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Synthesis and antimycobacterial activity of some 4*H*-1,2,4-triazin-5-one derivatives☆

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Abstract

6-[(Arylmethylenamino)carbonyl]-3-(pyridin-2-yl)-4*H*-1,2,4-triazin-5-one derivatives were synthesized and tested for their in vitro antimycobacterial activity. Some compounds showed interesting activity against a strain of *Mycobacterium tuberculosis*.

Keywords: 4*H*-1,2,4-Triazin-5-one derivatives; Antimycobacterial activity

1. Introduction

During the course of our studies on new drugs for the treatment of diseases induced by mycobacteria, we described the synthesis and the antimycobacterial properties of many amidrazone derivatives [1–6]. From these studies we claimed the importance of the 2-pyridinecarboxamidrazone moiety for the antimycobacterial activity. These findings prompted us to prepare a series of new compounds in which the amidrazone moiety is constricted in the cyclic system of the 4*H*-1,2,4-triazin-5-one derivatives **1**–**22** (Table 1). Moreover, in the synthesized compounds **1**–**22** a substituted benzylamino group was connected by an amide linkage to the 1,2,4-triazine nucleus in consideration of the antimycobacterial activity of a great number of benzylamine derivatives [7]. The new synthesized compounds **1**–**22** were tested for their in vitro antimycobacterial activity towards a strain of *Mycobacterium tuberculosis* $H_{37}Rv$. Some compounds showed an interesting in vitro inhibitory activity, their MICs ranging from 8 to 32 μg/ml.

2. Chemistry

The synthesis of 6-[(arylmethylenamino)carbonyl]-3-(pyridin-2-yl)-4*H*-1,2,4-triazin-5-ones **1**–**22** (Table 1) was carried out (Scheme 1) by adding variously substituted benzylamines to 3-(pyridin-2-yl)-4*H*-1,2,4-triazin-5-one-6-carboxylic acid ethyl ester **23**, which in turn was prepared from 2-pyridinecarboxamidrazone and diethyl ketomalonate, according to the previously proposed method [8].

The triazinone derivative **23** may occur in three tautomeric forms, namely, two triazinone forms **23a** and **23b**, in which the NH group is in the *para* and *ortho* position with respect to the carbonyl group, and the hydroxytriazine form **23c**. From literature data [9] for similar compounds, the structures **23a** and **23b** are the predominant tautomeric forms. However, in the synthesis of compounds **1**–**22** the employed arylmethylenamines produced their water soluble salts **1a**–**22a** by interacting with the acidic hydroxy group of the hydroxytriazine tautomeric form causing a shifting of the tautomeric equilibrium. It was necessary for the formation of the compounds **1**–**22** to employ an excess of arylmethylenamines (see Section 3) and liberate the free compounds from the aqueous solution of their salts by adding hydrochloric acid. The formation of the arylmethylenamine salts of compounds **1**–**22** was demon-

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strated by isolating and purifying the benzylamine salt **1a** of compound **1**. The structure of **1a** was confirmed by the IR spectrum, which showed the absence of the carbonyl function in the triazine cycle, and by NMR.

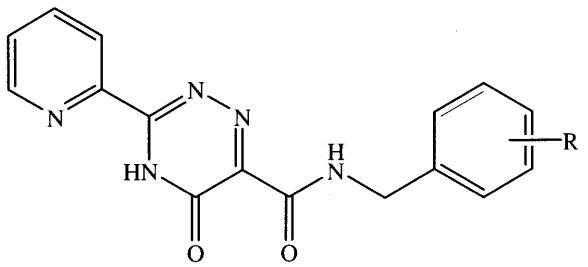
Compounds **19–22** were similarly obtained by using arylmethylenamine hydrochlorides in the presence of triethylamine.

3. Experimental

3.1. Chemistry

Melting points were determined with a Büchi 510 capillary apparatus, and are uncorrected. Infrared spectra in nujol mulls were recorded on a Jasco FT 200 spectrophotometer. Proton nuclear magnetic resonance (^{1}H NMR) spectra were determined on a Varian Gemini 200 spectrometer; chemical shifts are reported as δ (ppm) relative to tetramethylsilane as internal standard and with the indicated solvent. Reaction courses and product mixture were routinely monitored by thin-layer chromatography (TLC) on silica gel precoated F_{254} Merck plates. EI–MS spectra (70 eV) were taken on a VG 7070 spectrometer. Elemental analyses (C, H, N) were performed on a Carlo Erba analyzer and were within $\pm$0.3 of the theoretical value.

Table 1
Chemical formulae, yields and melting points for compounds **1–22**



1-22

Comp.	R	Yield (%)	M.p. (°C)	Formula
1	H	51	255–256	$C_{16}H_{13}N_5O_2$
2	2-CH_3	32	233–234	$C_{17}H_{15}N_5O_2$
3	3-CH_3	75	345–247	$C_{17}H_{15}N_5O_2$
4	4-CH_3	61	257–258	$C_{17}H_{15}N_5O_2$
5	2-OCH_3	95	210–212	$C_{17}H_{15}N_5O_3$
6	3-OCH_3	81	237–238	$C_{17}H_{15}N_5O_3$
7	4-OCH_3	64	246–248	$C_{17}H_{15}N_5O_3$
8	2-Cl	78	257–259	$C_{16}H_{12}N_5O_2Cl$
9	3-Cl	67	277–278	$C_{16}H_{12}N_5O_2Cl$
10	4-Cl	61	224–226	$C_{16}H_{12}N_5O_2Cl$
11	2,4-Cl_2	24	249–251	$C_{16}H_{11}N_5O_2Cl$
12	3,4-Cl_2	35	265–266	$C_{16}H_{11}N_5O_2Cl$
13	2-F	91	244–245	$C_{16}H_{12}N_5O_2F$
14	3-F	68	276–278	$C_{16}H_{12}N_5O_2F$
15	4-F	63	272–274	$C_{16}H_{12}N_5O_2F$
16	2-CF_3	76	241–243	$C_{17}H_{12}N_5O_2F_3$
17	3-CF_3	58	274–276	$C_{17}H_{12}N_5O_2$
18	4-CF_3	59	325–327	$C_{16}H_{12}N_5O_2$
19	2-Br	28	246–248	$C_{16}H_{12}N_5O_2$
20	3-Br	29	275–276	$C_{16}H_{12}N_5O_2$
21	4-Br	33	278–280	$C_{16}H_{12}N_5O_2$
22	3-NO_2	26	306–307	$C_{16}H_{12}N_6O_4$

3.1.1. 3-(Pyridin-2-yl)-4H-1,2,4-triazin-5-one-6-carboxylic acid ethyl ester (**23**)

To a solution of 6 g (44 mmol) of 2-pyridinecarboxamidrazone in 30 ml of absolute ethanol were added dropwise 7.6 g (44 mmol) of diethyl ketomalonate under cooling (0–5°C) and stirring. After addition, the reaction mixture was stirred for 48 h keeping the temperature at about 4°C. The reaction mixture was heated under reflux for 1 h and concentrated under reduced pressure. After 24 h the solid precipitate was collected by filtration, washed with ether and dissolved in chloroform. After further filtration, the solvent was removed under reduced pressure to give a yellow solid, m.p. 138–140°C. Yield: 5.6 g (51%). IR (nujol, cm^{-1}): 3460, 1740, 1650. ^{1}H NMR ($CDCl_3$/TMS): δ 1.4 (t, 3H, CH_2 4.4 (q, 2H, CH_2), 7.6 (m, 1H, pyr), 7.9 (m, 1H, pyr), 8.5 (d, 1H, pyr), 8.7 (d, 1H, pyr), 12.2 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 246 [M^+]. *Anal.* ($C_{11}H_{10}N_4O_3$): C, H, N.

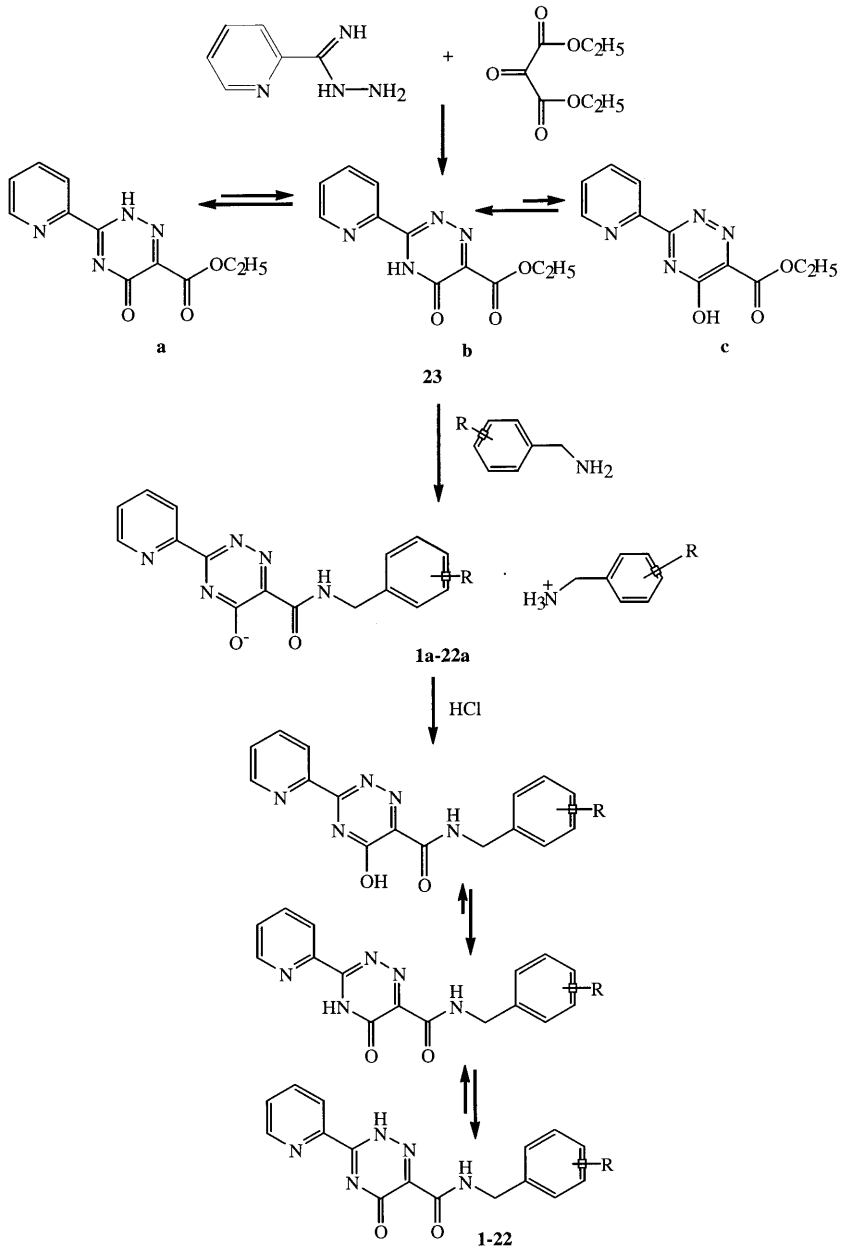
3.1.2. 6-[(Benzylamino)carbonyl]-5-hydroxy-3-(pyridin-2-yl)-1,2,4-triazine benzylamine salt (**1a**)

To a solution of 3.5 g (14 mmol) of 3-(pyridin-2-yl)-4*H*-1,2,4-triazin-5-one-6-carboxylic acid ethyl ester **23** in 20 ml of methanol, 3.3 ml (30 mmol) of benzylamine were added dropwise and the reaction mixture was heated at reflux for 24 h. Thereafter, the reaction mixture was cooled at room temperature (r.t.) and the precipitate was collected by filtration and recrystallized from absolute ethanol to give 3.3 g (57%) of a yellow solid; m.p. 184–185°C.

IR (nujol, cm^{-1}): 3160, 3060, 1680; (KBr, cm^{-1}): 3160, 3060, 300–2810 (br.s.), 1680. 1HNMR (DMSO/TMS): δ 4.15 (s, 2H, $H_3N^+-CH_2-$), 4,50 (d, 2H, NH–CH_2–), 7.10–7.50 (m, 1H, 10H arom. and 1H pyr.), 7.80–8.75 (m, 3H, pyr.), 7.40–9.25 (br.s., 3H, $^+NH_3$, disappearing on deuteration), 11.0 (t, 1H, N*H*–CH_2, disappearing on deuteration). MS: m/z 414 [M^+]. *Anal.* ($C_{23}H_{22}N_6O_2$): C, H, N.

3.1.3. 6-[(Benzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**1**)

To a solution of 3.95 g (16 mmol) of 3-(pyridin-2-yl)-4*H*-1,2,4-triazin-5-one-6-carboxylic acid ethyl ester **23** in 20 ml of methanol, 3.8 ml (35 mmol) of benzylamine were added dropwise and the reaction mixture was heated at reflux for 24 h. Thereafter, the reaction mixture was cooled at r.t. and the precipitate was



Scheme 1.

collected by filtration and dissolved in water. The aqueous solution was acidified with 1 M HCl and the precipitated solid was filtered off, washed with cold water, dried and recrystallized from ethanol to obtain 2.5 g (51%) of **1**; m.p. 255°C. IR (nujol, cm^{-1}): 3200, 3120, 3060, 1690, 1680. ^1H NMR ($CDCl_3$/TMS): δ 4.5 (d, 2H, CH_2) 7.3–8.8 (m, 5H arom. and 4H pyr.), 9.7 (t, 1H, NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 307 [M^+]. *Anal.* ($C_{16}H_{13}N_5O_2$): C, H, N.

In an analogous way the following compounds (**2**–**18**) have been prepared. Yields and melting points are reported in Table 1.

*3.1.4. 6-[(2-Methylbenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**2**)*

IR (nuiol, cm^{-1}): 3240, 3120, 3080, 1685, 1660. ^1H NMR (DMSO/TMS): δ 1.5 (s, 3H, CH_3), 3.8 (d, 2H, CH_2) 6.4–8.1 (m, 4H arom. and 4H pyr.) 8.8 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 321 [M^+]. *Anal.* ($C_{17}H_{15}N_5O_2$): C, H, N.

*3.1.5. 6-[(3-Methylbenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**3**)*

IR (nujol, cm^{-1}): 3200, 3160, 3080, 1690, 1680. ^1H NMR (DMSO/TMS): δ 2.2 (s, 3H, CH_3), 4.5 (d, 2H, CH_2), 7.0–8.6 (m, 4H arom. and 4H pyr), 9.6 (t, 1H,

NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 321 [M^+]. *Anal.* ($C_{17}H_{15}N_5O_2$): C, H, N.

*3.1.6. 6-[(4-Methylbenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**4**)*

IR (nujol, cm^{-1}): 3200, 3160, 3060, 1690, 1680. ^1H NMR (DMSO/TMS): δ 2.2 (s, 3H, CH_3), 4.4 (d, 2H, CH_2) 7.1–8.8 (m, 4H arom. and 4H pyr.), 9.6 (t, 1H, NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 321 [M^+]. *Anal.* ($C_{17}H_{15}N_5O_2$): C, H, N.

*3.1.7. 6-[(2-Methoxybenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**5**)*

IR (nujol, cm^{-1}): 3160, 3120, 3080, 1695, 1670. ^1H NMR (DMSO/TMS): δ 3.8 (s, 3H, CH_3), 4.4 (d, 2H, CH_2), 6.9–8.8 (m, 4H arom. and 4H pyr.), 9.7 (t, 1H, NH disappearing on deuteration), 14.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 337 [M^+]. *Anal.* ($C_{17}H_{15}N_5O_3$): C, H, N.

*3.1.8. 6-[(3-Methoxybenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**6**)*

IR (nujol, cm^{-1}): 3280, 3120, 3060, 1680, 1660. ^1H NMR (DMSO/TMS): δ 3.7 (s, 3H, CH_3), 4.4 (d, 2H, CH_2), 6.0–8.8 (m, 4H arom. and 4H pyr), 8.9 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 337 [M^+]. *Anal.* ($C_{17}H_{15}N_5O_3$): C, H, N.

*3.1.9. 6-[(4-Methoxybenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**7**)*

IR (nujol, cm^{-1}): 3280, 3120, 3060, 1680, 1660. ^1H NMR (DMSO/TMS): δ 3.7 (s, 3H, CH_3), 4.4 (d, 2H, CH_2), 6.0–8.2 (m, 4H arom. and 4H pyr), 8.8 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 337 [M^+]. *Anal.* ($C_{17}H_{15}N_5O_3$): C, H, N.

*3.1.10. 6-[(2-Chlorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**8**)*

IR (nujol, cm^{-1}): 3200, 3130, 3070, 1690, 1660. ^1H NMR (DMSO/TMS): δ 3.9 (d, 2H, CH_2), 6.5–7.9 (m, 4H arom. and 4H pyr), 8.9 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 341 [M^+], 343. *Anal.* ($C_{16}H_{12}N_5O_2C1$): C, H, N.

*3.1.11. 6-[(3-Chlorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazlin-5-one (**9**)*

IR (nujol, cm^{-1}): 3260, 3180, 3080, 1670, 1660. ^1H NMR (DMSO/TMS): δ 4.5 (d, 2H, CH_2), 7.2–8.8 (m, 4H arom. and 4H pyr), 9.6 (t, 1H, NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 341 [M^+], 343. *Anal.* ($C_{16}H_{12}N_5O_2Cl$): C, H, N.

*3.1.12. 6-[(4-Chlorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**10**)*

IR (nuiol, cm^{-1}): 3240, 3140, 3060, 1690, 1680. ^1H NMR (DMSO/TMS): δ 4.5 (d, 2H, CH_2), 7.1–8.8 (m, 4H arom. and 4H pyr), 9.6 (t, 1H, NH disappearing on deuteration), 14.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 341 [M^+], 343. *Anal.* ($C_{16}H_{12}N_5O_2Cl$): C, H, N.

*3.1.13. 6-[(2,4-Dichlorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**11**)*

IR (nujol, cm^{-1}): 3220, 3150, 3080, 1680, 1670. ^1H NMR (DMSO/TMS): δ4.5 (d, 2H, CH_2), 7.4–8.8 (m, 3H arom. and 4H pyr.), 9.9 (t, 1H, NH disappearing on deuteration), 14.2 (br.s., 1H, NH, disappearing on deuteration). MS: m/z, 375 [M^+], 377, 379. Anal. ($C_{16}H_{11}N_5O_2Cl_2$): C, H, N.

*3.1.14. 6-[(3,4-Dichlorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**12**)*

IR (nujol, cm^{-1}): 3220, 3140, 3070, 1690, 1670. ^1H NMR (DMSO/TMS): δ 4.8 (d, 2H, CH_2), 7.4–8.9 (m, 3H arom. and 4H pyr.), 9.8 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z, 375 [M^+], 377, 379. *Anal.* ($C_{16}H_{11}N_5O_2Cl_2$): C, H, N.

*3.1.15. 6-[(2-Fluorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**13**)*

IR (nujol, cm^{-1}): 3230, 3150, 3060, 1690, 1680. ^1H NMR (DMSO/TMS): δ 4.5 (d, 2H, CH_2), 7.1–8.8 (m, 4H arom. and 4H pyr.), 9.7 (t, 1H, NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 325 [M^+]. *Anal.* ($C_{16}H_{12}N_5O_2F$): C, H, N.

*3.1.16. 6-[(3-Fluorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**14**)*

IR (nujol, cm^{-1}): 3240, 3120, 3080, 1680, 1660. ^1H NMR (DMSO/TMS): δ 4.8 (d, 2H, CH_2), 6.9–8.8 (m, 4H arom. and 4H pyr.), 9.6 (t, 1H, NH disappearing on deuteration), 14.7 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 325 [M^+]. *Anal.* ($C_{16}H_{12}N_5O_2F$): C, H, N.

*3.1.17. 6-[(4-Fluorobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**15**)*

IR(nujol, cm^{-1}): 3240, 3120, 3080, 1680, 1665. ^1H NMR (DMSO/TMS): δ 3.6(d, 2H, CH_2), 6.3–8.0 (m, 4H arom. and 4H pyr.), 8.9 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: m/z 325 [M^+]. *Anal.* ($C_{16}H_{12}N_5O_2F$): C, H, N.

*3.1.18. 6-[(2-Trifluoromethylbenzylamino)-carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**16**)*

IR(nujol, cm^{-1}): 3200, 3120, 3060, 1680, 1670. ^1H NMR (DMSO/TMS): δ 3.9(d, 2H, CH_2), 6.6–8.0 (m, 4H arom. and 4H pyr.), 9.0 (t, 1H, NH disappearing on deuteration), 14.0 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 375 [M^+]. *Anal.* ($C_{17}H_{12}N_5O_2F_3$): C, H, N.

*3.1.19. 6-1(3-Trifluoromethylbenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**17**)*

IR (nujol, cm^{-1}): 3260, 3120, 3080, 1680, 1660. ^1H NMR (DMSO/TMS): δ 3.8 (d, 2H, CH_2), 6.7–8.0 (m, 4H arom. and 4H pyr.), 8.9 (t, 1H, NH disappearing on deuteration), 13.9 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 375 [M^+]. *Anal.* ($C_{17}H_{12}N_5O_2F_3$): C, H, N.

*3.1.20. 6-[(4-Trifluoromethylbenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**18**)*

IR (nujol, cm^{-1}): 3260, 3120, 3080, 1680, 1660. ^1H NMR (DMSO/TMS): δ 4.5 (d, 2H, CH_2), 7.5–8.9 (m, 4H arom. and 4H pyr.), 9.8 (t, 1H, NH disappearing on deuteration), 14.9 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 375 [M^+]. *Anal.* ($C_{17}H_{12}N_5O_2F_3$): C, H, N.

*3.1.21. 6-[(2-Bromobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**19**)*

IR (nujol, cm^{-1}): 3190, 3120, 3060, 1680, 1670. ^1H NMR (DMSO/TMS): δ 3.9 (d, 2H, CH_2), 6.4–8.1 (m, 4H arom. and 4H pyr.), 9.0 (t, 1H, NH disappearing on deuteration), 14.2 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 385 [M^+], 387. *Anal.* ($C_{16}H_{12}N_5O_2Br$): C, H, N.

*3.1.22. 6-[(3-Bromobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**20**)*

IR (nujol, cm^{-1}): 3260, 3120, 3080, 1680, 1670. ^1H NMR (DMSO/TMS): δ 4.5 (d, 2H, CH_2), 7.2–8.8 (m, 4H arom. and 4H pyr.), 9.7 (t, 1H, NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 385 [M^+], 387. *Anal.* ($C_{16}H_{12}N_5O_2Br$): C, H, N.

*3.1.23. 6-[(4-Bromobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-one (**21**)*

IR(nujol, cm^{-1}): 3200, 3140, 3040, 1690, 1680. ^1H NMR (DMSO/TMS): δ 4.4(d, 2H, CH_2), 7.2–8.8 (m, 4H arom. and 4H pyr.), 9.7 (t, 1H, NH disappearing on deuteration), 14.8 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 385 [M^+], 387. *Anal.* ($C_{16}H_{12}N_5O_2Br$): C, H, N.

*3.1.24. 6-[(3-Nitrobenzylamino)carbonyl]-3-(pyridin-2-yl)-4H-1,2,4-triazin-5-one (**22**)*

IR (nujol, cm^{-1}): 3240, 3150, 3070, 1680, 1665. ^1H NMR (DMSO/TMS): δ 3.8 (d, 2H, CH_2), 6.8–7.9 (m, 4H arom. and 4H pyr), 9.0 (t, 1H, NH disappearing on deuteration), 14.0 (br.s., 1H, NH, disappearing on deuteration). MS: *m/z* 352 [M^+]. *Anal.* ($C_{16}H_{12}N_6O_4$): C, H, N.

3.2. Microbiology

The determination of antimycobacterial activity was performed by viable count employing the agar dilution method [10]. Midlebrook and Cohn 7H10 agar, supplemented with Midlebrook OADC enrichment, was used to prepare quadrant plates with serial dimethylsulfoxide twofold dilutions of the different chemicals tested.

We employed two strains of *Mycobacterium* spp.: *M. tuberculosis* reference strain $H_{37}Rv$ and *M. avium*, strain 485, from our bacterial collection, emulsified in diluting fluid containing 0.2% fatty acid free albumin and 0.02% polysorbate 80, pH 6.9.

Control plates were included with known antimycobacterial drugs; all plates were incubated at 35°C in 5% CO_2 from 3 to 4 weeks.

The MIC was defined as the lowest chemical dilution associated with at least a 99% reduction in the number of the visible colonies. The results are indicated in Table 2.

Table 2
Activity of the 4*H*-1,2,4-triazin-5-one derivatives **1**–**22** against *M. tuberculosis* $H_{37}Rv$ (MIC μg/ml) [a]

Comp.	MIC (μg/ml)
1	>32
2	8
3	16
4	32
5	32
6	32
7	>32
8	>32
9	>32
10	>32
11	>32
12	>32
13	32
14	>32
15	>32
16	>32
17	32
18	16
19	>32
20	>32
21	>32
22	>32

[a] *M. tuberculosis* strain was sensitive to isoniazid (5 μg disk), rifampicin (25 μg disk) and ofloxacin (30 μg disk).

4. Results and discussion

A series of 6-[(arylmethylenamino)carbonyl]-3-(pyridin-2-yl)-4*H*-1,2,4-triazin-5-one derivatives **1–22** (Table 1) have been synthesized with the aim to evaluate their antimycobacterial activity (Table 2) towards a strain of *M. tuberculosis* ($H_{37}Rv$) sensitive to isoniazid, rifampicin and ofloxacin and a strain of *M. avium* resistant to isoniazid and rifampicin. Only compounds **2–6**, **13**, **17** and **18** exhibit a moderate in vitro antimycobacterial activity, against the tested strain of *M. tuberculosis* $H_{37}Rv$, their MIC values ranging from 8 to 32 μg/ml. However, the activity of the compounds towards the tested strain of *M. avium* is very low or absent, because only compounds **2** and **16** produce a 90% reduction in the number of visible colonies at the 80 μg/ml concentration.

In order to evaluate the significance of the 2-pyridinecarboxamidrazone sequence constricted in the triazinone system with respect to the antimycobacterial activity, the search for new modified triazinone derivatives is in progress.

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