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Short communication

Synthesis and biological activities of *N,N*-dimethyl-2-propen-1-amine derivatives

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Summary — The synthesis of several 3-(4'-bromo[1,1'-biphenyl]-4-yl)-3-(4-*X*-phenyl)-*N,N*-dimethyl-2-propen-1-amine derivatives is described. These compounds are potential trypanocide agents with relative low acute toxicities. The inhibition of growth of *Escherichia coli* by these drugs with different *para*-substitution on the phenyl moiety and their trypanocidal activities against epimastigote from *Trypanosoma cruzi* were investigated.

Chagas's disease / trypanocide / *Trypanosoma cruzi* / aminopropene

Introduction

Chagas's disease (American Trypanosomiasis), which the etiological agent is the flagellate *Trypanosoma cruzi*, has a prevalence estimated at around 16–18 million cases in Latin America. Despite intensive research in the field of chemotherapy of Chagas's disease [1–3], only two nitro heterocyclic drugs, Nifurtimox and Benznidazole, are in clinical use, with severely restricted applicability for chronic patients and a high toxicity. The transmission vector, *Triatoma infestant*, operates by sucking blood, but the second most important route of infection is by blood transfusion [4]. In endemic areas, the use of crystal violet for the chemoprophylaxis of banked blood is recommended [5]. In this direction, 3-(4'-bromo[1,1'-biphenyl]-4-yl)-3-(4-chlorophenyl)-*N,N*-dimethyl-2-propen-1-amine and 3-(4'-bromo[1,1'-biphenyl]-4-yl)-3-(4-bromophenyl)-*N,N*-dimethyl-2-propen-1-amine were described as active against *T cruzi* [6–8], but there have been no further systematic studies related to other biological activities.

We now report the synthesis of new derivatives of 3-*N,N*-dimethyl-2-propen-1-amines characterized by *para*-substitution on the phenyl moiety, their trypanocidal activities on epimastigote culture of *T cruzi*, and their acute toxicities acting on *Escherichia coli* (ATCC 25922 strain).

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Chemistry

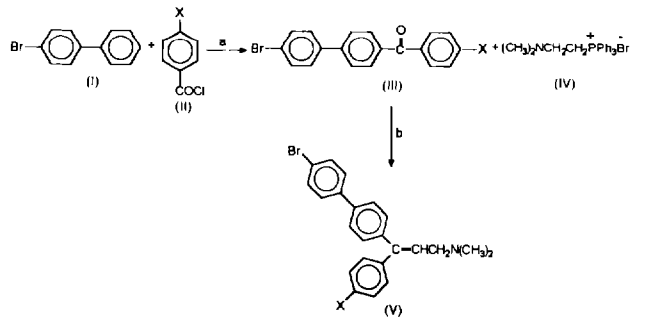
3-*N,N*-Dimethyl-2-propen-1-amine derivatives were prepared by Friedel-Crafts reaction between 4-bromobiphenyl and the corresponding benzoyl chloride to give the ketones **III** and by subsequent Wittig reaction of the latter with β-(*N,N*-dimethylamino)ethyltriphenylphosphonium bromide (scheme 1). 2-Propen-1-amines derivatives were obtained with high purity and good yields (50–62%) [8].

Biology

Epimastigote, amastigote and trypomastigote forms from *T cruzi* (Y strain) were used [9–11]. Acute toxicities on *E coli* (ATCC 25922 strain) were measured by flow injection analysis [12, 13].

Results and discussion

Table I shows the effect of compounds **V** on the epimastigote culture (ID_{50}). The most effective



Scheme 1. a) $AlCl_3$ in CS_2 or nitrobenzene; refluxing 5–8 h: CH_2Cl_2 extraction/NaOH. b) *n*BuLi/THF/compound IV, 30 min 0 °C; addition compound **III**, 12 h 25 °C; ether/chloroform extraction. X: a: Cl; b: H; c: OCH_3; d: NO_2; e: CH_3; f: SO_2CH_3; g: Br; h: I.

compound was **Vb** (X = H) (12.8 ± 0.1 μM) compared to that of X = Cl (17.5 ± 0.7 μM) and to X = Br (14.8 ± 0.6 μM). Compound **Vb** was 8.1-fold more active than Nifurtimox, a known drug for Chagas's disease (table I). Preliminary studies of **Vb** acting on trypomastigote and amastigote forms were also quite efficient (18.8 ± 1.2 and 6.6 ± 1.6 μM, respectively).

In these experiments Nifurtimox exhibited for trypomastigote and amastigote ID_{50} values of 157.0 and 207.3 μM, respectively.

E coli (ATCC 25922 strain) was used in the acute toxicity assay. The results are presented in table I. The most toxic compounds were **Vd** (X = NO_2) and **Ve** (X = Me) with IC_{50} values of 5.7 and 4.4 μM, respectively. Nifurtimox, halogens, SO_2Me derivative and non-substituted *para*-X-phenyl groups were the least toxic compounds by this technique. We are aware that these data are limited in terms of the complex cellular system, but a significant correlation was established for sets of multicenter evaluation of in vitro cytotoxicity (MEIC) chemicals between minimal inhibitory concentration (MIC) in bacteria and the relevant end point concentration in various tests on human cells [14].

Table I. Physical properties, toxicity and trypanocidal activities (Epimastigotes) of compounds **V**[a].

Substituent para-X	*Acute toxicity IC_{50}/1 h (μM)*	*Trypanocide activity ID_{50}/24 h (μM)*
Va-Cl	19.2	17.5
Vb-H	12.8	12.8
Vc-OMe	10.7	29.3
Vd-NO_2	5.7	26.3
Ve-Me	4.4	28.6
Vf-SO_2Me	110.0	21.2
Vg-Br	24.0	14.8
Vh-I	28.0	19.9
Nifurtimox	35.0	103.7

[a]Mixture of *Z*/*E* isomers (nearly 1:1).

In summary, based on acute toxicity series and trypanocidal activities, the unsubstituted phenyl group and the SO_2Me groups in the new 2-propen-1-amine derivatives make them interesting compounds with a great potential as a trypanocide new drug. Studies of lethal dose, in vivo trypanocidal activity in mice and QSAR correlation with other biological properties are presently in progress.

Experimental protocols

Chemistry

Syntheses of 3-*N*,*N*-dimethyl-2-propen-1-amines were achieved by the Wittig reaction between 4'-bromo[1,1'-biphenyl]-4-yl 4-X-phenyl ketone and the ylid from β-(*N*,*N*-dimethylamino) ethyl triphenylphosphonium bromide by a modified process from that previously published [7]. The solvent, temperature, separation method and reaction time were modified to optimize yields [8] (scheme 1).

*General procedure for the preparation of the ketones **IIIa–h** [4-bromo-[4-biphenyl]-4-yl X-phenyl methanone]*

The acid chloride (2 mmol) was added dropwise under agitation at room temperature to a solution containing 4-bromobiphenyl (1.72 mmol), aluminium chloride (3 mmol) and carbon disulfide (4 mL). The mixture was refluxed for 7 h and the solvent distilled. The mixture was then extracted with dichloromethane (40 mL), 10% NaOH solution (20 mL) and water (2 x 40 mL). The organic layer was separated and dried under $MgSO_4$. After filtration, the solvent was evaporated under reduced pressure to give a solid product (ketone **III**), which was recrystallized. The results are shown in table II.

*4'-Bromo-[1,1'-biphenyl]-4-yl p-chlorophenyl methanone **IIIa**.* ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.2–8.0 (m, 12H, aromatic) ppm; MS *(m/z)*: 372 (M^+, 85), 259 (60), 152 (100), 11 (28), 18 (28); IR (KBr): 3452 (C=O stretch, overtone), 1643 (C=O stretch) and 1558 (C=C stretch aromatic) cm^{-1}.

*4'-Bromo-[1,1'-biphenyl]-4-yl phenyl methanone **IIIb**.* ^{1}H-NMR (80 MHz; $CDCl_3$/TMS): δ 7.2–8.0 (m, 13H, aromatic) ppm; MS *(m/z)*: 337 (M^+, 100), 262 (72), 152 (39), 77 (29), 18 (21); IR (KBr): 3418 (C=O stretch, overtone), 1644 (C=O stretch) and 1596 (C=C stretch, aromatic) cm^{-1}.

*4'-Bromo-[1,1'-biphenyl]-4-yl p-methoxyphenyl methanone **IIIc**.* ^{1}H-NMR (80 MHz; $CDCl_3$/TMS): δ 6.8–8.0 (m, 12H, aromatic), 4.0 (s, 3H, OCH_3) ppm; MS *(m/z)*: 368 (M^+, 12), 259 (6), 152 (32), 135 (100), 43 (31); IR (KBr): 3628 (C=O stretch, overtone), 1638 (C=O stretch), 1577 (C=C stretch, aromatic) and 1261 (C-O stretch) cm^{-1}.

*4'-Bromo-[1,1'-biphenyl]-4-yl p-nitrophenyl methanone **IIId**.* ^{1}H-NMR (80 MHz; $CDCl_3$/TMS): δ 7.0–8.4 (m, 12H, aromatic) ppm; MS *(m/z)*: 383 (M^+, 99), 381 (100), 259 (88), 76 (17), 18 (96); IR (KBr): 3452 (C=O stretch, overtone), 1646 (C=O stretch), 1586 (C=C stretch, aromatic), 1348 (N-O stretch) and 850 (C-N stretch) cm^{-1}.

Table II. Characteristics of the synthesis and physical properties of the compounds **IIIa–h**.

Compound	*RS*	*RT (h)*	*SS*[a]	*Yield (%)*	*Mp (°C)*	*UV*[b] λ_{max} *(nm)*
IIIa	CS_2	7	1	79	204	266
IIIb	CS_2	5	1	72	156–157	292
IIIc	CS_2	8	2	62	219–220	258
IIId	CS_2	7	1	75	182–183	272
IIIe	CS_2	7	1	82	195	290
IIIf	$C_6H_5NO_2$	7	3	50	201–202	246
IIIg	CS_2	5	1	68	215	294
IIIh	CS_2	7	1	80	239	290

RS: reaction solvent. RT: reaction time. SS: solvent system for recrystallization. [a]1: CH_2Cl_2/hexane; 2: benzene; 3: ethanol. [b]CH_2Cl_2 as solvent.

4'-Bromo-[1,1'-biphenyl]-4-yl p-methylphenyl methanone ***IIIe***. ^{1}H-NMR (80 MHz; $CDCl_3$/TMS): δ 7.2–7.9 (m, 12H, aromatic), 2.5 (s, 3H, CH_3) ppm; MS *(m/z)*: 352 (M^+, 98), 351 (20), 350 (100), 152 (36), 199 (38), 43 (18), 18 (81); IR (KBr): 3429 (C=O stretch, overtone), 1643 (C=O stretch) and 1604 (C=C stretch, aromatic) cm^{-1}.

4'-Bromo-[1,1'-biphenyl]-4-yl methylsulfonyl methanone ***IIIf***
^{1}H-NMR (300 MHz, $CDCl_3$/TMS): δ 8.10 (d, 2H, aromatic), 8.00 (d, 2H, aromatic), 7.70 (d, 2H, aromatic), 7.60 (d, 2H, aromatic), 7.50 (d, 2H, aromatic), 3.10 (s, 3H, SO_2CH_3) ppm; MS: 416 (M^+, 19), 414 (33), 260 (13), 149 (13), 58 (26), 43 (100); IR (KBr) 3422 (C=O stretch, overtone), 1648 (C=O, stretch), 1602 (C=C stretch, aromatic), 1289 (S=O stretch) and 1151 (C=S stretch) cm^{-1}.

4'-Bromo-[1,1'-biphenyl]-4-yl p-bromophenyl methanone ***IIIg***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.8–8.0 (m, 12H, aromatic) ppm; MS *(m/z)*: 416 (M^+, 19), 414 (33), 260 (13), 149 (13), 58 (26), 43 (100); IR (KBr): 3449 (C=O stretch, overtone), 1643 (C=O stretch) and 1581 (C=C stretch, aromatic) cm^{-1}.

4'-Bromo-[1,1'-biphenyl]-4-yl p-iodophenyl methanone ***IIIh***. ^{1}H- NMR (300 MHz; $CDCl_3$/TMS): δ 7.5–8.4 (m, 12H, aromatic) ppm; MS *(m/z)*: 310 (M^+, 11), 231 (7), 152 (13), 58 (22), 43 (100), 18 (29); IR (KBr): 3418 (C=O stretch, overtone), 1643 (C=O stretch) and 1577 (C=C stretch, aromatic) cm^{-1}.

General procedure for the preparation of the derivatives ***Va–h*** *{3-(4'-bromo[1,1'-biphenyl]-4-yl)-3-(4-X-phenyl)-N,N-dimethyl-2-propen-1-amine}*
To a solution containing β-dimethylamino ethyl triphenyl phosphonium bromide (2.15 mmol) in THF (12 mL), a solution of *n*-butyllithium in 2.5 mmolar hexane (2.21 mmol) was slowly added at 0 °C under an argon atmosphere. The reaction mixture was kept at 0 °C for a further 30 min. The intermediate ketone **IIIa–h** (1.07 mmol) was added to the mixture under an argon atmosphere and stirred overnight at room temperature. The mixture was extracted with chloroform (20 mL) and water (2 x 20 mL), the organic layer separated and dried over $MgSO_4$. After filtration, the solvent was evaporated under reduced pressure and the crude product obtained as a dark yellow oil. The product was purified by percolation in a silica-gel column. The yields and physical properties of compounds **Va–h** are shown in table III.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-chlorophenyl)-N,N-dimethyl-2-propen-1-amine ***Va***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.1–7.7 (m, 12H, aromatic), 6.3 (t, 1H, CH-*trans*), 6.2 (t, 1H, CH-*cis*), 3.1 (d, 2H, CH_2-*cis*), 3.0 (d, 2H, CH_2-*trans*), 2.3 (2s, 6H, $N(CH_3)_2$) ppm; MS *(m/z)*: 427 (M^+, 47), 383 (13), 302 (49), 265 (36), 194 (76), 165 (40), 70 (100), 58 (91), 42 (55); IR (KBr): 1654 and 1634 (C=C stretch, olefin, trisubstituted), 1168 (C-N stretch) and 1487 (C=C stretch, aromatic) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(phenyl)-N,N-dimethyl-2-propen-1-amine ***Vb***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.2–7.7 (m, 13H, aromatic), 6.3 (t, 1H, CH-*trans*), 6.25 (t, 1H, CH-*cis*), 3.1 (d, 2H, CH_2-*cis*), 3.0 (d, 2H, CH_2-*trans*), 2.3 (s, 3H, $N(CH_3)_2$-*cis*) ppm; MS *(m/z)*: 392 (M^+, 34), 314 (20), 268 (32), 252 (27), 160 (52), 91 (59), 70 (100), 58 (93), 42 (52); IR (KBr): 1654, 1636 (C=C stretch, olefin), 1167 (C-N stretch) and 1487 (C=C stretch, aromatic) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-methoxyphenyl)-N,N-dimethyl-2-propen-1-amine ***Vc***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 6.8–7.1 (m, 12H, aromatic), 6.2 (t, 1H, CH-*trans*), 6.1 (t, 1H, CH-*cis*), 3.9 (s, 3H, OCH_3-*trans*), 3.85 (s, 3H, OCH_3-

Table III. Characteristics of the synthesis and physical properties of the compounds **Va–h**.

Compound	*SS*[a]	*Yield (%)*	*Mp (°C)*	*UV*[d] λ_{max} *(nm)*
Va	1 (8:2:1)[b]	50	115–120	272
Vb	1 (9:1:1)[b]	60	100–102	276
Vc	1 (9:1:1)[b]	45	Oil	268
Vd	1 (100:1)[b]	58	100–102	280
Ve	1 (8:2:1)[b]	58	85–87	272
Vf	2 (5–15%)[c]	62	Oil	264
Vg	2 (1–5%)[c]	50	101	268
Vh	2 (1–5%)[c]	56	142	268

SS: solvent system for column chromatography. [a]1: hexane/diethyl ether/triethylamine; 2: ethyl acetate/methanol. [b]Volume ratio. [c]Percentage ratio. [d]CH_2Cl_2 as solvent.

cis), 3.0–3.1 (2d, 2H, CH_2), 2.3 (s, 6H, $N(CH_3)_2$) ppm; MS *(m/z)*: 423 (M^+, 34), 377 (32), 298 (16), 239 (18), 190 (73), 145 (34), 121 (100), 70 (54), 58 (52), 42 (32); IR (KBr): 1654 (C=C stretch, olefin), 1509, 1480 (C=C stretch, aromatic) and 1167 (C-N stretch) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-nitrophenyl)-N,N-dimethyl-2-propen-1-amine ***Vd***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.0–8.4 (m, 12H, aromatic), 6.35–6.45 (2t, 1H, CH), 3.15 (d, 2H, CH_2-*cis*), 3.0 (d, 2H, CH_2-*trans*), 2.35 (s, 6H, $N(CH_3)_2$-*cis*), 2.25 (s, 6H, $N(CH_3)_2$-*trans*) ppm; MS *(m/z)*: 436 (M^+, 33), 346 (9), 314 (27), 265 (33), 205 (58), 165 (24), 131 (16), 70 (100), 58 (93), 42 (50); IR (KBr): 1174 (C-N stretch), 1343 (N-O stretch) and 853 (C-N stretch) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-methyl)-N,N-dimethyl-2-propen-1-amine ***Ve***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.05–7.6 (m, 12H, aromatic), 6.3 (t, 1H, CH-*trans*), 6.2 (t, 1H, CH-*cis*), 6.2–6.3 (2t, 1H, CH), 3.1 (d, 2H, CH_2), 2.25 (s, 6H, $N(CH_3)_2$), 2.25 ppm; MS *(m/z)*: 405 (M^+, 36), 361 (16), 314 (18), 256 (27), 189 (20), 174 (66), 105 (75), 70 (100), 58 (88), 43 (95); IR (KBr): 1654 (C=C stretch, olefin), 1508, 1488 (C=C stretch, aromatic) and 1168 (C=N stretch) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-methylsulphonylphenyl)-N,N-dimethyl-2-propen-1-amine ***Vf***. ^{1}H NMR (300 MHz; $CDCl_3$/TMS): δ 7.2–8.1 (m, 12H, aromatic), 6.35–6.45 (2t, 1H, CH-*cis* and -*trans*), 3.2 (d, 2H, CH_2-*cis*), 3.15 (s, 3H, CH_3SO_2-*cis*), 3.1 (d, 2H, CH_2-*trans*), 3.05 (s, 3H, CH_3SO_2-*trans*), 2.4 (s, 6H, $N(CH_3)_2$-*trans*), 2.3 (s, 6H, $N(CH_3)_2$-*cis*) ppm; MS *(m/z)*: 470 (M^+, 7), 256 (28), 239 (10), 227 (9), 213 (11), 185 (14), 167 (28), 149 (100), 83 (70), 47 (59); IR (KBr): 1654 (C=C stretch, olefin), 1481, 1438 (C-N stretch, amine), 1264 (S=O stretch), 1147 (C=N stretch) and 1116 (C=C, C=S stretches) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-bromophenyl)-N,N-dimethyl-2-propen-1-amine ***Vg***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.0–7.6 (m, 12H, aromatic), 6.3 (t, 1H, CH-*trans*), 6.2 (t, 1H, CH-*cis*), 3.1 (d, 2H, CH_2-*cis*), 3.0 (d, 2H, CH_2-*trans*), 2.25 (s, 6H, $N(CH_3)_2$) ppm; MS *(m/z)*: 471 (M^+, 29), 392 (2), 346 (27), 265 (32), 238 (45), 154 (93), 125 (46), 70 (100), 58 (68), 42 (59); IR (KBr): 1654 (C=C stretch, olefin), 1460, 1450 (C=C stretch) and 1171 (C-N stretch) cm^{-1}.

3-(4'-Bromo-[1,1'-biphenyl]-4-yl)-3-(4-iodophenyl)-N,N-dimethyl-2-propen-1-amine ***Vh***. ^{1}H-NMR (300 MHz; $CDCl_3$/TMS): δ 7.1–7.6 (m, 12H, aromatic), 6.3 (t, 1H, CH-*trans*), 6.2 (t, 1H, CH-*cis*), 3.1 (d, 2H, CH_2-*cis*), 3.0 (d, 2H, CH_2-*trans*), 2.25 (s, 6H, $N(CH_3)_2$) ppm; MS *(m/z)*: 517 (M^+, 20), 439 (4), 348 (29), 286 (27), 265 (36), 165 (33), 115 (35), 70 (100), 58 (80), 42 (52); IR (KBr): 1654 (C=C stretch, olefin), 1450, 1480 (C=C stretching aromatic) and 1172 (C-N stretch) cm^{-1}.

NOE experiments

The configurational assignment of the stereomers formed in the Wittig olefination was carried out by differencial NOE experiments, in the previously isolated isomers, eg, in (*E*)-**Vf**, irradiation of the olefinic proton (δ 6.4 ppm) led to a 13% increment in the signal of the *ortho* protons of the methylsulfonyl group (δ 7.4 ppm) while irradiation of the olefinic proton (δ 6.4 ppm) in the (*Z*)-**Vf** isomer gave rise to a 9% increment in the signal of the *ortho* protons of the biphenyl ring (δ 7.3 ppm) (fig 1).

On the basis of their ^{1}H-NMR spectra compounds **Va–h** were formed as a 1:1 mixture of geometrical isomers in all cases studied, and these isomeric mixtures were used as free bases for the assay of acute toxicity and trypanocidal activities.

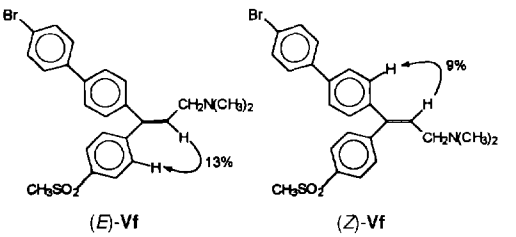


Fig 1. Selected NOE differences for (*Z*)-**Vf** and (*E*)-**Vf**.

Biology

Epimastigote forms from *T cruzi* (Y strain) [9], maintained in LIT medium, were harvested during the exponential growth phase. Stock solutions of the drug were prepared in dimethylsulphoxide and the final concentration never exceeded 0.1%.

The assay with epimastigote, in LIT medium, were performed at 28 °C. Cell counts were performed after 24 h of incubation with the drug using a Neubauer chamber. The drug concentration corresponding to 50% parasite elimination was expressed as ID_{50} [10]. Bloodstream trypomastigotes from infected mice and amastigote from supernatant of infected J774G-8 macrophages were obtained as previously described [11]. *E coli* (ATCC 25922 strain) was used in the acute toxicity assay and the method for determination of bacterial cytotoxicty by flow injection analysis [12, 13] proved to be reliable for a rapid assessment of the toxicity of the compounds by analyzing alterations of the amount of CO_2 produced and trapped in the culture medium. Although these values did not correlate with trypanocidal activity, the technique is important in order to assess the toxicity relationship in the same series.

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