C-3 and -3′), 142.94 (C-7a and -7′a), 140.66 (C-5 and -5′), 134.75 (C-1″ and -2″), 128.02 (C-4″ and -5″), 127.73 (C-3″ and -6″), 122.51 (C-6 and -6′), 117.65 (C-4 and -4′), 111.05 (C-3a and -3′a), 110.38 (C-7 and -7′), 56.61 (CH_3), 49.43 ppm (CH_2); MS (EI): *m/z* (%): 473 ([*M*−Me]$^+$, 27), 294 (43), 280 (100), 264 (6), 248 (28), 234 (30), 218 (9), 206 (8), 104 (15). Anal. calcd. for $C_{24}H_{20}N_6O_6$ (488.45): C, 59.01; H, 4.13; N, 17.21. Found: C, 59.30; H, 4.22; N, 16.95.

1,1′-(*m*-Xylylene)bis(3-methoxy-5-nitro-1*H*-indazole) (21): Yield: 0.69 g (94%); mp: 209–210 °C (2-butanone). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.38 (2H, d, J = 2.1, 4- and 4′-H), 8.10 (2H, dd, J = 9.3, J = 2.1, 6- and 6′-H), 7.53 (2H, d, J = 9.3, 7- and 7′-H), 7.33 (1H, t, J = 7.3, 5″-H), 7.20 (2H, d, J = 7.3, 4″- and 6″-H), 5.94 (1H, s, 2″-H), 5.48 (4H, s, CH_2), 3.86 ppm (6H, s, CH_3); ^{13}C NMR [100 MHz, $(CD_3)_2SO$]: δ = 157.45 (C-3 and -3′), 142.83 (C-7a and -7′a), 140.39 (C-5 and -5′), 137.60 (C-1″ and -3″), 128.86 (C-5″), 125.95 (C-4″ and -6″), 123.26 (C-2″), 122.16 (C-6 and -6′), 117.42 (C-4 and -4′), 110.62 (C-3a and -3′a), 110.17 (C-7 and -7′), 56.39 (CH_3), 51.23 ppm (CH_2); MS (EI): *m/z* (%): 488 ([*M*]$^+$, 100), 458 (5), 427 (5), 311 (7), 296 (73), 280 (21), 249 (28), 104 (39). Anal. calcd. for $C_{24}H_{20}N_6O_6$ (488.45): C, 59.01; H, 4.13; N, 17.21. Found: C, 59.23; H, 4.41; N, 17.43.

1,1′-(*p*-Xylylene)bis(3-methoxy-5-nitro-1*H*-indazole) (22): Yield: 0.69 g (94%); mp: 275–277 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.49 (2H, d, J = 2.0, 4- and 4′-H), 8.17 (2H, dd, J = 9.4, J = 2.0, 6- and 6′-H), 7.73 (2H, d, J = 9.4, 7- and 7′-H), 7.18 (4H, s, 2″-, 3″-, 5″- and 6″-H), 5.49 (4H, s, CH_2), 4.02 ppm (6H, s, CH_3); ^{13}C NMR (125 MHz, C_5D_5N): δ = 158.62 (C-3 and -3′), 143.19 (C-7a and -7′a), 141.33 (C-5 and -5′), 137.10 (C-1″ and -4″), 128.21 (C-2″, -3″, -5″ and -6″), 122.78 (C-6 and -6′), 118.59 (C-4 and -4′), 112.29 (C-3a and -3′a), 109.75 (C-7 and -7′), 56.63 (CH_3), 52.32 ppm (CH_2); MS (EI): *m/z* (%): 488 ([M]$^+$, 100), 458 (6), 296 (90), 281 (100), 250 (33), 235 (21), 179 (6), 118 (5), 104 (78). Anal. calcd. for $C_{24}H_{20}N_6O_6$ (488.45): C, 59.01; H, 4.13; N, 17.21. Found: C, 59.17; H, 3.95; N, 17.39.

1,1′-[(2,2′-Biphenyldiyl)bismethylene]bis(3-methoxy-5-nitro-1*H*-indazole) (23): Yield: 0.79 g (93%); mp: 162–164 °C (2-butanone). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.40 (2H, d, J = 2.2, 4- and 4′-H), 8.00 (2H, dd, J = 9.3, J = 2.2, 6- and 6′-H), 7.30 (4H, m, 4″-, 5″-, 4‴- and 5‴-H), 7.12 (4H, m, 3″-, 6″-, 3‴and 6‴-H), 7.09 (2H, d, J = 9.3, 7- and 7′-H), 5.29 (2H, d, J = -15.7, NCH_A), 5.20 (2H, d, J = -15.7, NCH_B), 3.93 ppm (6H, s, CH_3); ^{13}C NMR [100 MHz, $(CD_3)_2SO$]: δ = 157.64 (C-3 and -3′), 142.61 (C-7a and -7′a), 140.24 (C-5 and -5′), 138.82 (C-2″ and -2‴), 134.24 (C-1″ and -1‴), 129.99 and 128.75 (C-3″, -6″, -3‴ and -6‴), 128.07 and 127.82 (C-4″, -5″, -4‴ and -5‴), 122.04 (C-6 and -6′), 117.38 (C-4 and -4′), 110.58 (C-3a and -3′a), 109.70 (C-7 and -7′), 56.47 (CH_3), 49.88 ppm (CH_2); MS (EI): *m/z* (%): 564 ([*M*]$^+$, 4), 549 ([*M*−Me]$^+$, 80), 371 (24), 356 (100), 339 (10), 324 (10), 310 (28), 294 (8), 206 (19), 179 (68), 165 (27), 160 (19), 152 (4), 103 (4). Anal. calcd. for $C_{30}H_{24}N_6O_6$ (564.55): C, 63.82; H, 4.28; N, 14.89. Found: C, 64.02; H, 4.27; N, 14.68.

1,1′-[(2,6-Pyridinediyl)bismethylene]bis(3-methoxy-5-nitro-1*H*-indazole) (24): Yield: 0.70 g (95%); mp: 164–166 °C (2-butanone). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.27 (2H, d, J = 2.0, 4- and 4′-H), 7.97 (2H, dd, J = 9.5, J = 2.0, 6- and 6′-H), 7.79 (1H, t, J = 7.8, 4″-H), 7.29 (2H, d, J = 9.5, 7- and 7′-H), 7.23 (2H, d, J = 7.8, 3″- and 5″-H), 5.51 (4H, s, CH_2), 3.84 ppm (6H, s, CH_3); ^{13}C NMR [100 MHz, $(CD_3)_2SO$]: δ = 157.16 (C-3 and -3′), 155.61 (C-2″ and -6″), 143.20 (C-7a and -7′a), 140.01 (C-5 and -5′), 137.85 (C-4″), 121.58 (C-6 and -6′), 120.50 (C-3″ and -5″), 117.19 (C-4 and -4′), 110.37 (C-3a and -3′a), 110.26 (C-7 and -7′), 56.28 (CH_3), 52.79 ppm (CH_2); MS (EI): *m/z* (%): 489 ([*M*]$^+$, 100), 459 (10), 428 (17), 312 (20), 251 (11), 220 (9), 206 (83), 193 (9), 160 (38), 105 (12). Anal. calcd. for $C_{23}H_{19}N_7O_6$ (489.44): C, 56.44; H, 3.91; N, 20.03. Found: C, 56.69; H, 3.67; N, 19.85.

1,1′-Ethylenebis(3-methoxy-5-nitro-1*H*-indazole) (25): Yield: 0.38 g (61%); mp: 247–250 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.28 (2H, d, J = 2.0, 4- and 4′-H), 8.00 (2H, dd, J = 9.3, J = 2.0, 6- and 6′-H), 7.26 (2H, d, J = 9.3, 7- and 7′-H), 4.69 (4H, s, 1″- and 2″-H), 3.92 ppm (6H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$, 80 °C]: δ = 157.54 (C-3 and -3′), 142.73 (C-7a and -7′a), 140.16 (C-5 and -5′), 121.28 (C-6 and -6′), 116.47 (C-4 and -4′), 110.74 (C-3a and -3′a), 109.22 (C-7 and -7′), 56.05 (CH_3), 47.18 ppm (C-1″ and -2″); MS (EI): *m/z* (%): 412 ([*M*]$^+$, 8), 219 (68), 206 (100), 193 (28), 160 (43), 131 (3), 117 (3), 103 (5). Anal. calcd. for $C_{18}H_{16}N_6O_6$ (412.36): C, 52.43; H, 3.91; N, 20.38. Found: C, 52.39; H, 3.94; N, 20.62.

1,1′-Tetramethylenebis(3-methoxy-5-nitro-1*H*-indazole) (26): Yield: 0.60 g (91%); mp: 221–223 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.47 (2H, d, J = 2.1, 4- and 4′-H), 8.16 (2H, dd, J = 9.3, J = 2.1, 6- and 6′-H), 7.69 (2H, d, J = 9.3, 7- and 7′-H), 4.30 (4H, br s, 1″- and 4″-H), 3.98 (6H, s, CH_3), 1.75 ppm (4H, br s, 2″- and 3″-H); ^{13}C NMR (75 MHz, C_5D_5N, 70 °C): δ = 158.55 (C-3 and -3′), 143.17 (C-7a and -7′a), 141.53 (C-5 and -5′), 122.47 (C-6 and -6′), 118.47 (C-4 and -4′), 112.09 (C-3a and -3′a), 109.40 (C-7 and -7′), 56.63 (CH_3), 48.51 (C-1″ and -4″), 26.97 ppm (C-2″ and -3″); MS (EI): *m/z* (%): 440 ([*M*]$^+$, 2), 425 ([*M*−Me]$^+$, 47), 246 (2), 232 (19), 221 (12), 220 (10), 219 (12), 206 (100), 190 (10), 174 (7), 160 (41), 123 (10), 111 (10), 109 (10), 97 (11). Anal. calcd. for $C_{20}H_{20}N_6O_6$ (440.41): C, 54.54; H, 4.58; N, 19.08. Found: C, 54.50; H, 4.67; N, 18.82.

1,1′-Pentamethylenebis(3-methoxy-5-nitro-1*H*-indazole) (27): Yield: 0.63 g (92%); mp: 221–223 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: δ = 8.41 (2H, d, J = 2.1, 4- and 4′-H), 8.08 (2H,

dd, $J=9.3$, $J=2.1$, 6- and 6′-H), 7.57 (2H, d, $J=9.3$, 7- and 7′-H), 4.22 (4H, t, $J=6.6$, 1″- and 5″-H), 3.99 (6H, s, CH_3), 1.80 (4H, m, 2″- and 4″-H), 1.15 ppm (2H, m, 3″-H); ^{13}C NMR (75 MHz, C_5D_5N, 80 °C): $\delta=158.53$ (C-3 and -3′), 143.18 (C-7a and -7′a), 141.54 (C-5 and -5′), 122.42 (C-6 and -6′), 118.42 (C-4 and -4′), 112.07 (C-3a and -3′a), 109.34 (C-7 and -7′), 56.63 (CH_3), 48.81 (C-1″ and -5″), 29.20 (C-2″ and -4″), 24.35 ppm (C-3″); MS (EI): *m/z* (%): 454 ([*M*]$^+$, 7), 439 ([*M*−Me]$^+$, 48), 260 (3), 246 (10), 232 (4), 230 (5), 206 (100), 193 (5), 190 (4), 160 (33), 103 (4). Anal. calcd. for $C_{21}H_{22}N_6O_6$ (454.44): C, 55.50; H, 4.88; N, 18.49. Found: C, 55.60; H, 4.61; N, 18.65.

1,1′-Hexamethylenebis(3-methoxy-5-nitro-1*H*-indazole) (28): Yield: 0.67 g (95%); mp: 227–229 °C (nitromethane). ^{1}H NMR [500 MHz, $(CD_3)_2SO$]: $\delta=8.42$ (2H, d, $J=2.0$, 4- and 4′-H), 8.13 (2H, dd, $J=9.3$, $J=2.0$, 6- and 6′-H), 7.62 (2H, d, $J=9.3$, 7- and 7′-H), 4.21 (4H, t, $J=6.3$, 1″- and 6″-H), 3.97 (6H, s, CH_3), 1.71 (4H, br s, 2″- and 5″-H), 1.16 ppm (4H, br s, 3″- and 4″-H); ^{13}C NMR [125 MHz, $(CD_3)_2SO$]: $\delta=157.36$ (C-3 and -3′), 142.42 (C-7a and -7′a), 140.14 (C-5 and -5′), 122.00 (C-6 and -6′), 117.61 (C-4 and -4′), 110.30 (C-3a and -3′a), 110.06 (C-7 and -7′), 56.49 (CH_3), 48.05 (C-1″ and -6″), 28.88 (C-2″ and -5″), 25.70 ppm (C-3″ and -4″); MS (EI): *m/z* (%): 468 ([*M*]$^+$, 1), 453 ([*M*−Me]$^+$, 5), 206 (100), 192 (5), 176 (4), 160 (23), 103 (3); MS (ES+): *m/z* (%): 491 ([*M*+Na]$^+$, 73), 469 ([*M*H]$^+$, 100). Anal. calcd. for $C_{22}H_{24}N_6O_6$ (468.46): C, 56.40; H, 5.16; N, 17.94. Found: C, 56.51; H, 5.39; N, 17.69.

1,1′-(*o*-Xylylene)bis(3-methoxy-1*H*-indazole) (29): Yield: 0.51 g (85%); mp: 118–119 °C (1-propanol). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta=7.63$ (2H, d, $J=7.8$, 4- and 4′-H), 7.51 (2H, d, $J=8.5$, 7- and 7′-H), 7.37 (2H, dd, $J=8.5$, $J=7.0$, 6- and 6′-H), 7.11 (2H, m, 4″- and 5″-H), 7.06 (2H, dd, $J=7.8$, $J=7.0$, 5- and 5′-H), 6.70 (2H, m, 3″- and 6″-H), 5.71 (4H, s, CH_2), 4.00 ppm (6H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta=155.77$ (C-3 and -3′), 141.62 (C-7a and -7′a), 135.61 (C-1″ and -2″), 127.53 (C-6 and -6′), 127.39 (C-4″ and -5″), 127.27 (C-3″ and -6″), 119.49 (C-4 and -4′), 119.35 (C-5 and -5′), 111.65 (C-3a and -3′a), 109.50 (C-7 and -7′), 55.97 (CH_3), 48.82 ppm (CH_2); MS (EI): *m/z* (%): 398 ([*M*]$^+$, 1), 383 ([*M*−Me]$^+$, 44), 366 (5), 249 (60), 235 (100), 219 (15), 104 (14). Anal. calcd. for $C_{24}H_{22}N_4O_2$ (398.46): C, 72.34; H, 5.57; N, 14.06. Found: C, 72.43; H, 5.71; N, 14.15.

3-Methoxy-5-nitro-1-vinyl-1*H*-indazole: Mp: 127–129 °C (EtOH). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta=8.45$ (1H, d, $J=2.2$, 4-H), 8.25 (1H, dd, $J=9.3$, $J=2.2$, 6-H), 7.92 (1H, d, $J=9.3$, 7-H), 7.62 (1H, dd, $J=14.9$, $J=8.8$, 1′-H), 5.47 (1H, d, $J=14.9$, 2′-H_{trans}), 4.83 (1H, d, $J=8.8$, 2′-H_{cis}), 4.10 ppm (3H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta=158.85$ (C-3), 141.34 and 141.16 (C-5 and -7a), 129.41 (C-1′), 123.18 (C-6), 117.17 (C-4), 112.30 (C-3a), 110.33 (C-7), 97.48 (C-2′), 56.68 ppm (CH_3); MS (EI): *m/z* (%): 219 ([*M*]$^+$, 100), 204 (21), 189 (7), 173 (23), 158 (7), 116 (7), 102 (10). Anal. calcd. for $C_{10}H_9N_3O_3$ (219.20): C, 54.79; H, 4.14; N, 19.17. Found: C, 54.60; H, 3.97; N, 18.93.

Preparation of bis(indazol-3-ol) derivatives 5 and 30–38: A suspension of the required 3-alkoxyindazole (1.00 mmol) in 48% aq HBr (10 mL) was refluxed for 2–3 h (from **15**–**19**) or 1 h (from **20**–**29**). The reaction was cooled, and water was added (30 mL). The precipitate was collected by filtration, washed with water (3× 20 mL) and EtOH (3×10 mL) and air dried to give the desired indazolol. The crystalline solids obtained through recrystallization (DMF or DMF/water) retained DMF, however, in most cases residual DMF could be removed after several days of heating under vacuum at 120 °C. For compound **34**, the DMF could not be completely removed, a solvated crystal structure corresponding to **34**×0.25 DMF was obtained.

1,1′-(*o*-Xylylene)bis(5-nitro-1*H*-indazol-3-ol) (5): Yield: 428 mg (93%) (from **15**), 451 mg (98%) (from **20**); mp: 308–311 °C (decomp) (DMF/water) [lit. 302–305 °C (decomp)[2]]. ^{1}H NMR: ref.,[2] ^{13}C NMR [100 MHz, $(CD_3)_2SO$]: $\delta=156.71$ (C-3 and -3′), 142.35 (C-7a and -7′a), 140.17 (C-5 and -5′), 135.02 (C-1″ and -2″), 127.94 (C-3″, -4″, -5″ and -6″), 122.00 (C-6 and -6′), 118.69 (C-4 and -4′), 111.76 (C-3a and -3′a), 110.12 (C-7 and -7′), 49.09 ppm (CH_2); MS (EI): *m/z* (%): 460 ([*M*]$^+$, 5), 443 (4), 426 (4), 280 (100), 264 (9), 249 (17), 234 (40), 205 (20), 179 (29), 149 (17), 104 (28), 91 (20), 78 (18).

1,1′-(*m*-Xylylene)bis(5-nitro-1*H*-indazol-3-ol) (30): Yield: 419 mg (91%) (from **16**), 447 mg (97%) (from **21**); mp: 325–328 °C (decomp) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta=11.43$ (2H, br s, OH), 8.60 (2H, d, $J=2.1$, 4- and 4′-H), 8.08 (2H, dd, $J=9.3$, $J=2.1$, 6- and 6′-H), 7.61 (2H, d, $J=9.3$, 7- and 7′-H), 7.27 (1H, t, $J=7.6$, 5″-H), 7.12 (2H, d, $J=7.6$, 4″- and 6″-H), 6.83 (1H, s, 2″-H), 5.41 ppm (4H, s, CH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta=156.56$ (C-3 and -3′), 142.19 (C-7a and -7′a), 139.97 (C-5 and -5′), 137.56 (C-1″ and -3″), 128.84 (C-5″), 126.58 (C-4″ and -6″), 125.75 (C-2″), 121.76 (C-6 and -6′), 118.49 (C-4 and -4′), 111.64 (C-3a and -3′a), 109.92 (C-7 and -7′), 51.30 ppm (CH_2); MS (EI): *m/z* (%): 460 ([*M*]$^+$, 39), 444 (4), 430 (9), 296 (13), 282 (100), 236 (51), 179 (23), 148 (23), 133 (11), 119 (24), 104 (55), 91 (13), 78 (24); MS (ES+): *m/z* (%): 483 ([*M*+Na]$^+$, 57), 461 ([*M*+H]$^+$, 100). Anal. calcd. for $C_{22}H_{16}N_6O_6$ (460.40): C, 57.39; H, 3.50; N, 18.25. Found: C, 57.30; H, 3.67; N, 17.99.

1,1′-(*p*-Xylylene)bis(5-nitro-1*H*-indazol-3-ol) (31): Yield: 437 mg (95%) (from **17**), 451 mg (98%) (from **22**); mp: > 340 °C (darkening is observed from ~320 °C but a clear melting or decomposition point cannot be determined) (DMF). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta=11.50$ (2H, br s, OH), 8.62 (2H, d, $J=2.0$, 4- and 4′-H), 8.13 (2H, dd, $J=9.5$, $J=2.0$, 6- and 6′-H), 7.73 (2H, d, $J=9.5$, 7- and 7′-H), 7.17 (4H, s, 2″-, 3″-, 5″- and 6″-H), 5.42 ppm (4H, s, CH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta=156.58$ (C-3 and -3′), 142.12 (C-7a and -7′a), 140.04 (C-5 and -5′), 136.50 (C-1″ and -4″), 127.76 (C-2″, -3″, -5″ and -6″), 121.88 (C-6 and -6′), 118.57 (C-4 and -4′), 111.72 (C-3a and -3′a), 110.08 (C-7 and -7′), 51.20 ppm (CH_2); MS (EI): *m/z* (%): 460 ([*M*]$^+$, 14), 297 (15), 282 (72), 265 (14), 236 (15), 179 (35), 149 (29), 133 (19), 119 (30), 104 (63), 84 (72), 66 (100). Anal. calcd. for $C_{22}H_{16}N_6O_6$ (460.40): C, 57.39; H, 3.50; N, 18.25. Found: C, 57.41; H, 3.73; N, 18.27.

1,1′-[(2,2′-Biphenyldiyl)bismethylene]bis(5-nitro-1*H*-indazol-3-ol) (32): Yield: 526 mg (98%) (from **18**), 520 mg (97%) (from **23**); mp: 308–311 °C (decomp) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta=11.50$ (2H, s, OH), 8.60 (2H, d, $J=2.2$, 4- and 4′-H), 7.99 (2H, dd, $J=9.3$, $J=2.2$, 6- and 6′-H), 7.20 (4H, m, 4‴-, 5‴-, 4⁗- and 5⁗-H), 7.16 (2H, d, $J=9.3$, 7- and 7′-H), 7.02 (4H, m, 3‴-, 6‴-, 3⁗- and 6⁗-H), 5.21 (2H, d, $J=-15.6$, NCH_A), 5.11 ppm (2H, d, $J=-15.6$, NCH_B); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta=156.70$ (C-3 and -3′), 142.30 (C-7a and -7′a), 139.84 (C-5 and -5′), 138.75 (C-2″ and -2‴), 134.33 (C-1″ and -1‴), 129.59 and 128.63 (C-3″, -6″, -3‴ and -6‴), 127.86 and 127.56 (C-4″, -5″, -4‴ and -5‴), 121.55 (C-6 and -6′), 118.32 (C-4 and -4′), 111.27 (C-3a and -3′a), 109.45 (C-7 and -7′), 49.50 ppm (CH_2); MS (EI): *m/z* (%): 536 ([*M*]$^+$, 6), 519 (8), 356 (23), 341 (18), 310 (8), 235 (22), 192 (22), 179 (100), 165 (62), 146 (17), 133 (7), 105 (8), 91 (8), 76 (9). Anal. calcd. for $C_{28}H_{20}N_6O_6$ (536.50): C, 62.68; H, 3.76; N, 15.66. Found: C, 62.57; H, 3.67; N, 15.62.

1,1′-[(2,6-Pyridinediyl)bismethylene]bis(5-nitro-1*H*-indazol-3-ol) (33): Yield: 447 mg (97%) (from **19** or **24**); mp: 286–288 °C

(decomp) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 11.40$ (2H, br s, OH), 8.57 (2H, d, $J = 2.1$, 4- and 4′-H), 8.00 (2H, dd, $J = 9.3$, $J = 2.1$, 6- and 6′-H), 7.72 (1H, t, $J = 7.6$, 4″-H), 7.48 (2H, d, $J = 9.3$, 7- and 7′-H), 7.06 (2H, d, $J = 7.6$, 3″- and 5″-H), 5.46 ppm (4H, s, CH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.56$ (C-3 and -3′), 156.11 (C-2″ and -6″), 142.69 (C-7a and -7′a), 139.88 (C-5 and -5′), 138.06 (C-4″), 121.55 (C-6 and -6′), 120.77 (C-3″ and -5″), 118.32 (C-4 and -4′), 111.58 (C-3a and -3′a), 110.17 (C-7 and -7′), 53.17 ppm (CH_2); MS (EI): *m/z* (%): 461 ([*M*]$^+$, 68), 298 (26), 284 (83), 266 (13), 252 (14), 236 (26), 219 (16), 207 (9), 192 (27), 179 (100), 164 (35), 149 (68), 133 (44), 121 (73), 106 (85), 91 (48), 78 (54). Anal. calcd. for $C_{21}H_{15}N_7O_6$ (461.39): C, 54.67; H, 3.28; N, 21.25. Found: C, 54.53; H, 3.57; N, 21.51.

1,1′-Ethylenebis(5-nitro-1*H*-indazol-3-ol) (34): Yield: 373 mg (97%) (from **25**); mp: >340 °C (darkening is observed from ~300 °C but a clear melting or decomposition point cannot be determined) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 11.42$ (2H, br s, OH), 8.45 (2H, d, $J = 2.2$, 4- and 4′-H), 7.93 (0.25H, s, DMF CHO), 7.91 (2H, dd, $J = 9.3$, $J = 2.2$, 6- and 6′-H), 7.10 (2H, d, $J = 9.3$, 7- and 7′-H), 4.59 (4H, s, 1″- and 2″-H), 2.87 (0.75H, s) and 2.71 ppm (0.75H, s) (DMF CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 162.33$ (DMF CHO), 156.81 (C-3 and -3′), 142.39 (C-7a and -7′a), 139.70 (C-5 and -5′), 121.27 (C-6 and -6′), 118.21 (C-4 and -4′), 111.65 (C-3a and -3′a), 109.24 (C-7 and -7′), 47.61 (C-1″ and -2″), 35.79 and 30.77 ppm (DMF CH_3); MS (EI): *m/z* (%): 384 ([*M*]$^+$, 15), 354 (6), 206 (53), 192 (100), 179 (21), 162 (19), 146 (54), 133 (9), 118 (7), 105 (10), 91 (14), 75 (15). Anal. calcd. for $C_{16}H_{12}N_6O_6 \times 0.25$ DMF (402.58): C, 49.97; H, 3.44; N, 21.75. Found: C, 49.70; H, 3.57; N, 21.82.

1,1′-Tetramethylenebis(5-nitro-1*H*-indazol-3-ol) (35): Yield: 408 mg (99%) (from **26**); mp: >340 °C (darkening is observed from ~300 °C but a clear melting or decomposition point cannot be determined) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 11.41$ (2H, br s, OH), 8.62 (2H, d, $J = 2.2$, 4- and 4′-H), 8.10 (2H, dd, $J = 9.5$, $J = 2.2$, 6- and 6′-H), 7.63 (2H, d, $J = 9.5$, 7- and 7′-H), 4.23 (4H, br s, 1″- and 4″-H), 1.71 ppm (4H, br s, 2″- and 3″-H); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.23$ (C-3 and -3′), 141.85 (C-7a and -7′a), 139.75 (C-5 and -5′), 121.44 (C-6 and -6′), 118.46 (C-4 and -4′), 111.21 (C-3a and -3′a), 109.70 (C-7 and -7′), 47.30 (C-1″ and -4″), 26.24 ppm (C-2″ and -3″); MS (EI): *m/z* (%): 412 ([*M*]$^+$, 16), 396 (9), 395 (9), 382 (6), 322 (7), 278 (15), 248 (19), 232 (59), 220 (21), 203 (38), 192 (100), 179 (65), 162 (28), 146 (66), 131 (39), 119 (27), 105 (38), 91 (38), 75 (43). Anal. calcd. for $C_{18}H_{16}N_6O_6$ (412.36): C, 52.43; H, 3.91; N, 20.38. Found: C, 52.30; H, 3.76; N, 20.61.

1,1′-Pentamethylenebis(5-nitro-1*H*-indazol-3-ol) (36): Yield: 418 mg (98%) (from **27**); mp: 294–297 °C (decomp) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 11.33$ (2H, br s, OH), 8.58 (2H, d, $J = 2.1$, 4- and 4′-H), 8.03 (2H, dd, $J = 9.4$, $J = 2.1$, 6- and 6′-H), 7.52 (2H, d, $J = 9.4$, 7- and 7′-H), 4.15 (4H, t, $J = 6.6$, 1″- and 5″-H), 1.75 (4H, m, 2″- and 4″-H), 1.10 ppm (2H, m, 3″-H); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.32$ (C-3 and -3′), 141.95 (C-7a and -7′a), 139.64 (C-5 and -5′), 121.43 (C-6 and -6′), 118.54 (C-4 and -4′), 111.09 (C-3a and -3′a), 109.74 (C-7 and -7′), 47.63 (C-1″ and -5″), 28.76 (C-2″ and -4″), 23.31 ppm (C-3″); MS (EI): *m/z* (%): 426 ([*M*]$^+$, 24), 409 (6), 396 (8), 246 (38), 132 (14), 218 (11), 203 (10), 192 (100), 179 (14), 162 (19), 146 (59), 134 (7), 119 (7), 118 (7), 105 (7), 91 (12), 76 (12). Anal. calcd. for $C_{19}H_{18}N_6O_6$ (426.38): C, 53.52; H, 4.26; N, 19.71. Found: C, 53.51; H, 4.32; N, 19.50.

1,1′-Hexamethylenebis(5-nitro-1*H*-indazol-3-ol) (37): Yield: 427 mg (97%) (from **28**); mp: 303–306 °C (decomp) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 11.39$ (2H, br s, OH), 8.62 (2H, d, $J = 2.1$, 4- and 4′-H), 8.09 (2H, dd, $J = 9.3$, $J = 2.1$, 6- and 6′-H), 7.57 (2H, d, $J = 9.3$, 7- and 7′-H), 4.16 (4H, t, $J = 6.6$, 1″- and 6″-H), 1.70 (4H, br m, 2″- and 5″-H), 1.18 ppm (4H, br s, 3″- and 4″-H); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 156.35$ (C-3 and -3′), 141.92 (C-7a and -7′a), 139.70 (C-5 and -5′), 121.53 (C-6 and -6′), 118.65 (C-4 and -4′), 111.15 (C-3a and -3′a), 109.76 (C-7 and -7′), 47.67 (C-1″ and -6″), 28.88 (C-2″ and -5″), 25.59 ppm (C-3″ and -4″); MS (EI): *m/z* (%): 440 ([*M*]$^+$, 18), 410 (6), 278 (13), 260 (17), 248 (12), 232 (30), 226 (33), 206 (20), 192 (100), 179 (41), 162 (26), 149 (82), 146 (55), 131 (33), 121 (22), 105 (87), 91 (33), 73 (94). Anal. calcd. for $C_{20}H_{20}N_6O_6$ (440.41): C, 54.54; H, 4.58; N, 19.08. Found: C, 54.71; H, 4.67; N, 18.88.

1,1′-(*o*-Xylylene)bis(1*H*-indazol-3-ol) (38): Yield: 356 mg (96%) (from **29**), mp: 280–284 °C (decomp) (DMF/water). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 10.75$ (2H, br s, OH), 7.65 (2H, d, $J = 7.9$, 4- and 4′-H), 7.45 (2H, d, $J = 8.5$, 7- and 7′-H), 7.31 (2H, dd, $J = 8.5$, $J = 6.9$, 6- and 6′-H), 7.12 (2H, m, 4″- and 5″-H), 7.00 (2H, dd, $J = 7.9$, $J = 6.9$, 5- and 5′-H), 6.77 (2H, m, 3″- and 6″-H), 5.55 ppm (4H, s, CH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 154.48$ (C-3 and -3′), 141.30 (C-7a and -7′a), 135.94 (C-1″ and -2″), 127.84 (C-3″ and -6″), 127.44 (C-4″ and -5″), 127.16 (C-6 and -6′), 120.20 (C-4 and -4′), 118.85 (C-5 and -5′), 112.47 (C-3a and -3′a), 109.40 (C-7 and -7′), 48.73 ppm (CH_2); MS (EI): *m/z* (%): 370 ([*M*]$^+$, 12), 235 (100), 219 (8), 206 (10), 180 (5), 165 (7), 104 (15), 77 (13). Anal. calcd. for $C_{22}H_{18}N_4O_2$ (370.40): C, 71.34; H, 4.90; N, 15.13. Found: C, 71.53; H, 4.97; N, 15.02.

Preparation of bis(indazolin-3-one) derivatives 39–42: A mixture of the required 2-substituted indazolinone (**11** or **12**) (3.00 mmol), the corresponding dibromo derivative (1.54 mmol) and K_2CO_3 (0.80 g, excess) in acetone (20 mL) was refluxed until consumption of the starting indazolinone (~10–20 h). The reaction mixture was allowed to reach RT and the precipitate was collected by filtration, washed with acetone (3×5 mL) and water (3×50 mL), and air dried to afford the title derivatives.

1,1′-(*o*-Xylylene)bis(2-benzyl-5-nitro-1,2-dihydro-3*H*-indazol-3-one) (39): Yield: 884 mg (92%); mp: 234–236 °C (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.62$ (2H,d, $J = 2.2$, 4- and 4′-H), 8.39 (2H, dd, $J = 9.3$, $J = 2.2$, 6- and 6′-H), 7.35 (2H, d, $J = 9.3$, 7- and 7′-H), 7.18 (6H, m, Bn 3-, 4- and 5-H), 7.07 (2H, m, 4″- and 5″-H), 7.01 (4H, m, Bn 2- and 6-H), 6.44 (2H, m, 3″- and 6″-H), 5.30 (4H, s, 1″- and 2″-CH_2), 5.06 ppm (4H, s, Bn CH_2); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 160.55$ (C-3 and -3′), 148.49 (C-7a and -7′a), 141.48 (C-5 and -5′), 135.71 (Bn C-1), 132.97 (C-1″ and -2″), 128.62 (Bn C-3 and -5), 127.99 (C-4″ and -5″), 127.78 (Bn C-4), 127.61 (C-6 and -6′), 126.98 (Bn C-2 and -6), 126.20 (C-3″ and -6″), 120.85 (C-4 and -4′), 115.23 (C-3a and -3′a), 111.50 (C-7 and -7′), 47.77 (1″- and 2″-CH_2), 45.06 ppm (Bn CH_2); MS (ES+): *m/z* (%): 1281 ([2*M*+H]$^+$, 33), 641 ([*M*+H]$^+$, 100). Anal. calcd. for $C_{36}H_{28}N_6O_6$ (640.64): C, 67.49; H, 4.41; N, 13.12. Found: C, 67.67; H, 4.23; N, 13.01.

1,1′-(*o*-Xylylene)bis(2-methyl-5-nitro-1,2-dihydro-3*H*-indazol-3-one) (40): Yield: 535 mg (73%); mp: 284–287 °C (decomp) (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.56$ (2H, d, $J = 2.3$, 4- and 4′-H), 8.41 (2H, dd, $J = 9.3$, $J = 2.3$, 6- and 6′-H), 7.61 (2H, d, $J = 9.3$, 7- and 7′-H), 7.15 (2H, m, 4″- and 5″-H), 6.54 (2H, m, 3″- and 6″-H), 5.65 (4H, s, CH_2), 3.39 ppm (6H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 159.58$ (C-3 and -3′), 147.05 (C-7a and -7′a), 140.96 (C-5 and -5′), 133.34 (C-1″ and -2″), 128.10 (C-4″ and -5″), 127.18 (C-6 and -6′), 125.47 (C-3″ and -6″), 120.61 (C-4 and -4′), 114.80 (C-3a and -3′a), 110.88 (C-7 and -7′), 46.80 (CH_2), 29.16 ppm (CH_3); MS (ES+): *m/z* (%): 999 ([2*M*+Na]$^+$, 53), 511 ([*M*+Na]$^+$, 80), 489 ([*M*

H]$^+$, 100). Anal. calcd. for $C_{24}H_{20}N_6O_6$ (488.45): C, 59.01; H, 4.13; N, 17.21. Found: C, 59.29; H, 3.87; N, 17.42.

1,1′-(*m*-Xylylene)bis(2-methyl-5-nitro-1,2-dihydro-3*H*-indazol-3-one) (41): Yield: 601 mg (82%); mp: 285–288 °C (decomp) (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.35$ (2H, d, $J = 2.1$, 4- and 4′-H), 8.23 (2H, dd, $J = 9.3$, $J = 2.1$, 6- and 6′-H), 7.67 (2H, d, $J = 9.3$, 7- and 7′-H), 7.24 (1H, t, $J = 7.3$, 5″-H), 7.11 (2H, d, $J = 7.3$, 4″- and 6″-H), 6.59 (1H, s, 2″-H), 5.30 (4H, s, CH_2), 3.27 ppm (6H, s, CH_3); ^{13}C NMR [75 MHz, $(CD_3)_2SO$]: $\delta = 160.12$ (C-3 and -3′), 147.68 (C-7a and -7′a), 141.06 (C-5 and -5′), 135.22 (C-1″ and -3″), 129.14 (C-5″), 127.47 (C-4″ and -6″), 126.75 (C-6 and -6′), 125.48 (C-2″), 120.12 (C-4 and -4′), 115.46 (C-3a and -3′a), 111.63 (C-7 and -7′), 49.37 (CH_2), 29.09 ppm (CH_3); MS (ES+): *m/z* (%): 999 ([2*M*+Na]$^+$, 59), 511 ([*M*+Na]$^+$, 53), 489 ([*M*+H]$^+$, 100). Anal. calcd. for $C_{24}H_{20}N_6O_6$ (488.45): C, 59.01; H, 4.13; N, 17.21. Found: C, 58.77; H, 4.37; N, 17.50.

1,1′-(*p*-Xylylene)bis(2-methyl-5-nitro-1,2-dihydro-3*H*-indazol-3-one) (42): Yield: 593 mg (81%); mp: 295–298 °C (decomp) (nitromethane). ^{1}H NMR [300 MHz, $(CD_3)_2SO$]: $\delta = 8.40$ (2H, d, $J = 2.3$, 4- and 4′-H), 8.33 (2H, dd, $J = 9.1$, $J = 2.3$, 6- and 6′-H), 7.78 (2H, d, $J = 9.1$, 7- and 7′-H), 7.01 (4H, s, 2″-, 3″-, 5″- and 6″-H), 5.30 (4H, s, CH_2), 3.35 ppm (6H, s, CH_3); ^{13}C NMR [125 MHz, $(CD_3)_2SO$]: $\delta = 160.43$ (C-3 and -3′), 148.15 (C-7a and -7′a), 141.32 (C-5 and -5′), 134.69 (C-1″ and -4″), 127.90 (C-2″, -3″, -5″ and -6″), 127.01 (C-6 and -6′), 120.31 (C-4 and -4′), 115.85 (C-3a and -3′a), 112.17 (C-7 and -7′), 49.49 (CH_2), 29.34 ppm (CH_3); MS (ES+): *m/z* (%): 999 ([2*M*+Na]$^+$, 38), 511 ([*M*+Na]$^+$, 64), 489 ([*M*+H]$^+$, 100), 247 (100). Anal. calcd. for $C_{24}H_{20}N_6O_6$ (488.45): C, 59.01; H, 4.13; N, 17.21. Found: C, 59.29; H, 4.19; N, 17.22.

Determination of pK_a values: Typically, 100–200 mL of 0.5–1 mM solutions of indazolols in water or dimethyl sulfoxide/water (2:1 *v/v*) containing 0.15 M NaCl for constant ionic strength, were titrated by addition of 20–50 μL aliquots of 50–100 mM aq NaOH. Titration experiments were carried out in triplicate at 25 ± 0.5 °C for each indazolol. The experimental data (pH vs. volume of titrant solution, ~60 points) were analyzed with the CurTiPot software,[26] with interpolation of 300 points and a degree of smoothing of 60–70%, to afford the pK_a values (Table 2).

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[1] a) V. J. Arán, J. L. Asensio, J. R. Ruiz, M. Stud, *J. Chem. Res. (S)* **1993**, 218; b) V. J. Arán, J. L. Asensio, J. R. Ruiz, M. Stud, *J. Chem. Res. (M)* **1993**, 1322–1345; c) V. J. Arán, J. L. Asensio, J. R. Ruiz, M. Stud, *J. Chem. Soc. Perkin Trans. 1* **1993**, 1119–1127.

[2] V. J. Arán, M. Flores, P. Muñoz, J. R. Ruiz, P. Sánchez-Verdú, M. Stud, *Liebigs Ann.* **1995**, 817–824.

[3] V. J. Arán, M. Flores, P. Muñoz, J. A. Páez, P. Sánchez-Verdú, M. Stud, *Liebigs Ann.* **1996**, 683–691.

[4] V. J. Arán, C. Ochoa, L. Boiani, P. Buccino, H. Cerecetto, A. Gerpe, M. González, D. Montero, J. J. Nogal, A. Gómez-Barrio, A. Azqueta, A. López de Ceráin, O. E. Piro, E. E. Castellano, *Bioorg. Med. Chem.* **2005**, *13*, 3197–3207.

[5] J. R. Ruiz, V. J. Arán, J. L. Asensio, M. Flores, M. Stud, *Liebigs Ann. Chem.* **1994**, 679–684.

[6] V. J. Arán, J. L. Asensio, J. Molina, P. Muñoz, J. R. Ruiz, M. Stud, *J. Chem. Soc. Perkin Trans. 1* **1997**, 2229–2235.

[7] a) S. de Castro, R. Chicharro, V. J. Arán, *J. Chem. Soc. Perkin Trans. 1* **2002**, 790–802; b) R. Chicharro, S. de Castro, J. L. Reino, V. J. Arán, *Eur. J. Org. Chem.* **2003**, 2314–2326.

[8] a) A. Montero-Torres, M. C. Vega, Y. Marrero-Ponce, M. Rolón, A. Gómez-Barrio, J. A. Escario, V. J. Arán, A. R. Martínez-Fernández, A. Meneses-Marcel, *Bioorg. Med. Chem.* **2005**, *13*, 6264–6275; b) M. C. Vega, A. Montero-Torres, Y. Marrero-Ponce, M. Rolón, A. Gómez-Barrio, J. A. Escario, V. J. Arán, J. J. Nogal, A. Meneses-Marcel, F. Torrens, *Bioorg. Med. Chem. Lett.* **2006**, *16*, 1898–1904.

[9] a) A. Meneses-Marcel, Y. Marrero-Ponce, Y. Machado-Tugores, A. Montero-Torres, D. Montero Pereira, J. A. Escario, J. J. Nogal-Ruiz, C. Ochoa, V. J. Arán, A. R. Martínez-Fernández, R. N. García Sánchez, *Bioorg. Med. Chem. Lett.* **2005**, *15*, 3838–3843; b) Y. Marrero-Ponce, Y. Machado-Tugores, D. Montero Pereira, J. A. Escario, A. Gómez Barrio, J. J. Nogal-Ruiz, C. Ochoa, V. J. Arán, A. R. Martínez-Fernández, R. N. García Sánchez, A. Montero-Torres, F. Torrens, A. Meneses-Marcel, *Curr. Drug Discov. Technol.* **2005**, *2*, 245–265; c) Y. Marrero-Ponce, A. Meneses-Marcel, J. A. Castillo-Garit, Y. Machado-Tugores, J. A. Escario, A. Gómez Barrio, D. Montero Pereira, J. J. Nogal-Ruiz, V. J. Arán, A. R. Martínez-Fernández, F. Torrens, R. Rotondo, F. Ibarra-Velarde, Y. J. Alvarado, *Bioorg. Med. Chem.* **2006**, *14*, 6502–6524; d) O. M. Rivera-Borroto, Y. Marrero-Ponce, A. Meneses-Marcel, J. A. Escario, A. Gómez Barrio, V. J. Arán, M. A. Martins Alho, D. Montero Pereira, J. J. Nogal, F. Torrens, F. Ibarra-Velarde, Y. Vera Montenegro, A. Huesca-Guillén, N. Rivera, C. Vogel, *QSAR Comb. Sci.* **2008**; DOI: 10.1002/qsar.200610165; e) Y. Marrero-Ponce, A. Meneses-Marcel, O. M. Rivera-Borroto, R. García-Domenech, J. V. de Julián-Ortiz, A. Montero, J. A. Escario, A. Gómez Barrio, D. Montero Pereira, J. J. Nogal, R. Grau, F. Torrens, C. Vogel, V. J. Arán, *J. Comput. Aided Mol. Des.* **2008**, *22*, 523–540.

[10] A. Montero-Torres, R. N. García-Sánchez, Y. Marrero-Ponce, Y. Machado-Tugores, J. J. Nogal-Ruiz, A. R. Martínez-Fernández, V. J. Arán, C. Ochoa, A. Meneses-Marcel, F. Torrens, *Eur. J. Med. Chem.* **2006**, *41*, 483–493.

[11] E. Deharo, R. N. García, P. Oporto, A. Gimenez, M. Sauvain, V. Jullian, H. Ginsburg, *Exp. Parasitol.* **2002**, *100*, 252–256.

[12] a) A. C. Chou, R. Chevli, C. D. Fitch, *Biochemistry* **1980**, *19*, 1543–1549; b) A. Dorn, S. R. Vippagunta, H. Matile, C. Jaquet, J. L. Vennerstrom, R. G. Ridley, *Biochem. Pharmacol.* **1998**, *55*, 727–736; c) M. Foley, L. Tilley, *Pharmacol. Ther.* **1998**, *79*, 55–87.

[13] S. Kumar, M. Guha, V. Choubey, P. Maity, U. Bandyopadhyay, *Life Sci.* **2007**, *80*, 813–828.

[14] a) P. A. Winstanley, *Parasitol. Today* **2000**, *16*, 146–153; b) G. A. Biagini, P. M. O'Neill, A. Nzila, S. A. Ward, P. G. Bray, *Trends Parasitol.* **2003**, *19*, 479–487; c) R. Arav-Boger, T. A. Shapiro, *Annu. Rev. Pharmacol. Toxicol.* **2005**, *45*, 565–585.

[15] J. Schmutz, F. Hunziker, W. Michaelis, *Helv. Chim. Acta* **1964**, *47*, 1986–1996.

[16] L. Baiocchi, G. Corsi, G. Palazzo, *Synthesis* **1978**, 633–648.

[17] P. Bruneau, C. Delvare, M. P. Edwards, R. M. McMillan, *J. Med. Chem.* **1991**, *34*, 1028–1036.

[18] B. Cottineau, P. Toto, C. Marot, A. Pipaud, J. Chenault, *Bioorg. Med. Chem. Lett.* **2002**, *12*, 2105–2108.

[19] S. Pagola, P. W. Stephens, D. S. Bohle, A. D. Kosar, S. K. Madsen, *Nature* **2000**, *404*, 307–310.

[20] A. Leed, K. DuBay, L. M. B. Ursos, D. Sears, A. C. de Dios, P. D. Roepe, *Biochemistry* **2002**, *41*, 10245–10255.

[21] The different acidities of 1-substituted indazol-3-ols and the corresponding 5-nitro analogues is reflected in the ^{1}H and ^{13}C NMR spectra of these compounds; in $(CD_3)_2SO$, the signals corresponding to 3-OH and C-3 appear at δ_H ~10.65 ppm (10.63–10.68 ppm) and δ_C ~154.60 ppm (154.48–154.68 ppm), respectively, for the former, and at δ_H ~11.40 ppm (11.33–11.50 ppm) and δ_C ~156.50 ppm (156.23–

156.83 ppm) for the latter (ref. [1], [3] and [4], this paper and unpublished results).
[22] P. Bruneau, P. J. Taylor, A. J. Wilkinson, *J. Chem. Soc. Perkin Trans. 2* **1996**, 2263–2269.
[23] a) I. Constantinidis, J. D. Satterlee, *J. Am. Chem. Soc.* **1988**, *110*, 927–932; b) I. Constantinidis, J. D. Satterlee, *J. Am. Chem. Soc.* **1988**, *110*, 4391–4395; c) H.-J. Schneider, M. Wang, *J. Org. Chem.* **1994**, *59*, 7464–7472; d) A. C. de Dios, R. Tycko, L. M. B. Ursos, P. D. Roepe, *J. Phys. Chem. A* **2003**, *107*, 5821–5825.
[24] W. Peters, B. L. Robinson, *Ann. Trop. Med. Parasitol.* **1992**, *86*, 455–465.
[25] S. D. Wyrick, P. J. Voorstad, G. Cocolas, I. H. Hall, *J. Med. Chem.* **1984**, *27*, 768–772.
[26] I. G. R. Gutz, "CurTiPot (pH and Acid-Base Titration Curves: Analysis and Simulation)", version 3.3.2 for MS-Excel, available online at http://www2.iq.usp.br/docente/gutz/Curtipot_.html (last accessed October 27, 2008).

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