Available online at www.sciencedirect.com
SCIENCE DIRECT
Bioorganic & Medicinal Chemistry 14 (2006) 5850–5865
Bioorganic & Medicinal Chemistry

Synthesis and antimicrobial evaluation of new 2-substituted 5,7-di-*tert*-butylbenzoxazoles

Jarmila Vinsova,[a,*] Katerina Cermakova,[a] Alexandra Tomeckova,[a] Martina Ceckova,[b] Josef Jampilek,[c] Pavel Cermak,[d] Jiri Kunes,[a] Martin Dolezal[e] and Frantisek Staud[b]

[a]*Department of Inorganic and Organic Chemistry, Faculty of Pharmacy, Charles University, 500 05 Hradec Kralove, Czech Republic*
[b]*Department of Pharmacology and Toxicology, Faculty of Pharmacy, Charles University, 500 05 Hradec Kralove, Czech Republic*
[c]*Zentiva a.s., U kabelovny 130, 102 37 Prague, Czech Republic*
[d]*Department of Clinical Microbiology, University Hospital and Faculty of Medicine, 500 05 Hradec Kralove, Czech Republic*
[e]*Department of Pharmaceutical Chemistry and Drug Control, Faculty of Pharmacy, Charles University, 500 05 Hradec Kralove, Czech Republic*

Received 3 April 2006; revised 9 May 2006; accepted 15 May 2006
Available online 19 June 2006

Abstract—Various synthetic pathways of the 30 novel 2-substituted 5,7-di-*tert*-butylbenzoxazoles as new potential antimicrobial drugs are discussed. The 28 intermediates are described as well. The compounds were characterized by ^{1}H and ^{13}C NMR spectra, MS spectra, IR/UV spectra and by means of CHN analysis. The purity of the final compounds was checked by HPLC and their lipophilicity ($\log K$) was also determined by means of RP-HPLC. In the present study, the correlation between RP-HPLC retention parameter $\log K$ (the logarithm of capacity factor K) and various calculated $\log P$ data is shown. The target compounds were tested for their in vitro antimycobacterial activity. Several compounds showed antituberculotic activity comparable with or higher than the standard isoniazide. In vitro cytotoxicity testing of the most active benzoxazoles and isoniazide as a reference drug was performed using MTT assay and compared with isoniazide as a reference drug. Structure–activity relationships among the chemical structures, the physical properties and the biological activities of the evaluated compounds are discussed in the article.

1. Introduction

Tuberculosis (TB), the world's leading infectious disease, caused by *Mycobacterium tuberculosis* represents a major global health problem. One-third of the world's population is currently infected; more than 5000 people die of TB every day. A great number of people are carriers of the latent form that creates dangerous source of illness for the future. Therefore, there is an urgent need to develop new structural classes of anti-tuberculosis drugs. The development stimulates also a problem of multi-drug resistant strains (MDR-TB) that challenge preparation of new type of compounds with unique mechanism of action, different from antitubercular drugs currently used.

Benzoxazoles belong to biologically very active skeletons.[1] Various benzoxazole derivatives were extensively studied for their antibacterial and antifungal activity[2–5] anticancer activity[6,7] also as new non-nucleoside *topoisomerase I* poisons[8] and *HIV-1 reverse transcriptase* inhibitors.[9,10]

Benzoxazoles are also interesting fluorescent probes which show high Stokes shift and present thermal and photophysical stability due to an excited state intramolecular proton transfer mechanism.[11,12] Since they interfere with biosynthesis of coloured carotenoids by inhibiting the enzyme phytoene desaturase, they are studied as potential bleaching herbicides.[13]

Benzoxazoles can be considered as structural bioisosters of naturally occurring nucleotides such as adenine and guanine, which allow them to interact easily with the biopolymers of a living system. They have shown low toxicity in warm-blooded animals.[14] In a previous paper we reported the synthesis of a series of lipophilic 2-substituted 5,7-di-*tert*-butylbenzoxazoles, which were

Keywords: 5,7-Di-*tert*-butylbenzoxazole; In vitro antimycobacterial activity; Lipophilicity determination; Cytotoxicity; Structure–activity relationships.
* Corresponding author. Tel.: +420 49 5067343; fax: +420 49 5514330; e-mail: vinsova@faf.cuni.cz

0968-0896/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmc.2006.05.030

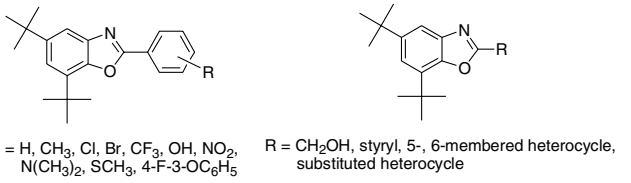


Figure 1. General formula of the target 2-substituted 5,7-di-*tert*-butylbenzoxazoles.

prepared by the reaction of 3,5-di-*tert*-butyl-1,2-benzoquinone with amino acids and dipeptides carrying *N*-terminal glycine.[15] They have shown promising activity against *M. tuberculosis* and some non-tuberculous strains where isoniazide, the first line antituberculous drug, has been inactive. This fact has prompted us to turn our attention to the preparation of a new lipophilic series of 5,7-di-*tert*-butylbenzoxazoles substituted in the $C_{(2)}$ position with aromatic substituent. The first series of benzoxazoles possess in the $C_{(2)}$ position benzene ring substituted by various electron-accepting or donor properties and lipophilic–hydrophilic balance, see Figure 1. In particular the electronegative substituents OH, Cl, Br and the most electron-withdrawing groups NO_2, CF_3 were compared with unsubstituted terms or with those bearing electron-releasing groups such as CH_3. The substitution on the benzene ring was chosen in accordance with Topliss.[16] The other series of prepared compounds possess in the $C_{(2)}$ position of benzoxazole the heterocyclic moiety, see Figure 1. Tertiary butyl groups increase lipophilicity of the molecule which is very important for passing through the extraordinary thick and tight mycobacterial cell wall. Unusually low cell membrane permeability also contributes to the resistance to therapeutic agents.

One of the major prerequisites for pharmacological screening and drug development is the prediction of permeability, that is the transport of a molecule through cellular membranes. Drugs cross biological barriers most frequently through passive transport, which strongly depends on their lipophilicity. Therefore, hydrophobicity is one of the most important physical properties of biologically active compounds. This thermodynamic parameter describes the partitioning of a compound between an aqueous and an organic phase and is characterized by the partition coefficient ($\log P$). Reversed phase high performance liquid chromatography (RP-HPLC) methods have become popular and are widely used for lipophilicity measurement. The general procedure consists of the measurement of the directly accessible retention time under isocratic conditions with varying amounts of an organic modifier in the mobile phase using end-capped non-polar C_{18} stationary RP columns and calculating the capacity factor K. $\log K$, calculated from the capacity factor K, is used as the lipophilicity index converted to $\log P$ scale.[17] The hydrophobicity evaluation of the target compounds by means of RP-HPLC and computational techniques is reported in this communication.

This presented study is a follow-up paper to the previous articles dealing with the *N*-heterocyclic derivatives as potential drugs.[18–20]

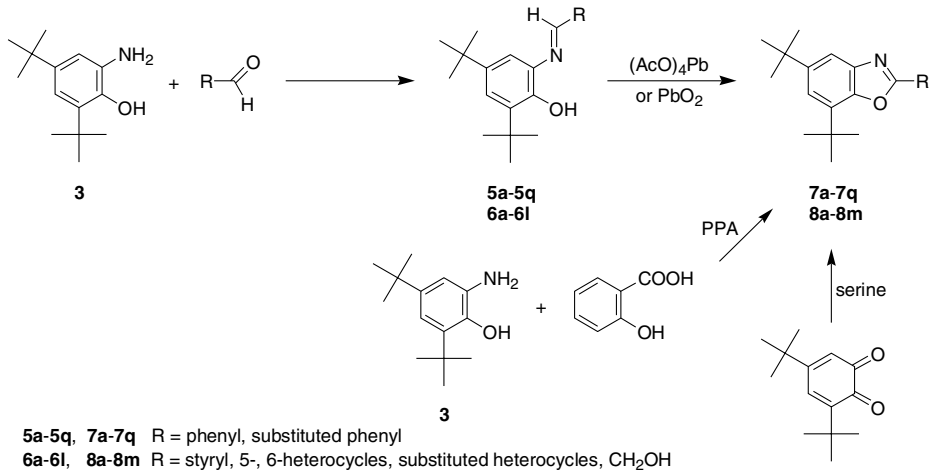
2. Results and discussion

2.1. Chemistry

There are three general methods for synthesis of target 2-substituted benzoxazoles: (i) consists of coupling 2-aminophenols with carboxylic acid derivatives, which is either catalyzed by strong acids[21] or requires microwave conditions;[22] (ii) is oxidative cyclization of phenolic Schiff bases obtained from 2-amino-4,6-di-*tert*-butylphenol (**3**) condensation with the appropriate aromatic aldehydes; (iii) is the reaction of 3,5-di-*tert*-butyl-1,2-benzoquinone (DTBBQ) with amino acids.[15]

2-Amino-4,6-di-*tert*-butylphenol (**3**) is not commercially available and was therefore synthesized via several possible procedures. Nitration of 2,4-di-*tert*-butylphenol gave according to TLC a mixture of two yellow components. The large excess of 2,4-di-*tert*-butyl-6-nitrophenol (**1**), the less polar compound, was isolated by continuous column chromatography on silica gel with hexane as eluent. The second more polar compound was identified as 2-*tert*-butyl-4,6-dinitrophenol (**2**). Hydrogenation of nitrophenol **1** on 5% Pd/C gave starting 2-amino-4,6-di-*tert*-butylphenol (**3**). Further synthetic pathways of aminophenol **3** generation, deamination of ethylene-1,2-diamine with 3,5-di-*tert*-butyl-1,2-benzoquinone in boiling propan-2-ol[23], gave on the contrary to the good yields presented in the literature[23] a mixture of required aminophenol **3** with fluorescent 5,7,5′,7′-tetra-*terc*-butyl-2,2′-bi-benzoxazole (**4**). More convenient one-pot preparation was reduction of 3,5-*tert*-butyl-1,2-benzoquinone monoimine by means of complex hydride $NaBH_4$.[24]

Synthesis of 2-substituted 5,7-di-*tert*-butylbenzoxazoles **7a**–**7q** and **8a**–**8m** is shown in Scheme 1. The majority of the target compounds were prepared via cyclization of the Schiff bases (method A), which were generated by the reaction of aminophenol **3** with the appropriate aromatic aldehydes. Schiff bases **5a**–**5q** and **6a**–**6l** were isolated and characterized. The target phenyl substituted derivatives **7a**–**7q** were obtained by means of **5a**–**5q** catalytic cyclization using PbO_2 in glacial AcOH, whereas the final heterocyclic substituted derivatives **8a**–**8l** were prepared through cyclization of **6a**–**6l** under $(AcO)_4Pb$ catalysis in glacial AcOH. (5,7-Di-*tert*-butyl-benzoxazol-2-yl)-methanol (**8m**) was obtained by the reaction of serine with DTBBQ,[21] method C. Direct condensation by PPA (method B) was tried out only for the preparation of 2-(5,7-di-*tert*-butylbenzoxazol-2-yl)-phenol (**7l**) with a much lower yield (15%) than by classical Shiff base oxidative cyclization (75%), method A. Straightforward method taking aminophenol **3** and carboxylic acid in the presence of polyphosphoric acid (PPA)[21] has not been found optimal.



Scheme 1. Synthesis of the target 2-substituted 5,7-di-*tert*-butylbenzoxazoles **7a**–**7q** and **8a**–**8m**.

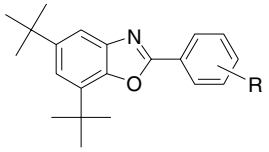
2.2. Lipophilicity

Hydrophobicities (log *P*/C log *P* values) of the studied compounds **7a**–**7q** and **8a**–**8m** were calculated using two commercially available programs and measured by means of RP-HPLC determination of capacity factors *K* with a subsequent calculation of log *K*. The results are shown in Tables 1, 2 and illustrated in Figures 2 and 3.

The results show that the experimentally determined log *K* values correlate relatively poorly with all computed lipophilicity data using ChemOffice software or ACD/Log P program. Nevertheless, log *P* values calculated by the ACD/Log P program agree better with target benzoxazoles **8a**–**8m**. All the showed differences between experimental and calculated lipophilicity values are probably caused by interactions of the substituents with heteroatoms of benzoxazole nucleus in individual compounds.

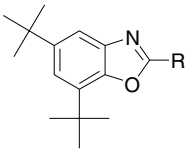
5,7-Di-*tert*-butyl-2-(2-nitrophenyl)-benzoxazole (**7m**) possesses the lowest lipophilicity, as expected. The phenolic derivative **7l** shows the highest lipophilicity in contrast with our expectations, see Figure 2. Great differences between experimental log *K* value and all computed data log *P*/C log *P* were also observed for

Table 1. Comparison of the determined log *K* values with the calculated lipophilicities (log *P*/C log *P*) of the synthesized ring substituted 2-phenyl-5,7-di-*tert*-butylbenzoxazoles **7a**–**7q**



Compound	R	log *K*	log *P*/C log *P* ChemOffice	log *P* ACD/Log P
7a	H	0.7515	6.35/7.165	7.39 ± 0.59
7b	3-Me	0.8370	6.83/7.664	7.85 ± 0.59
7c	4-Me	0.8350	6.83/7.664	7.85 ± 0.59
7d	3-Cl	0.9254	6.90/7.8948	8.18 ± 0.60
7e	4-Cl	0.9118	6.90/7.8948	8.15 ± 0.60
7f	2-Br	1.0021	7.17/7.7448	7.74 ± 0.63
7g	3-Br	1.0005	7.17/8.0448	8.36 ± 0.63
7h	4-Br	0.9981	7.17/8.0448	8.33 ± 0.63
7i	2-CF_3	0.8507	7.27/8.0774	8.36 ± 0.62
7j	3-CF_3	0.8487	7.27/8.0774	8.44 ± 0.62
7k	4-CF_3	0.8696	7.27/8.0774	8.36 ± 0.62
7l	2-OH	1.0221	5.96/6.1888	7.66 ± 0.61
7m	2-NO_2	0.5331	5.00/3.873	6.89 ± 0.60
7n	4-NO_2	0.7543	5.04/3.873	7.35 ±0.60
7o	4-$N(CH_3)_2$	0.7555	6.63/7.4398	7.81 ± 0.60
7p	4-SCH_3	0.8445	6.79/7.76	8.23 ± 0.61
7q	4-F-3-OC_6H_5	0.9245	8.04/9.2228	9.81 ± 0.66

Table 2. Comparison of the determined log *K* values with the calculated lipophilicities (log *P*/C log *P*) of the prepared compounds **8a**–**8m**



Compound	R	log *K*	log *P*/C log *P*ChemOffice	log *P*ACD/Log P
8a	Styryl	0.8325	7.13/7.759	7.93 ± 0.59
8b	1*H*-Pyrazol-3-yl	0.4065	4.87/5.669	5.29 ± 0.61
8c	Pyridin-2-yl	0.5044	5.43/5.986	6.31 ± 0.60
8d	Pyridin-3-yl	0.5726	6.14/6.485	6.77 ± 0.60
8e	pyridin-4-yl	0.6473	5.01/5.776	6.13 ± 0.60
8f	6-CH_3-pyridin-2-yl	0.5726	6.14/6.485	6.77 ± 0.60
8g	1*H*-Indol-3-yl	0.6064	5.89/7.299	7.31 ± 0.84
8h	Furan-2-yl	0.6234	4.84/6.551	6.18 ± 0.61
8i	5-CH_3-furan-2-yl	0.6783	5.30/7.050	6.64 ± 0.61
8j	5-C_2H_5-furan-2-yl	0.6972	5.78/7.579	7.17 ± 0.61
8k	Thiophen-2-yl	0.6723	6.33/7.060	6.99 ± 0.61
8l	Thiophen-3-yl	0.6870	6.27/6.8500	7.07 ± 0.84
8m	—CH_2OH	0.3281	4.37/3.824	4.40 ± 0.60

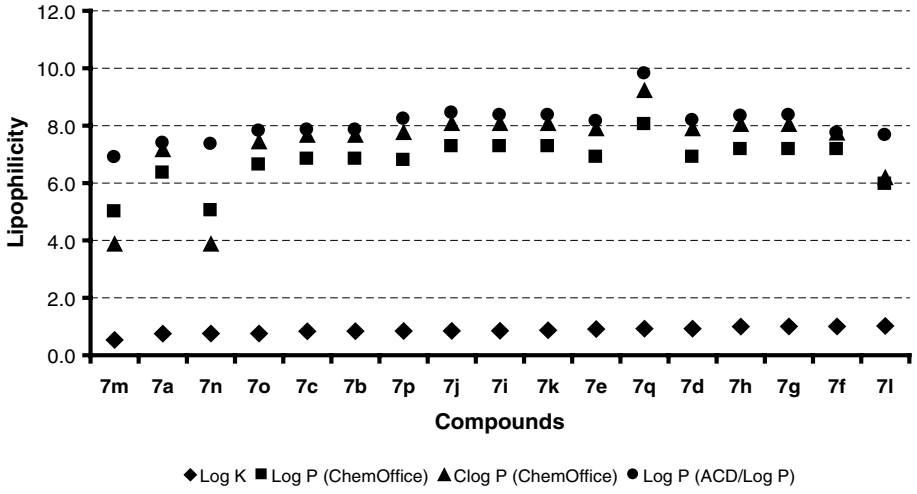


Figure 2. Comparison of the calculated log *P*/C log *P* data using the two programs with the experimentally found log *K* values of compounds **7a**–**7q**.

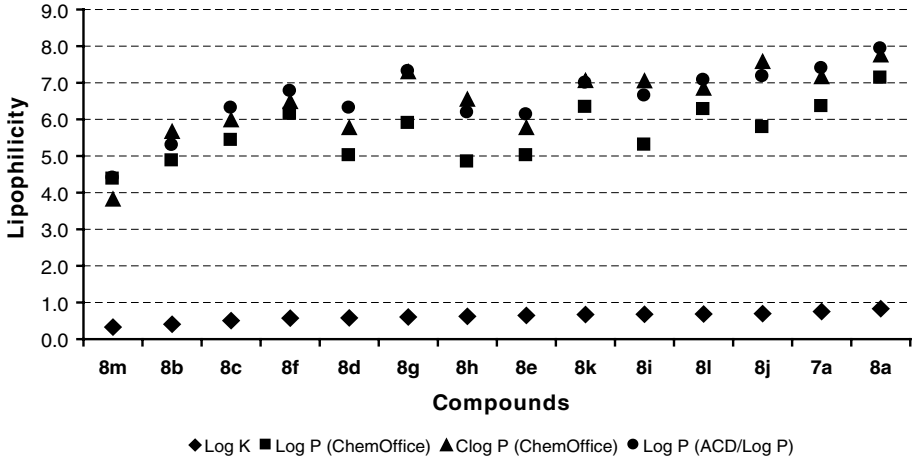


Figure 3. Comparison of the calculated log *P*/C log *P* data using the two programs with the experimentally found log *K* values of compounds **8a**–**8l** and **7a**.

5,7-di-*tert*-butyl-2-(4-fluoro-3-phenoxyphenyl)-benzoxazole (**7q**). According to the calculations compound **7q** shows the highest lipophilicity of all compounds in Figure 2, but according to the experiment this disubstituted derivative shows lipophilicity comparable with chloro derivatives **7e** and **7d**, it seems to be much more hydrophilic.

The program ChemOffice does not resolve $C_{(2)}/C_{(3)}/C_{(4)}$ substitutions in phenyl ring. According to the program ACD/Log P lipophilicity of the compounds substituted by CF_3, Cl, Br groups in phenyl ring increases $C_{(2)} < C_{(4)} < C_{(3)}$, but experimental log *K* for these benzoxazole derivatives increases $C_{(4)} < C_{(3)} < C_{(2)}$. This fact is not in good agreement with calculated data according to the program ACD/Log P. On the other hand, position derivatives of CH_3 or NO_2 moieties possess similar log *P* (ACD/Log P) to log *K*. As mentioned above, great differences were found for 2-OH moiety probably because of intramolecular interactions of $C_{(2)}$ phenolic moiety with heteroatoms of benzoxazole nucleus.

Figure 3 shows that (5,7-di-*tert*-butyl-benzoxazol-2-yl)-methanol (**8m**) possesses the lowest log *K*, whereas styryl derivative **8a** possesses the highest lipophilicity, as expected. The pyrazole derivative **8b** shows the lowest lipophilicity parameter log *K* of all heteroaromatic substituents, whereas 5,7-di-*tert*-butyl-2-(5-ethylfuran-2-yl)-benzoxazole (**8j**) possesses the highest lipophilicity among heterocycle substituted benzoxazoles **8b**–**8l** discussed in Figure 3. Great differences between log *K* and calculated log *P*/C log *P* were also observed for **8d**, **8e** and **8g**.

As expected, the dependence between log *K* and the length of the alkyl substituents in compounds **8h**–**8j** and **8c**, **8f** (H, CH_3, C_2H_5) is approximately linear. Log *K* values of 2-pyridinyl **8c**, 3-pyridinyl **8d** and 4-pyridinyl **8e** derivatives show also approximate linearity as well as 2-thiophenyl **8k** and 3-thiophenyl **8l** derivatives. The 4-pyridinyl derivative **8e** is the most lipophilic compound of the pyridinyl benzoxazoles in contrast to computed data. Great differences were observed for lipophilicity of 6-methylpyridin-2-yl derivative **8f**. According to the computed data this compound shows the highest lipophilicity of pyridine series, but in accordance with log *K* is less lipophilic than 5,7-di-*tert*-butyl-2-(pyridin-3-yl)-benzoxazole (**8d**), see Figure 3. Contrary to our expectations, all computed data for 5,7-di-*tert*-butyl-2-(1*H*-indole-3-yl)-benzoxazole (**8g**) show great differences from experimental log *K* value. According to the calculations, the compound **8g** shows the highest lipophilicity of all heterocyclic substituents, but in accordance with the experiment the compound **8g** shows medium lipophilicity in the series of benzoxazoles **8a**–**8m**.

2.3. Biological activity

All the prepared benzoxazole derivatives were tested for their antituberculotic activity at the GWL Hansen's Disease Center (Colorado State University) within the Tuberculosis Antimicrobial Acquisition and Coordinating Facility (TAACF) screening program for the discovery of novel drugs for treatment of TB. Primary screening was conducted at a single concentration, 6.25 μg/mL against *M. tuberculosis* $H_{37}Rv$ (ATCC2729) in BACTEC 12B medium using a broth microdilution assay, the Microplate Almar Blue Assay (MABA). Compounds affecting >90% inhibition in the primary screening (MIC < 6.25 μg/mL) were evaluated further. The active compounds were re-tested by serial dilution beginning at the concentration of 6.25 μg/mL against *M. tuberculosis* $H_{37}Rv$ to determine the actual minimum concentration (MIC) in the BACTEC 460 radiometric system and BACTEC 12B medium. The MIC is defined as the lowest concentration affecting a reduction in fluorescence of 90% relative to controls.

The results are reported in Table 3 and show an interesting activity for several compounds. Compounds **7f**, **7h**, **7l**, **7o**, **8a**–**8c**, **8e** exhibited an inhibitory effect against *M. tuberculosis* at a concentration of 6.25 μg/mL or lower in the range 50–100%. Five most active compounds **7f**, **7h**, **8a**, **8c**, **8e** were evaluated to determine the MIC and their activity against opportunistic pathogen *Mycobacterium kansasii* and two strains of *Mycobacterium avium* in comparison with isoniazide (INH).[15] Results of the latter tests are also reported in Table 3. Schiff bases **5f**, **5h**, **6a**, **6c**, **6e** of the most active benzoxazoles showed much lower activity against *M. tuberculosis*, the range 14–21% indicates benzoxazole structure's necessity.

Table 3. In vitro biological properties of the selected 2-substituted 5,7-di-*tert*-butylbenzoxazoles: antimycobacterial activities against *Mycobacterium tuberculosis* and atypical strains *Mycobacterium kansasii* and *Mycobacterium avium* in comparison with standard isoniazide (INH) as well as cytotoxicity of the compounds on HCT-8 cells

Compound	*M. tuberculosis* $H_{37}Rv$		*M. kansasii* 235/80			*M. avium* 80/72		*M. avium* 152/74		Cytotoxicity IC_{50} (μg/mL)
	MIC (μg/mL)	Inhibition (%)	7d	14d	21d	21d	28d	21d	28d	
7f	6.25	99	>128	>128	>128	>128	>128	>128	>128	53.6
7h	6.25	94	>128	>128	>128	>128	>128	>128	>128	80.4
7l	>6.25	70	—	—	—	—	—	—	—	—
7o	>6.25	54	—	—	—	—	—	—	—	—
8a	3.13	100	>128	>128	>128	>128	>128	>128	>128	902.2
8b	>6.25	72	—	—	—	—	—	—	—	—
8c	>6.25	79	32	64	64	64	64	64	64	32.9
8e	6.25	100	32	32	32	32	32	32	32	39.3
INH	0.02	100	>128	>128	>128	>128	>128	>128	>128	>100

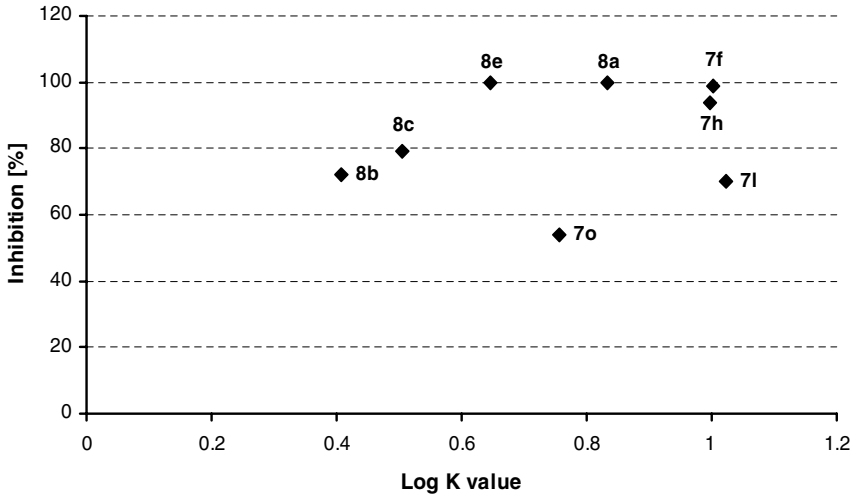


Figure 4. Dependence between growth inhibition (%) of *M. tuberculosis* $H_{37}Rv$ and the logarithm of the retention factor ($\log K$) of the studied most active compounds.

The cytotoxicity to human intestinal cells HCT-8 of the most active benzoxazoles **7f**, **7h**, **8a**, **8c**, **8e** and isoniazide as a reference drug was determined using MTT assay. The IC_{50} values of all the evaluated compounds are shown in Table 3. Among the tested compounds, **8e** and **8c** were the most toxic to the HCT-8 cells with low IC_{50} values. In contrast, the compound **8a** was the least toxic, revealing considerably higher IC_{50} value than the others. Toxicity of isoniazide to HCT-8 cells was not observed up to a concentration of 100 μg/mL. These data show for the most active styryl derivative **8a** MIC 3.13 μg/mL and the lowest toxicity to human intestinal cells HCT-8 (IC_{50} = 902.2 μg/mL).

From analysis of the data reported in Table 3 it can be generally deduced that bromine substituent in positions $C_{(2)}$ and $C_{(4)}$ has been found as the most active phenyl derivative. Hydrophilic electron-withdrawing groups CF_3, NO_2, OH, SCH_3 or electron-releasing group CH_3, $N(CH_3)_2$ with both lipophilic and hydrophilic properties are ineffective. 5,7-Di-*tert*-butyl-2-(pyridin-4-yl)-benzoxazole (**8e**) has been found as the most efficient heterocyclic derivative with 100% of inhibition of *M. tuberculosis* growth at 6.25 μg/mL as well as with the best activity also against *M. kansasii* and both strains of *M. avium*. In the light of the above results we can conclude that the 5,7-di-*tert*-butyl-benzoxazoles appear to be useful for antitubercular agents, the most active **8a** has even the lowest cytotoxicity. The highest cytotoxicity was found to correlate with the activity against nontuberculotic mycobacterial strains.

Figure 4 describes the dependence between in vitro growth inhibition (%) of *M. tuberculosis* and the logarithm of the retention factor ($\log K$) of the most active compounds **7f**, **7h**, **7l**, **7o** and **8a**–**8c**, **8e**. Generally, it could be assumed that the higher lipophilicity of the compounds results in positive effect for antituberculotic activity, but a straight correlation between the activity and lipophilicity of 2-substituted 5,7-di-*tert*-butylbenzoxazoles was not found.

3. Conclusion

The 30 2-substituted 5,7-di-*tert*-butylbenzoxazoles were prepared by means of various synthetic pathways. In addition, 28 intermediates were described. All final compounds and some representative Schiff bases were characterized by analytical and spectroscopic data. Lipophilicity of the final compounds was determined by means of RP-HPLC and the correlation between RP-HPLC retention parameter $\log K$ and $\log P$ data calculated in various ways was discussed. Several compounds showed high antituberculotic activity. 5,7-Di-*tert*-butyl-2-(pyridin-4-yl)-benzoxazole (**8e**) and especially 5,7-di-*tert*-butyl-2-styrylbenzoxazole (**8a**) showed the highest in vitro antituberculotic activities. The antimycobacterial activity seems to be independent of lipophilicity and dependent on chemical structure.

4. Experimental

4.1. Chemistry

4.1.1. Instrumentation and chemicals. The chemicals were purchased from Aldrich. Melting points (uncorrected) were determined on a Kofler block. Elemental analyses were performed on an automatic microanalyser CHNS-O CE instrument (FISONS EA 1110, Milano, Italy). UV spectra (λ, nm) of intermediates were measured on a Polarimeter ADP 220 (BS Bellingham Stanley Ltd) in ethanol. UV spectra (λ, nm) of the final compounds were determined on a Waters Photodiode Array Detector 2996 (Waters Corp., Milford, MA, USA) in ca 8×10^{-4} mol methanolic solution and $\log \varepsilon$ (the logarithm of molar absorption coefficient ε) was calculated for the absolute maximum λ_{max} of individual target compounds. Infrared spectra were recorded in Nicolet Impact 400 spectrometer in KBr pellets, Nujol mulls or $CHCl_3$ solutions. NMR spectra were measured in $CDCl_3$ or DMSO-d_6 solutions at ambient temperature on a Varian Mercury-VxBB 300

spectrometer (299.95 MHz for ^{1}H and 75.43 MHz for ^{13}C), Varian Comp. (Palo Alto, CA, USA). The chemical shifts δ are given in ppm related to tetramethylsilane (TMS) as internal standard. The coupling constants (*J*) are reported in Hz. Mass spectra were measured on ABI/MSD SCIEX API 3000™ LC/MS/MS System (MSD SCIEX, Concord, Ont., Canada). The reactions were monitored and the purity of the products was checked by TLC (Silufol UV 254, Kavalier Votice, Czech Republic and Merck TLC plates Silica gel 60 F_{254}, aluminium back) using two types of developing solvents S_1 (petroleum ether/EtOAc 9:1) and S_2 (toluene/EtOAc 4:1). The plates were visualized using UV light. Preparative thin-layer chromatography was carried out on 20 × 20 cm plates coated by silica gel. Silica gel 60 (0.015–0.040 mm, Merck, Darmstadt, Germany) was used for column chromatography.

The purity of the final compounds was checked by HPLC, see Section 2.2. The detection wavelength 210 nm was chosen. Peaks in the chromatogram of the solvent (blank) were deducted from peaks in the chromatogram of the sample solution. A purity of the individual compounds was determined from area peaks in the chromatogram of the sample solution.

4.1.2. General procedure for Schiff base synthesis (compounds 5a–5q and 6b–6l). The appropriately substituted aldehyde (0.01 mol) was added to the mixture of aminophenol **3** (0.01 mol) and boiled for 2 h. After evaporation, residue was crystallized from the mixture EtOH/water.

4.1.3. General procedure for benzoxazole synthesis (compounds 7a–7q and 8a–8m). *Method A—Schiff base cyclization.* $(AcO)_4Pb$ or PbO_2 (0.003 mol) was added to the mixture of the corresponding Schiff base (0.003 mol) in glacial AcOH (12 mL) and after 1 h stirring at the laboratory temperature the reaction mixture was evaporated, dissolved in EtOAc (25 mL) and extracted with water (25 mL), 5% $NaHCO_3$ (15 mL) and water (25 mL). EtOAc extract was dried over anhydrous Na_2SO_4 and evaporated to dryness. Purification was done by column chromatography on Silica gel using EtOAc/petroleum ether 1:9 as an eluent.

Method B—used for synthesis of ***7l***. The mixture of aminophenol **3** (0.01 mol) and salicylic acid (0.02 mol) was heated in PPA (12 g) while stirring for 2.5 h. At the end of the reaction period, the residue was poured into ice-water and neutralized with excess of 10% NaOH solution. After extraction with EtOAc, the EtOAc solution was dried over anhydrous Na_2SO_4 and evaporated under reduced pressure. The residue was recrystallized from EtOH/water.

Method C—used for synthesis of ***8m***. Serine (1 mmol) and DTBBQ (1 mmol) were dissolved in EtOH (50 mL, 60%) and heated for 5 h at 50 °C. The solvent was removed and the residue separated by column chromatography with mixture of EtOAc/petroleum ether in the appropriate ratio.

4.1.4. Data of prepared compounds

4.1.4.1. 2,4-Di-*tert*-butyl-6-nitrophenol (1). 2,4-Di-*tert*-butylphenol 19.2 g (0.093 mol) was dissolved in glacial AcOH (240 mL) and concd nitric acid (6.6 mL, 0.183 mol) was added with stirring at 25 °C. A rapid change from yellow to deep red was observed. After 50 min of stirring, water (150 mL) was added. $NaHCO_3$ was added to the orange solution until pH was raised to 5; a brown-red oily precipitate was extracted into $CHCl_3$, dried over anhydrous Na_2SO_4 and concentrated to the red oil. Continuous column chromatography (silica gel, hexane) separated the product mixture into two main components. Yield 90%. Anal. Calcd for $C_{14}H_{21}NO_3$ (251.32): 66.91% C, 8.42% H, 5.57% N. Found: 66.79% C, 8.18% H, 5.42% N. Mp 59–60 °C, mp 59–60 °C.[25] UV (nm), λ_{max}: 220, 289, 372. IR (KBr) cm^{-1}: 2955, 2911, 2871 (C–H), 1541 (N–O), 1460 (C=C), 1366 (CH_3), 1317, 1272, 1236, 1202, 1179, 1139, 924, 886, 773, 709, 641. MS: the peak 251.5 was found.

4.1.4.2. 2-*tert*-Butyl-4,6-dinitrophenol (2). Yield 14%. Anal. Calcd for $C_{10}H_{12}N_2O_5$ (240.22): 50.00% C, 5.04% H, 11.66% N. Found: 49.78% C, 4.98% H, 11.50% N. Mp 124–126 °C. Mp 124–125 °C.[26] TLC: R_f (S_1) 0.07. IR (KBr) cm^{-1}: 2975, 2907, 2880 (C–H), 1600, 1550, 1460, 1440 (C=C), 1395, 1365 (CH_3), 1335, 1270 (NO_2), 1150 (C–O), 935, 920, 735, 710 (Ar–H). MS: the peak 240.3 was found. ^{1}H NMR ($CDCl_3$), δ: 12.01 (s, 1H, OH), 8.96 (d, 1H, *J* = 2.80 Hz, Ar), 8.46 (d, 1H, *J* = 2.80 Hz, Ar), 1.50 (s, 9H, CH_3). ^{13}C NMR ($CDCl_3$), δ: 158.8, 142.8, 139.1, 133.0, 128.5, 119.5, 36.1, 29.0.

4.1.4.3. 2-Amino-4,6-di-*tert*-butylphenol (3). Prepared from 3,5-di-*tert*-butyl-1,2-benzoquinone (0.01 mol) by reaction with ammonia (20 mL) and subsequent addition of powdered $NaBH_4$ in MeOH (50 mL) according to the Ref. 27 Yield 80%. Anal. Calcd for $C_{14}H_{23}NO$ (221.35): 75.97% C, 10.47% H, 6.33% N. Found: 75.63% C, 10.15% H, 6.34% N. Mp 171–172 °C. Mp 170 °C.[28] TLC: R_f (S_1) 0.26. UV (nm), λ_{max}: 210, 287, 342. (KBr) cm^{-1}: 3395, 3383 (N–H), 2950, 2900, 2860 (C–H), 1590, 1482 (C=C), 1392, 1364 (CH_3), 909, 869, 833 (Ar–H). MS: the peak 221.5 was found. ^{1}H NMR ($CDCl_3$), δ: 6.90 (d, 1H, *J* = 2.30 Hz, Ar), 6.79 (d, 1H, *J* = 2.30 Hz, Ar), 1.40 (s, 9H, CH_3), 1.27 (s, 9H, CH_3). ^{13}C NMR ($CDCl_3$), δ: 144.2, 142.4, 135.5, 132.2, 116.0, 117.1, 34.7, 34.3, 31.7, 29.9.

4.1.4.4. 5,7,5′,7′-Tetra-*tert*-butyl-2,2′-bi-benzoxazole (4). Yield 13%. Anal. Calcd for $C_{30}H_{40}N_2O_2$ (460.66): 78.22% C, 8.75% H, 6.08% N. Found: 78.00% C, 8.74% H, 5.89% N. Mp 210–212 °C. TLC: R_f (S_2) 0.13. UV (nm), λ_{max}: 202, 241, 333. IR (KBr) cm^{-1}: 2961, 2908, 2871 (C–H), 1617 (C=N), 1482 (C=C), 1392, 1364 (CH_3), 909, 869, 833 (Ar–H). MS: the peak 460.8 was found. ^{1}H NMR ($CDCl_3$), δ: 7.80 (d, 2H, *J* = 1.92 Hz, Ar), 7.44 (d, 2H, *J* = 1.92 Hz, Ar), 1.58 (s, 18H, CH_3), 1.41 (s, 18H, CH_3). ^{13}C NMR ($CDCl_3$), δ: 151.9, 148.8, 147.4, 141.7, 134.6, 121.7, 115.3, 35.2, 34.6, 31.7, 30.0.

4.1.4.5. 2,4-Di-*tert*-butyl-6-[(phenylmethylene)amino]-phenol (5a). Yield 86%. Anal. Calcd for $C_{21}H_{27}NO$ (309.45): 81.51% C, 8.79% H, 4.53% N. Found: 81.43% C, 8.43% H, 8.67% N. Mp 85 °C. Mp 85 °C.[28] TLC: R_f (S_1) 0.46, R_F (S_2) 0.75. UV (nm), λ_{max}: 209, 271, 364. IR (KBr) cm^{-1}: 3351 (O–H), 3066, 3034 (=C–H), 2961, 2904, 2869 (C–H), 1624, 1577 (C=N), 1475 (C=C), 1391, 1363 (CH_3), 1075 (O–H), 874, 857, 751 (Ar–H). MS: the peak 309.6 was found.

4.1.4.6. 2,4-Di-*tert*-butyl-6-{[(3-methylphenyl)methylene]amino}-phenol (5b). Yield 65%. Anal. Calcd for $C_{22}H_{29}NO$ (323.48): 81.69% C, 9.04% H, 4.33% N. Found: 81.45% C, 8.98% H, 3.95% N. Mp 98–99 °C. TLC: R_f (S_1) 0.77, R_F (S_2) 0.82. UV (nm), λ_{max}: 206, 274, 361. IR (KBr) cm^{-1}: 3424 (O–H), 2955, 2904, 2866 (C–H), 1619, 1583 (C=N), 1478 (C=C), 1390, 1361 (CH_3), 1120 (O–H), 865, 788, (Ar–H). MS: the peak 323.6 was found.

4.1.4.7. 2,4-Di-*tert*-butyl-6-{[(4-methylphenyl)methylene]amino}-phenol (5c). Yield 79%. Anal. Calcd for $C_{22}H_{29}NO$ (323.48): 81.69% C, 9.04% H, 4.33% N. Found: 81.5% C, 8.88% H, 4.05% N. Mp 137–139 °C. TLC: R_f (S_1) 0.77, R_F (S_2) 0.82. UV (nm), λ_{max}: 206, 279, 357. IR (KBr) cm^{-1}: 3408 (O–H), 2954, 2903, 2866 (C–H), 1620, 1570 (C=N), 1480 (C=C), 1390, 1362 (CH_3), 1121 (O–H), 811, (Ar–H). MS: the peak 323.5 was found.

4.1.4.8. 2,4-Di-*tert*-butyl-6-{[(3-chlorophenyl)methylene]amino}-phenol (5d). Yield 75%. Anal. Calcd for $C_{21}H_{24}ClNO$ (341.87): 73.78% C, 7.08% H, 4.10% N. Found: 73.67% C, 6.87% H, 4.02% N. Mp 106–108 °C. TLC: R_f (S_1) 0.71, R_F (S_2) 0.93. UV (nm), λ_{max}: 208, 270, 371. IR (KBr) cm^{-1}: 3386 (O–H), 2955, 2904, 2866 (C–H), 1618, 1569 (C=N), 1478 (C=C), 1390, 1361 (CH_3), 1074 (O–H), 788, 683 (Ar–H). MS: the peak 341.9 was found.

4.1.4.9. 2,4-Di-*tert*-butyl-6-{[(4-chlorophenyl)methylene]amino}-phenol (5e). Yield 89%. $C_{21}H_{24}ClNO$ (341.87): 73.78% C, 7.08% H, 4.10% N. Found: 73.54% C, 7.17% H, 3.79% N. Mp 138–140 °C. TLC: R_f (S_1) 0.71, R_f (S_2) 0.92. UV (nm), λ_{max}: 210, 277, 364. IR (KBr) cm^{-1}: 3424 (O–H), 2958, 2905, 2868 (C–H), 1624, 1591 (C=N), 1478 (C=C), 1362 (CH_3), 1087 (O=H), 824, (Ar–H). MS: the peak 341.9 was found.

4.1.4.10. 2,4-Di-*tert*-butyl-6-{[(2-bromophenyl)methylene]amino}-phenol (5f). Yield 58%. Anal. Calcd for $C_{21}H_{26}BrNO$ (388.34): 64.95% C, 6.75% H, 3.61% N. Found: 65.68% C, 6.46% H, 3.33% N. Mp 122–124 °C. TLC: R_f (S_1) 0.75, R_F (S_2) 0.90. UV (nm), λ_{max}: 207, 275, 370. IR (KBr) cm^{-1}: 3374 (O–H), 2953, 2906, 2865 (C–H), 1614, 1560 (C=N), 1480 (C=C), 1387, 1361 (CH_3), 1027 (O–H), 867 (Ar–H). MS: the peak 388.5 was found.

4.1.4.11. 2,4-Di-*tert*-butyl-6-{[(3-bromophenyl)methylene]amino}-phenol (5g). Yield 56%. Anal. Calcd for $C_{21}H_{26}BrNO$ (388.34): 64.95% C, 6.75% H, 3.61% N. Found: 65.18% C, 6.76% H, 3.56% N. Mp 124–126 °C. TLC: R_f (S_1) 0.71, R_F (S_2) 0.93. UV (nm), λ_{max}: 210, 270, 369. IR (KBr) cm^{-1}: 3393 (O–H), 2955, 2904, 2866 (C–H), 1619, 1562 (C=N), 1480 (C=C), 1390, 1362 (CH_3), 1068 (O–H), 786 (Ar–H). MS: the peak 388.4 was found.

4.1.4.12. 2,4-Di-*tert*-butyl-6-{[(4-bromophenyl)methylene]amino}-phenol (5h). Yield 90%. Anal. Calcd for $C_{21}H_{26}BrNO$ (388.34): 64.95% C, 6.75% H, 3.61% N. Found: 64.83% C, 6.38% H, 3.54% N. Mp 144–146 °C. TLC: R_f (S_1) 0.64, R_F (S_2) 0.87. UV (nm), λ_{max}: 208, 279, 369. IR (KBr) cm^{-1}: 3416 (O–H), 2957, 2904, 2867 (C–H), 1623, 1566 (C=N), 1478 (C=C), 1390, 1361(CH_3), 1068 (O–H), 820 (Ar–H). MS: the peak 388.5 was found.

4.1.4.13. 2,4-Di-*tert*-butyl-6-{[(2-trifluoromethylphenyl)methylene]amino}-phenol (5i). Yield 73%. Anal. Calcd for $C_{22}H_{26}F_3NO$ (377.20): 70.01% C, 6.94% H, 3.71% N. Found: 69.78% C, 6.80% H, 3.80% N. Mp 83–85 °C. TLC: R_f (S_1) 0.51, R_f (S_2) 0.95. UV (nm), λ_{max}: 230, 268, 364. IR (KBr) cm^{-1}: 3355 (O–H), 2965, 2906, 2870 (C–H), 1625 (C=N), 1482 (C=C), 1393, 1364 (CH_3), 1332 (CF_3), 1067 (O–H), 866, 799 (Ar–H). MS: the peak 377.3 was found.

4.1.4.14. 2,4-Di-*tert*-butyl-6-{[(3-trifluoromethylphenyl)methylene]amino}-phenol (5j). Yield 38%. Anal. Calcd for $C_{22}H_{26}F_3NO$ (377.20): 70.01% C, 6.94% H, 3.71% N. Found: 69.81% C, 6.56% H, 3.53% N. Mp 89–94 °C. TLC: R_f (S_1) 0.78, R_f (S_2) 0.85. UV (nm), λ_{max}: 219, 268, 364. IR (KBr) cm^{-1}: 3405 (O–H), 2964, 2907, 2870 (C–H), 1625 (C=N), 1481 (C=C), 1393, 1363 (CH_3), 1332 (CF_3), 1068 (O–H), 866, 800 (Ar–H). MS: the peak 377.3 was found.

4.1.4.15. 2,4-Di-*tert*-butyl-6-{[(4-trifluoromethylphenyl)methylene]amino}-phenol (5k). Yield 64%. Anal. Calcd for $C_{22}H_{26}F_3NO$ (377.20): 70.01% C, 6.94% H, 3.71% N. Found: 70.06% C, 6.67% H, 3.46% N. Mp 124–126 °C. TLC: R_f (S_1) 0.80, R_f (S_2) 0.88. UV (nm), λ_{max}: 209, 230, 287, 378. IR (KBr) cm^{-1}: 3397 (O–H), 2960, 2906, 2869 (C–H), 1624 (C=N), 1479 (C=C), 1391, 1362 (CH_3), 1065 (O–H), 836, (Ar–H). MS: the peak 377.4 was found.

4.1.4.16. 2,4-Di-*tert*-butyl-6-{[(2-hydroxyphenyl)methylene]amino}-phenol (5l). Yield 78%. Anal. Calcd for $C_{21}H_{27}NO_2$ (325.45): 77.50% C, 8.36% H, 4.30% N. Found: 77.15% C, 8.06% H, 4.15% N. Mp 124–125 °C. Mp 114–115 °C.[29] TLC: R_f (S_1) 0.52, R_F (S_2) 0.82. UV (nm), λ_{max}: 207, 269, 358. IR (KBr) cm^{-1}: 3503 (O–H), 2957, 2906, 2869 (C–H), 1608, 1571 (C=N), 1479 (C=C), 1391, 1362 (CH_3), 1151 (O–H), 866, 756 (Ar–H). MS: the peak 325.5 was found.

4.1.4.17. 2,4-Di-*tert*-butyl-6-{[(2-nitrophenyl)methylene]amino}-phenol (5m). Yield 87%. Anal. Calcd for $C_{21}H_{26}N_2O_3$ (354.44): 71.16% C, 7.39% H, 7.90% N. Found: 71.06% C, 7.15% H, 7.96% N. Mp 127–128 °C. TLC: R_f (S_1) 0.32, R_f (S_2) 0.88. UV (nm), λ_{max}: 204, 275, 379. IR (KBr) cm^{-1}: 3425 (O–H), 2957, 2907, 2868

(C–H), 1610, 1569 (C=N), 1527 (NO_2), 1481 (C=C), 1390, 1362 (CH_3), 1345 (NO_2), 1065 (O–H), 864, 785, 744 (Ar–H). MS: the peak 354.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 9.16 (s, 1H, OH), 8.28–8.24 (m, 1H, H3′), 8.09–8.04 (m, 1H, H6′), 7.78–7.71 (m, 1H, H5′), 7.66–7.51 (m, 2H, 4H′, NCH), 7.33 (d, 1H, J = 2.06 Hz, H3), 7.23 (d, 1H, J = 2.06 Hz, H5), 1.46 (s, 9H, CH_3), 1.34 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 150.7, 149.4, 141.8, 135.7, 133.9, 133.4, 131.1, 130.8, 129.5, 124.8, 124.7, 110.3, 35.0, 34.6, 31.5, 29.4.

4.1.4.18. 2,4-Di-*tert*-butyl-6-{[(4-nitrophenyl)methylene]amino}-phenol (5n). Yield 77%. Anal. Calcd for $C_{21}H_{26}N_2O_3$ (354.44): 71.16% C, 7.39% H, 7.90% N. Found: 70.96% C, 7.08% H, 7.87% N. Mp 185–187 °C. Mp 169–170 °C.[29] TLC: R_f (S_1) 0.39, R_f (S_2) 0.87. UV (nm), λ_{max}: 202, 285, 346, 400. IR (KBr) cm^{-1}: 3424 (O–H), 2957, 2906, 2869 (C–H), 1623, 1599 (C=N), 1522 (NO_2), 1479 (C=C), 1390, 1362 (CH_3), 1343 (NO_2), 1107 (O–H), 842, 747 (Ar–H). MS: the peak 354.6 was found.

4.1.4.19. 2,4-Di-*tert*-butyl-6-{[(4-dimethylaminophenyl)methylene]amino}-phenol (5o). Yield 85%. Anal. Calcd for $C_{23}H_{32}N_2O$ (352.51): 78.36% C, 9.15% H, 7.95% N. Found: 78.34% C, 9.04% H, 7.75% N. Mp 156–158 °C. Mp 158–159 °C.[29] TLC: R_f (S_1) 0.32, R_F (S_2) 0.75. UV (nm), λ_{max}: 204, 242, 379. IR (KBr) cm^{-1}: 3425 (O–H), 2957, 2904, 2867 (C–H), 1598, 1552 (C=N), 1478 (C=C), 1363 (CH_3), 1166 (O–H), 816, 764 (Ar–H). MS: the peak 352.6 was found.

4.1.4.20. 2,4-Di-*tert*-butyl-6-{[(4-methylsulfanylphenyl)methylene]amino}-phenol (5p). Yield 68%. Anal. Calcd for $C_{22}H_{28}NOS$ (354.53): 74.53% C, 7.96% H, 3.95% N. Found: 74.33% C, 7.80% H, 3.84% N. Mp 139–140 °C. TLC: R_f (S_1) 0.32, R_F (S_2) 0.75. UV (nm), λ_{max}: 207, 324, 364. IR (KBr) cm^{-1}: 3443 (O–H), 2957, 2918, 2867 (C–H), 1618, 1592 (C=N), 1478 (C=C), 1390, 1361 (CH_3), 1062 (O–H), 877, 815 (Ar–H). MS: the peak 354.6 was found.

4.1.4.21. 2,4-Di-*tert*-butyl-6-{[(3-fluoro-4-phenoxyphenyl)methylene]amino}-phenol (5q). Yield 62%. Anal. Calcd for $C_{27}H_{30}FNO_2$ (419.54): 77.30% C, 7.21% H, 3.34% N. Found: 77.06% C, 6.89% H, 3.01% N. Mp 114 °C. TLC: R_f (S_1) 0.18, R_f (S_2) 0.94. UV (nm), λ_{max}: 204, 269, 361. IR (KBr) cm^{-1}: 3397 (O–H), 2957, 2905, 2867 (C–H), 1628, 1586 (C=N), 1509, 1490, 1425 (C=C), 1391, 1362 (CH_3), 1105 (O–H), 870, 818, 750 (Ar–H). MS: the peak 419.6 was found.

4.1.4.22. 2,4-Di-*tert*-butyl-6-[(3-phenylallylidene)amino]-phenol (6a). Yield 80%. Anal. Calcd for $C_{23}H_{29}NO$ (335.49): 82.34% C, 8.71% H, 4.18% N. Found: 82.30% C, 8.54% H, 3.97% N. Mp 143 °C. TLC: R_f (S_1) 0.56, R_f (S_2) 0.75. UV (nm), λ_{max}: 207, 306, 375. IR (KBr) cm^{-1}: 3259 (O–H), 2953, 2906, 2866 (C–H), 1625, 1586 (C=N), 1478 (C=C), 1395, 1360 (CH_3), 1162 (O–H), 865 (Ar–H). MS: the peak 335.6 was found.

4.1.4.23. 2,4-Di-*tert*-butyl-6-[(1*H*-pyrazol-3-ylmethylene)amino]-phenol (6b). Yield 80%. Anal. Calcd for $C_{18}H_{25}N_3O$ (299.41): 72.21% C, 8.42% H, 14.03% N. Found: 72.08% C, 8.06% H, 13.88% N. Mp 174–176 °C. TLC: R_f (S_1) 0.04, R_f (S_2) 0.32. UV (nm), λ_{max}: 201, 227, 273, 356. IR (KBr) cm^{-1}: 3428 (O–H), 2958, 2907, 2868 (C–H), 1629 (C=N), 1481 (C=C), 1391, 1362 (CH_3), 1052 (O–H), 868, 821, 794, 764 (Ar–H). MS: the peak 299.6 was found.

4.1.4.24. 2,4-Di-*tert*-butyl-6-[(pyridin-2-ylmethylene)-amino]-phenol (6c). Yield 56%. Anal. Calcd for $C_{20}H_{26}N_2O$ (310.44): 77.38% C, 8.44% H, 9.02% N. Found: 77.22% C, 8.04% H, 8.86% N. Mp 159–161 °C. TLC: R_f (S_1) 0.15, R_f (S_2) 0.35. UV (nm), λ_{max}: 209, 226, 295, 381. IR (KBr) cm^{-1}: 3424 (O–H), 2955, 2904, 2866 (C–H), 1623, 1590 (C=N), 1481 (C=C), 1389, 1363 (CH_3), 1151 (O–H), 881, 864 (Ar–H). MS: the peak 310.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.87 (s, 1H, NCH), 8.72 (ddd, 1H, J = 4.94 Hz, J = 1.65 Hz, J = 1.10 Hz, Ar), 8.22 (dt, 1H, J = 7.69 Hz, J = 1.10 Hz, Ar), 7.90-7.81 (m, 2H, Ar, OH), 7.39 (ddd, 1H, J = 7.69 Hz, J = 4.94 Hz, J = 1.10 Hz, Ar), 7.34–7.30 (m, 2H, Ar), 1.46 (s, 9H, CH_3), 1.33 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 155.1, 154.1, 149.4, 149.3, 141.6, 137.2, 135.7 133.5, 125.1, 124.7, 121.4, 110.3, 35.0, 34.6, 31.5, 29.4.

4.1.4.25. 2,4-Di-*tert*-butyl-6-[(pyridin-3-ylmethylene)-amino]-phenol (6d). Yield 80%. Anal. Calcd for $C_{20}H_{26}N_2O$ (310.44): 77.38% C, 8.44% H, 9.02% N. Found: 76.98% C, 8.32% H, 8.79% N. Mp 180–182 °C. TLC: R_f (S_1) 0.20, R_f (S_2) 0.65. UV (nm), λ_{max}: 228, 269, 375. IR (KBr) cm^{-1}: 3424 (O–H), 2957, 2906, 2868 (C–H), 1623 (C=N), 1481 (C=C), 1391, 1362 (CH_3), 1026 (O–H), 806 (Ar–H). MS: the peak 310.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 9.07 (d, 1H, J = 1.64 Hz, Ar), 8.75 (s, 1H, NCH), 8.71 (dd, 1H, J = 4.67 Hz, J = 1.64 Hz, Ar), 8.32 (dt, 1H, J = 7.97 Hz, J = 1.65 Hz, Ar), 7.66 (br s, 1H, OH), 7.46 (dd, 1H, J = 7.96 Hz, J = 4.67 Hz, Ar), 7.31 (d, 1H, J = 2.19 Hz, Ar), 7.22 (d, 1H, J = 2.20 Hz, Ar), 1.46 (s, 9H, CH_3), 1.35 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 152.1, 151.3 150.2, 149.0, 141.6, 135.6, 135.0 133.9, 131.9, 124.4, 124.0, 109.8, 35.0, 34.6, 31.6, 29.4.

4.1.4.26. 2,4-Di-*tert*-butyl-6-[(pyridin-4-ylmethylene)-amino]-phenol (6e). Yield 79%. Anal. Calcd for $C_{20}H_{26}N_2O$ (310.44): 77.38% C, 8.44% H, 9.02% N. Found: 77.40% C, 8.26% H, 8.76% N. Mp 138–140 °C. TLC: R_f (S_1) 0.08, R_f (S_2) 0.30. UV (nm), λ_{max}: 228, 290, 384. IR (KBr) cm^{-1}: 3424 (O–H), 2958, 2906, 2868 (C–H), 1636, 1601 (C=N), 1480 (C=C), 1391, 1362 (CH_3), 1052 (O–H), 814 (Ar–H). MS: the peak 310.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.79–8.74 (m, 2H, Ar), 8.69 (s, 1H, NCH), 7.82–7.77 (m, 2H, Ar), 7.70 (br s, 1H, OH), 7.34 (d, 1H, J = 2.00 Hz, Ar), 7.23 (d, 1H, J = 2.00 Hz, Ar), 1.45 (s, 9H, CH_3), 1.35 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 152.5, 150.1, 149.5, 143.1, 141.7, 135.9, 133.4, 125.1, 122.1, 109.9, 35.0, 34.6, 31.5, 29.3.

4.1.4.27. 2,4-Di-*tert*-butyl-6-{[(6-methylpyridin-2-yl)-methylene]amino}-phenol (6f). Yield 52%. Anal. Calcd for $C_{21}H_{28}N_2O$ (324.46): 77.74% C, 8.70% H, 8.63% N. Found: 77.56% C, 8.46% H, 8.35% N. Mp 176–178 °C. TLC: R_f (S_1) 0.11, R_f (S_2) 0.39. UV (nm), λ_{max}: 208, 299, 379. IR (KBr) cm^{-1}: 3406 (O–H), 2952, 2903, 2867 (C–H), 1624, 1595 (C=N), 1462 (C=C), 1388, 1362 (CH_3),1025 (O–H), 874 (Ar–H). MS: the peak 324.6 was found. ^1H NMR (300 MHz, $CDCl_3$), δ: 8.85 (s, 1H, NCH), 8.05 (d, 1H, J = 7.69 Hz, Ar), 7.83 (s, 1H, OH), 7.71 (t, 1H, J = 7.69 Hz, Ar), 7.34 (d, 1H, J = 2.20 Hz, Ar), 7.30 (d, 1H, J = 2.20 Hz, Ar), 7.24 (d, 1H, J = 7.69 Hz, Ar), 2.64 (s, 3H, CH_3), 1.46 (s, 9H, CH_3), 1.32 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 158.4, 156.0, 154.0, 149.2, 141.6, 136.9, 135.4, 133.6, 124.8, 124.4, 118.2, 110.3, 34.9, 34.6, 31.6, 29.4, 24.3.

4.1.4.28. 2,4-Di-*tert*-butyl-6-[(1*H*-indol-3-ylmethylene)-amino]-phenol (6g). Yield 35%. Anal. Calcd for $C_{23}H_{28}N_2O$ (348.49): 79.27% C, 8.10% H, 8.04% N. Found: 79.06% C, 7.87% H, 7.76% N. Mp 187–189 °C. TLC: R_f (S_1) 0.08, R_f (S_2) 0.60. UV (nm), λ_{max}: 207, 225, 271, 354. IR (KBr) cm^{-1}: 3435 (O–H), 2957, 2905, 2867 (C–H), 1618, 1577 (C=N), 1478, 1457 (C=C), 1362 (CH_3), 1106 (O–H), 867, 766, 746 (Ar–H). MS: the peak 348.6 was found. ^1H NMR (300 MHz, $CDCl_3$), δ: 8.86 (s, 1H, NCH), 8.55–8.43 (m, 2H, Ar), 7.68 (br s, 1H, OH), 7.46–7.39 (m, 1H, Ar), 7.37–7.28 (m, 2H, Ar), 7.23 (d, 1H, J = 2.20 Hz, Ar), 7.18 (d, 1H, J = 2.20 Hz, Ar), 1.49 (s, 9H, CH_3), 1.37 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 151.8, 147.8, 141.3, 136.9, 136.6, 134.6, 131.0, 124.9, 123.9, 122.3, 122.1, 121.7, 116.8, 111.4, 109.9, 34.9, 34.6, 31.7, 29.5.

4.1.4.29. 2,4-Di-*tert*-butyl-6-[(furan-2-ylmethylene)-amino]-phenol (6h). Yield 61%. $C_{19}H_{25}NO_2$ (299.41): 76.22% C, 8.42% H, 4.68% N. Found 75.97% C, 8.22% H, 4.44% N. Mp 88–90 °C. TLC: R_f (S_1) 0.47, R_f (S_2) 0.88. UV (nm), λ_{max}: 208, 289, 364. IR (KBr) cm^{-1}: 3424 (O–H), 2955, 2906, 2868 (C–H), 1629, 1574 (C=N), 1475 (C=C), 1391, 1361 (CH_3), 1017 (O–H), 884, 865, 820 (Ar–H). MS: the peak 299.5 was found.

4.1.4.30. 2,4-Di-*tert*-butyl-6-{[(5-methylfuran-2-yl)-methylene]amino}-phenol (6i). Yield 98%. Anal. Calcd for $C_{20}H_{25}NO_2$ (311.42): 76.64% C, 8.68% H, 4.47% N. Found: 76.25% C, 8.52% H, 4.14% N. Mp 98–100 °C. TLC: R_f (S_1) 0.35, R_f (S_2) 0.76. UV (nm), λ_{max}: 204, 249, 315. IR (KBr) cm^{-1}: 3405 (O–H), 2953, 2906, 2866 (C–H), 1623, 1577 (C=N), 1478 (C=C), 1389, 1362 (CH_3), 1023 (O–H), 873, 821, 802 (Ar–H). MS: the peak 311.5 was found.

4.1.4.31. 2,4-Di-*tert*-butyl-6-{[(5-ethylfuran-2-yl)methylene]amino}-phenol (6j). Yield 40%. $C_{21}H_{27}NO_2$(325.44): 77.02% C, 8.93% H, 4.28% N. Found: 76.84% C, 8.72% H, 3.96% N. Viscous oil. TLC: R_f (S_1) 0.51, R_f (S_2) 0.75. UV (nm), λ_{max}: 202, 316. IR (KBr) cm^{-1}: 3424 (O–H), 2965, 2906, 2869 (C–H), 1619, 1529 (C=N), 1479 (C=C), 1386, 1361 (CH_3), 1018 (O–H), 801 (Ar–H). MS: the peak 325.6 was found.

4.1.4.32. 2,4-Di-*tert*-butyl-6-[(thiophen-2-ylmethylene)-amino]-phenol (6k). Yield 62%. Anal. Calcd for $C_{19}H_{25}NOS$ (315.47): 72.34% C, 7.99% H, 4.44% N. Found: 72.01% C, 7.50% H, 4.07% N. Mp 90–92 °C. TLC: R_f (S_1) 0.68, R_F (S_2) 0.89. UV (nm), λ_{max}: 211, 275, 307, 372. IR (KBr) cm^{-1}: 3406 (O–H), 2957, 2905, 2867 (C–H), 1612 (C=N), 1480 (C=C), 1391, 1362 (CH_3), 1045 (O–H), 861, 813 (Ar–H). MS: the peak 315.6 was found. ^1H NMR (300 MHz, $CDCl_3$), δ: 8.80 (s, 1H, NCH), 7.67 (br s, 1H, OH), 7.53–7.49 (m, 2H, Ar), 7.25 (d, 1H, J = 2.20 Hz, Ar), 7.18 (d, 1H, J = 2.20 Hz, Ar), 7.14 (dd, 1H, J = 4.94 Hz, J = 3.85 Hz, Ar), 1.46 (s, 9H, CH_3), 1.35 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 148.6, 148.5, 143.2, 141.3, 135.2, 134,1, 131.9, 130.3, 127.9, 123.3, 109.9, 34.9, 34.6, 31.6, 29.4.

4.1.4.33. 2,4-Di-*tert*-butyl-6-[(thiophen-3-ylmethylene)-amino]-phenol (6l). Yield 60%. Anal. Calcd for $C_{19}H_{25}NOS$ (315.47): 72.34% C, 7.99% H, 4.44% N. Found: 72.40% C, 7.68% H, 4.12% N. Mp 68–71 °C. TLC: R_f (S_1) 0.79, R_F (S_2) 0.90. UV (nm), λ_{max}: 212, 249, 355. IR (KBr) cm^{-1}: 3424 (O–H), 2957, 2904, 2867 (C–H), 1618, (C=N), 1480 (C=C), 1391, 1362 (CH_3), 1074 (O–H), 867, 831 (Ar–H). MS: the peak 315.6 was found.

4.1.4.34. 5,7-Di-*tert*-butyl-2-phenylbenzoxazole (7a). Method A. Yield 57%. Anal. Calcd for $C_{21}H_{25}NO$ (307.43): 82.04% C, 8.20% H, 4.56% N. Found: 82.10% C, 8.26% H, 4.54% N. Mp 59–60 °C. HPLC purity 98.98%. TLC: R_f (S_1) 0.52, R_f (S_2) 0.80. UV (nm), λ_{max}/log ε: 207/3.68, 239, 296. IR (KBr) cm^{-1}: 2957, 2907, 2868 (C–H), 1624, 1557 (C=N), 1482 (C=C), 1391, 1362 (CH_3), 1060 (C–O–C), 863, 706 (Ar–H). MS: the peak 307.5 was found. ^1H NMR (300 MHz, $CDCl_3$), δ: 8.30–8.23 (m, 2H, Ar), 7.67 (d, 1H, J = 1.79 Hz, Ar), 7.57–7.50 (m, 3H, Ar), 7.32 (d, 1H, J = 1.79 Hz, Ar), 1.56 (s, 9H, CH_3), 1.41 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 162.5, 147.7, 146.9, 142.3, 133.7, 131.2, 128.9, 127.5, 127.4, 119.5, 114.2, 35.1, 34.5, 31.8, 30.0.

4.1.4.35. 5,7-Di-*tert*-butyl-2-(3-methylphenyl)-benzoxazole (7b). Method A. Yield 56%. Anal. Calcd for $C_{22}H_{27}NO$ (321.46): 82.20% C, 8.47% H, 4.36% N. Found: 82.14% C, 8.53% H, 4.29% N. Mp 69–72 °C. HPLC purity 99.92%. TLC: R_f (S_1) 0.77, R_f (S2) 0.87. UV (nm), λ_{max}/log ε: 204/3.62, 243, 305. IR (KBr) cm^{-1}: 2957, 2908, 2870 (C–H), 1608, 1592 (C=N), 1479 (C=C), 1401, 1363 (CH_3), 1076 (C–O–C), 868, 790, 723 (Ar–H). MS: the peak 321.6 was found. ^1H NMR (300 MHz, $CDCl_3$), δ: 8.11–8.02 (m, 2H, Ar), 7.66 (d, 1H, J = 1.92 Hz, Ar), 7.42 (t, 1H, J = 7.69 Hz, Ar), 7.37–7.32 (m, 1H, Ar), 7.31 (d, 1H, J = 1.92 Hz, Ar), 2.47 (s, 3H, CH_3), 1.56 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 162.7, 147.7, 146.9, 142.2, 138.7, 133.7, 132.1, 128.8, 127.9, 127.3, 124.6, 119.5, 114.1, 35.1, 34.5, 31.8, 30.0, 21.4.

4.1.4.36. 5,7-Di-*tert*-butyl-2-(4-methylphenyl)-benzoxazole (7c). Method A. Yield 42%. Anal. Calcd for $C_{22}H_{27}NO$ (321.46): 82.20% C, 8.47% H, 4.36% N. Found: 82.28% C, 8.40% H, 4.23% N. Mp 92–93 °C. HPLC purity 99.85%. TLC: R_f (S_1) 0.52, R_f (S_2) 0.78. UV (nm), λ_{max}/log ε: 215, 246, 293/3.63. IR (KBr) cm^{-1}: 2954, 2907, 2869 (C–H), 1612, 1561 (C=N), 1482 (C=C), 1392, 1364 (CH_3), 1068 (C–O–C), 867, 825, 730 (Ar–H). MS: the peak 321.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.19–8.11 (m, AA′BB′, 2H, Ar), 7.65 (d, 1H, J = 1.92 Hz, Ar), 7.37–7.31 (m, AA′BB′, 2H, Ar), 7.30 (d, 1H, J = 1.92 Hz, Ar), 2.44 (s, 3H, CH_3), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 162.7, 147.7, 146.8, 142.2, 141.7, 133.6, 129.6, 127.4, 124.7, 119.3, 114.0, 35.1, 34.5, 31.8, 30.0, 21.6.

4.1.4.37. 5,7-Di-*tert*-butyl-2-(3-chlorophenyl)-benzoxazole (7d). Method A. Yield 45%. Anal. Calcd for $C_{21}H_{24}ClNO$ (341.87): 73.78% C, 7.08% H, 4.10% N. Found: 73.69% C, 7.15% H, 4.04% N. Mp 116–117 °C. HPLC purity 99.81%. TLC: R_f (S_1) 0.79, R_f (S_2) 0.82. UV (nm), λ_{max}/log ε: 207/3.67, 308. IR (KBr) cm^{-1}: 2954, 2906, 2869 (C–H), 1625, 1554 (C=N), 1475 (C=C), 1392, 1365 (CH_3), 1059 (C–O–C), 864, 782, 719 (Ar–H). MS: the peak 342.0 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.24–8.21 (m, 1H, Ar), 8.14 (dt, 1H, J = 6.87 Hz, J = 1.65 Hz, Ar), 7.66 (d, 1H,J = 1.92 Hz, Ar), 7.50–7.43 (m, 2H, Ar), 7.33 (d, 1H, J = 1.92 Hz, Ar), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.1, 148.0, 146.9, 142.1, 134.9, 133.9, 131.2, 130.2, 129.2, 127.3, 125.4, 120.0, 114.3, 35.1, 34.5, 31.8, 30.0.

4.1.4.38. 5,7-Di-*tert*-butyl-2-(4-chlorophenyl)-benzoxazole (7e). Method A. Yield 39%. Anal. Calcd for $C_{21}H_{24}ClNO$ (341.87): 73.78% C, 7.08% H, 4.10% N. Found: 73.87% C, 7.02% H, 4.03% N. Mp 109–111 °C. HPLC purity 99.91%. TLC: R_f (S_1) 0.69, R_f (S_2) 0.90. UV (nm), λ_{max}/log ε: 204/3.64, 245, 309. IR ($CHCl_3$) cm^{-1}: 2957, 2908, 2869 (C–H), 1600, 1553 (C=N), 1483 (C=C), 1399, 1364 (CH_3), 1064 (C–O–C), 868, 837, 733 (Ar–H). MS: the peak 341.9 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.21–8.15 (m, AA′BB′, 2H, Ar), 7.65 (d, 1H, J = 1.92 Hz, Ar), 7.53–7.47 (m, AA′ BB′, 2H, Ar), 7.32 (d, 1H, J = 1.92 Hz, Ar), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.5, 147.9, 146.9, 142.2, 137.4, 133.8, 129.2, 128.6, 126.0, 119.8, 114.3, 35.1, 34.5, 31.9, 30.0.

4.1.4.39. 5,7-Di-*tert*-butyl-2-(2-bromophenyl)-benzoxazole (7f). Method A. Yield 31%. Anal. Calcd for $C_{21}H_{24}BrNO$ (386.33): 65.29% C, 6.26% H, 3.63% N. Found: 65.35% C, 6.32% H, 3.56% N. Mp 157–158 °C. HPLC purity 99.61%. TLC: R_f (S_1) 0.73, R_f (S_2) 0.82. UV (nm), λ_{max}/log ε: 210/3.63, 308. IR (KBr) cm^{-1}: 2963, 2907, 2869 (C–H), 1624, 1551 (C=N), 1473 (C=C), 1392, 1364 (CH_3), 1058 (C–O–C), 866, 841, 722 (Ar–H). MS: the peak 386.4 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.11 (dd, 1H, J = 7.69 Hz, J = 1.37 Hz, Ar), 7.77 (dd, 1H, J = 7.69 Hz, J = 1.37 Hz, Ar), 7.72 (d, 1H, J = 1.92 Hz, Ar), 7.47 (dt, 1H, J = 7.69 Hz, J = 1.37 Hz, Ar), 7.40-7.33 (m, 1H, Ar), 7.36 (d overlapped, 1H, J = 1.92 Hz, Ar), 1.55 (s, 9H, CH_3), 1.41 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.1, 147.8, 147.0, 141.8, 134.6, 134.1, 132.2, 131.8, 128.8, 127.5, 121.6, 119.9, 114.5, 35.1, 34.4, 31.8, 29.9.

4.1.4.40. 5,7-Di-*tert*-butyl-2-(3-bromophenyl)-benzoxazole (7g). Method A. Yield 33%. Anal. Calcd for $C_{21}H_{24}BrNO$ (386.33): 65.29% C, 6.26% H, 3.63% N. Found: 65.20% C, 6.36% H, 3.52% N. Mp 100–101 °C. HPLC purity 99.47%. TLC: R_f (S_1) 0.65, R_f (S_2) 0.82. UV (nm), λ_{max}/log ε: 206/3.63, 290. IR (KBr) cm^{-1}: 2961, 2907, 2868 (C–H), 1629, 1568 (C=N), 1481 (C=C), 1391, 1331 (CH_3), 1082 (C=O=C), 868, 839, 768, 732 (Ar–H). MS: the peak 386.4 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.38 (t, 1H, J = 1.79 Hz, Ar), 8.20–8.16 (m, 1H, Ar), 7.67–7.63 (m, 1H, Ar), 7.65 (d, overlapped, 1H, J = 1.92 Hz, Ar), 7.40 (t, 1H, J = 7.69 Hz, Ar), 7.33 (d, 1H, J = 1.92 Hz, Ar), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 160.9, 148.0, 147.0, 142.1, 134.1, 133.9, 130.4, 130.2, 129.4, 125.9, 122.9, 120.0, 114.3, 35.1, 34.5, 31.8, 30.0.

4.1.4.41. 5,7-Di-*tert*-butyl-2-(4-bromophenyl)-benzoxazole (7h). Method A. Yield 39%. Anal. Calcd for $C_{21}H_{24}BrNO$ (386.33): 65.29% C, 6.26% H, 3.63% N. Found: 65.21% C, 6.19% H, 3.49% N. Mp 101–103 °C. HPLC purity 98.89%. TLC: R_f (S_1) 0.82, R_f (S_2) 0.85. UV (nm), λ_{max}/log ε: 207/3.64, 249, 311. IR (KBr) cm^{-1}: 2957, 2906, 2868 (C–H), 1620, 1596 (C=N), 1481 (C=C), 1397, 1363 (CH_3), 1072 (C–O–C), 868, 730 (Ar–H). MS: the peak 386.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.14–8.08 (m, AA′BB′, 2H, Ar), 7.69–7.64 (m, AA′BB′, 2H, Ar), 7.65 (d, overlapped, 1H, J = 1.92 Hz, Ar), 7.32 (d, 1H, J = 1.92 Hz, Ar), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.6, 147.9, 146.9, 142.2, 133.8, 132.2, 128.8, 126.5, 125.8, 119.9, 114.3, 35.1, 34.5, 31.8, 30.0.

4.1.4.42. 5,7-Di-*tert*-butyl-2-(2-trifluoromethylphenyl)-benzoxazole (7i). Method A. Yield 78%. Anal. Calcd for $C_{22}H_{24}F_3NO$ (375.43): 70.38% C, 6.44% H, 3.73% N. Found: 70.46% C, 6.58% H, 3.64% N. Mp 54-61 °C. HPLC purity 98.23%. TLC: R_f (S_1) 0.52, R_f (S_2) 0.78. UV (nm), λ_{max}/log ε: 207/3.63, 306. IR ($CHCl_3$) cm^{-1}: 3023, 2967, 2908, 2871 (C–H), 1610, 1559(C=N), 1480, 1455 (C=C), 1393, 1364 (CH_3). MS: the peak 375.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.53–8.49 (m, 1 H, Ar), 8.45–8.40 (m, 1H, Ar), 7.81–7.75 (m, 1H, Ar), 7.70–7.63 (m, 1H, Ar), 7.68 (d overlapped, 1H, J = 1.93 Hz, Ar), 7.35 (d, 1H, J = 1.93 Hz, Ar), 1.56 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.0, 148.2, 147.0, 142.1, 133.0, 131.6 (q, J = 32.9 Hz), 130.4 (q, J = 0.80 Hz), 129.5, 128.4, 127.6 (q, J = 3.50 Hz), 124.2 (q, J = 3.70 Hz), 123.7 (q, J = 272.6 Hz), 120.2, 114.4, 35.1, 34.5, 31.8, 30.0.

4.1.4.43. 5,7-Di-*tert*-butyl-2-(3-trifluoromethylphenyl)-benzoxazole (7j). Method A. Yield 87%. Anal. Calcd for $C_{22}H_{24}F_3NO$ (375.43): 70.38% C, 6.44% H, 3.73% N. Found: 70.44% C, 6.56% H, 3.67% N. Mp 62 °C. HPLC

purity 99.86%. TLC: R_f (S_1) 0.58, R_f (S_2) 0.85. UV (nm), λ_{max}/log ε: 211, 306/3.62. IR ($CHCl_3$) cm^{-1}: 3022, 2967, 2908, 2871 (C–H), 1610, 1559 (C=N), 1480, 1455 (C=C), 1393, 1364 (CH_3). MS: the peak 375.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.53–8.49 (m, 1H, Ar), 8.45–8.40 (m, 1H, Ar), 7.81–7.75 (m, 1H, Ar), 7.71–7.63 (m, 1H, Ar), 7.68 (d overlapped, 1H, J = 1.92 Hz, Ar), 7.35 (d, 1H, J = 1.92 Hz, Ar), 1.56 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.0, 148.2, 147.0, 142.1, 134.0, 131.6 (q, J = 32.9 Hz), 130.4, 130.3, 129.5, 128.4, 127.6 (q, J = 3.50 Hz), 124.2 (q, J = 3.70 Hz), 123.7 (q, J = 272.60 Hz), 120.2, 35.1, 34.5, 31.8, 30.0.

4.1.4.44. 5,7-Di-*tert*-butyl-2-(4-trifluoromethylphenyl)-benzoxazole (7k). Method A. Yield 37%. Anal. Calcd for $C_{22}H_{24}F_3NO$ (375.43): 70.38% C, 6.44% H, 3.73% N. Found: 70.31% C, 6.51% H, 3.62% N. Mp 79–81 °C. HPLC purity 99.68%. TLC: R_f (S_1) 0.48, R_f (S_2) 0.90. UV (nm), λ_{max}/log ε: 202, 307/3.67. IR (KBr) cm^{-1}: 2961, 2908, 2871 (C–H), 1627, 1562 (C=N), 1482 (C=C), 1393, 1365 (CH_3), 1324, 1167, 1129 (CF_3), 1073 (C–O–C), 851, 707 (Ar–H). MS: the peak 375.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.40–8.52 (m, AA′BB′, 2H, Ar), 7.83–7.76 (m, AA′BB′, 2H, Ar), 7.68 (d, 1H, J = 1.92 Hz, Ar), 7.36 (d, 1H, J = 1.92 Hz, Ar), 1.56 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ 160.9, 148.2, 147.1, 142.1, 134.0, 132.7 (q, J = 32.70 Hz), 130.8, 127.6, 125.9 (q, J = 3.70 Hz), 120.3, 120.2 (q, J = 272.60 Hz), 114.5, 35.1, 34.5, 31.8, 30.0.

4.1.4.45. 2-(5,7-Di-*tert*-butylbenzoxazol-2-yl)-phenol (7l). Method A. Yield 75%. Method B. Yield 15%. Anal. Calcd for $C_{21}H_{25}NO_2$ (323.43): 77.98% C, 7.79% H, 4.33% N. Found: 77.90% C, 7.88% H, 4.24% N. Mp 120–123 °C, Mp 99–100 °C.[25] HPLC purity 99.97%. TLC: R_f (S_1) 0.65, R_f (S_2) 0.90. UV (nm), λ_{max}/log ε: 208, 277, 290, 322/3.64. IR ($CHCl_3$) cm^{-1}: 3432 (OH), 2964, 2906, 2870 (C–H), 1635, 1548 (C=N), 1488 (C=C), 1393, 1364 (CH_3), 1063 (C–O–C), 867, 750 (Ar–H). MS: the peak 323.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 11.58 (br s, 1H, OH), 8.04 (dd, 1H, J = 7.28 Hz, J = 1.37 Hz, Ar), 7.61 (d, 1H, J = 1.92 Hz, Ar), 7.47–7.40 (m, 1H, Ar), 7.34 (d, 1H, J = 1.92 Hz, Ar), 7.13 (dd, 1H, J = 7.28 Hz, J = 1.37 Hz, Ar), 7.06–6.99 (m, 1H, Ar), 1.56 (s, 9H, CH_3), 1.41 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 162.3, 158.6, 148.3, 145.3, 140.2, 133.8, 133.2, 126.9, 119.9, 119.4, 117.3, 113.5, 110.9, 35.1, 34.5, 31.8, 30.0.

4.1.4.46. 5,7-Di-*tert*-butyl-2-(2-nitrophenyl)-benzoxazole (7m). Method A. Yield 32%. Anal. Calcd for $C_{21}H_{24}N_2O_3$ (352.43): 71.57% C, 6.86% H, 7.95% N. Found: 71.69% C, 6.98% H, 7.81% N. Mp 127–128 °C. HPLC purity 99.68%. TLC: R_F (S_1) 0.12, R_f (S_2) 0.71. UV (nm), λ_{max}/log ε: 211/3.65, 283, 311. IR (KBr) cm^{-1}: 2961, 2908, 2870 (C–H), 1629 (C=N), 1536 (NO_2), 1482, 1466 (C=C), 1392, 1365 (CH_3), 1307 (NO_2), 1061 (C–O–C), 869, 785, 756 (Ar–H). MS: the peak 352.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.23–8.19 (m, 1H, Ar), 7.86–7.82 (m, 1H, Ar), 7.77–7.63 (m, 2H, Ar), 7.68 (d, overlapped, 1H, J = 1.92 Hz, Ar), 7.36 (d, 1H, J = 1.92 Hz, Ar), 1.47 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 158.2, 149.3, 148.2, 147.4, 141.7, 134.3, 132.1, 131.6, 131.2, 123.9, 121.4 120.5, 114.7, 35.1, 34.3, 31.8, 29.9.

4.1.4.47. 5,7-Di-*tert*-butyl-2-(4-nitrophenyl)-benzoxazole (7n). Method A. Yield 78%. Anal. Calcd for $C_{21}H_{24}N_2O_3$ (352.43): 71.57% C, 6.86% H, 7.95% N. Found: 71.49% C, 6.95% H, 7.84% N. Mp 201–202 °C, Mp 199–200 °C.[29] HPLC purity 99.84%. TLC: R_F (S_1) 0.65, R_f (S_2) 0.90. UV (nm), λ_{max}/log ε: 202, 238, 341/3.67. IR (KBr) cm^{-1}: 2956, 2987, 2869 (C–H), 1622, 1606, 1558 (C=N), 1526 (NO_2), 1483 (C=C), 1390, 1362 (CH_3), 1345 (NO_2), 1067 (C–O–C), 858, 707 (Ar–H). MS: the peak 352.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.44–8.31 (m, 4H, Ar), 7.69 (d, 1H, J = 1.64 Hz, Ar), 7.38 (d, 1H, J = 1.64 Hz, Ar), 1.56 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 160.1, 149.1, 148.5, 147.3, 142.2, 134.1, 133.1, 128.1, 124.2, 120.9, 114.7, 35.2, 34.5, 31.7, 30.0.

4.1.4.48. 5,7-Di-*tert*-butyl-2-(4-dimethylaminophenyl)-benzoxazole (7o). Method A. Yield 72%. Anal. Calcd for $C_{23}H_{30}N_2O$ (350.50): 78.82% C, 8.63% H, 7.99% N. Found: 78.90% C, 8.68% H, 7.91% N. Mp 72 °C. Mp 149–150 °C.[29] HPLC purity 99.64%. TLC: R_f (S_1) 0.17, R_F (S_2) 0.72. UV (nm), λ_{max}/log ε: 206, 344/3.69. IR (KBr) cm^{-1}: 2957, 2905, 2868 (C–H), 1611, 1512 (C=N), 1481 (C=C), 1391, 1366 (CH_3), 1066 (C–O–C), 823, 741 (Ar–H). MS: the peak 350.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.17–8.05 (m AA′, BB′, 2H, Ar), 7.60 (d, 1H, J = 1.65 Hz, Ar), 7.23 (d, 1H, J = 1.65 Hz, Ar), 6.83–6.74 (m AA′, BB′, 2H, Ar), 3.07 (s, 6H, NCH_3), 1.54 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 163.6, 152.2, 147.2, 146.6, 142.7, 133.1, 128.8, 118.3, 114.7, 113.5, 111.6, 40.1, 35.0, 34.4, 31.8, 30.0.

4.1.4.49. 5,7-Di-*tert*-butyl-2-(4-methylsulfanylphenyl)-benzoxazole (7p). Method A. Yield 84%. Anal. Calcd for $C_{22}H_{27}NOS$ (353.52): 74.74% C, 7.70% H, 3.96% N. Found: 74.83% C, 7.79% H, 3.82% N. Mp 111–112 °C. HPLC purity 99.51%. UV (nm), λ_{max}/log ε: 205, 231, 325/3.69. IR (KBr) cm^{-1}: 2963, 2905, 2869 (C–H), 1624, 1598 (C=N), 1483 (C=C), 1384, 1364 (CH_3), 869, 828, 735 (Ar–H). MS: the peak 353.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.23–8.11 (m, AA′BB′, 2H, Ar), 7.64 (d, 1H, J = 1.79 Hz, Ar), 7.40–7.33 (m, AA′BB′, 2H, Ar), 7.30 (d, 1H, J = 1.79 Hz, Ar), 2.55 (s, 3H, SCH_3), 1.55 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 162.3, 147.7, 146.8, 143.2, 142.2, 133.6, 127.6, 125.8, 123.7, 119.4, 114.0, 35.1, 34.4, 31.8, 30.0, 15.0.

4.1.4.50. 5,7-Di-*tert*-butyl-2-(4-fluoro-3-phenoxyphenyl)-benzoxazole (7q). Method A. Yield 68%. Anal. Calcd for $C_{27}H_{28}FNO_2$ (417.52): 77.67% C, 6.76% H, 3.35% N. Found: 77.53% C, 6.84% H, 3.29% N. Mp 91–93 °C. HPLC purity 99.33%. UV (nm), λ_{max}/log ε: 207/3.73, 308. IR (KBr) cm^{-1}: 2961, 2907, 2870 (C–H), 1590 (C=N), 1491 (C=C), 1400, 1364 (CH_3),

1267, 1211 (–O–), 869, 803, 749, 731, 690 (Ar–H). MS: the peak 417.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.06–7.93 (2H, m, Ar), 7.61 (1H, d, J = 1.70 Hz, Ar), 7.42–7.32 (3H, m, Ar), 7.31 (1H, d, J = 1.70 Hz, Ar), 7.19–7.11 (1H, m, Ar), 7.10-7.03 (2H, m, Ar), 1.52 (9H, s, CH_3), 1.39 (9H, s, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 159.4 (d, J = 237.30 Hz), 156.8, 154.5, 147.9, 147.0, 144.4, (d, J = 12.00 Hz), 142.1, 133.7, 129.9, 124.6 (d, J = 3.70 Hz), 123.9, 123.8 (d, J = 22.60 Hz), 120.7 (d, J = 2.00 Hz), 119.8, 117.7 (d, J = 22.60 Hz), 117.8, 114.2, 35.1, 34.4, 31.8, 30.0.

4.1.4.51. 5,7-Di-*tert*-butyl-2-styrylbenzoxazole (8a). Method A. Yield 67%. Anal. Calcd for $C_{23}H_{27}NO$ (333.47): 82.84% C, 8.16% H, 4.20% N. Found: 82.91% C, 8.08% H, 4.13% N. Mp 85–87 °C. HPLC purity 99.52%. TLC: R_f (S_1) 0.43, R_f (S_2) 0.88. UV (nm), λ_{max}/log ε: 210, 327/3.73. IR (KBr) cm^{-1}: 2959, 2906, 2869 (C–H), 1639, 1578 (C=N), 1535, 1482 (C=C), 1392, 1364 (CH_3), 1165 (C–O–C), 868, 842, 757 (Ar–H). MS: the peak 333.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 7.76 (d, 1H, J = 16.35 Hz, CH), 7.66–7.60 (m, 2H, Ar), 7.60 (d, 1H, J = 1.92 Hz, Ar), 7.47–7.36 (m, 3H, Ar), 7.30 (d, 1H, J = 1.92 Hz, Ar), 7.11 (d, 1H, J = 16.35 Hz, CH), 1.54 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 162.3, 147.7, 146.6, 142.4, 138.4, 135.3, 133.5, 129.6, 128.9, 127.5, 119.7, 114.3, 114.0, 35.0, 34.4, 31.8, 29.9.

4.1.4.52. 5,7-Di-*tert*-butyl-2-(1*H*-pyrazol-3-yl)-benzoxazole (8b). Method A. Yield 47%. Anal. Calcd for $C_{18}H_{23}N_3O$ (297.39): 72.70% C, 7.80% H, 14.13% N. Found: 72.62% C, 7.91% H, 14.04% N. Mp 177–180 °C. HPLC purity 99.55%. TLC: R_f (S_1) 0.05, R_f (S_2) 0.65. UV (nm), λ_{max}/log ε: 203, 237, 293/3.65. IR (KBr) cm^{-1}: 2961, 2908, 2870 (C–H), 1609 (C=N), 1482, 1464 (C=C), 1392, 1364 (CH_3), 1050 (C–O–C), 867, 774, 740 (Ar–H). MS: the peak 297.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 11.09 (br s, 1H, NH), 7.98 (d, 1H, J = 2.48 Hz, Ar), 7.71 (d, 1H,J = 1.92 Hz, Ar), 7.35 (d, 1H, J = 1.92 Hz, Ar), 7.11 (d, 1H, J = 2.48 Hz, Ar), 1.54 (s, 9H, CH_3), 1.41 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 157.0, 148.1, 146.6, 141.6, 138.4, 134.0, 133.0, 120.0, 114.2, 106.3, 35.1, 34.5, 31.8, 30.0.

4.1.4.53. 5,7-Di-*tert*-butyl-2-(pyridin-2-yl)-benzoxazole (8c). Method A. Yield 31%. Anal. Calcd for $C_{20}H_{24}N_2O$ (308.42): 77.89% C, 7.84% H, 9.08% N. Found: 77.99% C, 7.94% H, 8.91% N. Mp 75–77 °C. HPLC purity 98.44%. TLC: R_f (S_1) 0.06, R_f (S_2) 0.30. UV (nm), λ_{max}/log ε: 215, 310/3.70. IR ($CHCl_3$) cm^{-1}: 2960, 2907, 2869 (C–H), 1623, 1556 (C=N), 1458 (C=C), 1392, 1364 (CH_3), 1082 (C–O–C), 866, 839 (Ar–H). MS: the peak 308.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.85–8.81 (m, 1H, Ar), 8.30–8.26 (m, 1H, Ar), 7.91–7.83 (m, 1H, Ar), 7.70–7.68 (m, 1H, Ar), 7.45–7.39 (m, 1H, Ar), 7.37–7.35 (m, 1H, Ar), 1.56 (s, 9H, CH_3), 1.40 (d, 9H, J = 0.55 Hz, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.1, 150.4, 148.0, 147.3, 146.5, 142.1, 136.8, 134.1, 125.1, 123.1, 120.4, 114.8, 35.1, 34.5, 31.8, 30.1.

4.1.4.54. 5,7-Di-*tert*-butyl-2-(pyridin-3-yl)-benzoxazole (8d). Method A. Yield 77%. Anal. Calcd for $C_{20}H_{24}N_2O$ (308.42): 77.89% C, 7.84% H, 9.08% N. Found: 77.96% C, 7.78% H, 8.98% N. Mp 94–96 °C. HPLC purity 99.55%. TLC: R_f (S_1) 0.20, R_f (S_2) 0.38. UV (nm), λ_{max}/log ε: 216, 306/3.68. IR ($CHCl_3$) cm^{-1}: 2965, 2906, 2869 (C–H), 1607, 1576, 1552 (C=N), 1480 (C=C), 1392, 1362 (CH_3), 1076 (C–O–C), 865, 841 (Ar–H). MS: the peak 308.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 9.47 (d, 1H, J = 1.64 Hz, Ar), 8.75 (dd, 1H, J = 4.81 Hz, J = 1.64 Hz, Ar), 8.50 (dt, 1H, J = 7.97 Hz, J = 1.64 Hz, Ar), 7.67 (d, 1H, J = 1.79 Hz, Ar), 7.47 (ddd, 1H, J = 7.97 Hz, J = 4.81 Hz, J = 0.82 Hz, Ar), 7.34 (d, 1H, J = 1.79 Hz, Ar), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 160.1, 151.8, 148.6, 148.2, 147.0, 142.0, 134.4, 133.9, 123.8, 123.7, 120.2, 114.4, 35.1, 34.5, 31.8, 30.0.

4.1.4.55. 5,7-Di-*tert*-butyl-2-(pyridin-4-yl)-benzoxazole (8e). Method A. Yield 78%. Anal. Calcd for $C_{20}H_{24}N_2O$ (308.42): 77.89% C, 7.84% H, 9.08% N. Found: 78.01% C, 7.98% H, 8.89% N. Mp 97–100 °C. HPLC purity 98.26%. TLC: R_f (S_1) 0.06, R_f (S_2) 0.41. UV (nm), λ_{max}/log ε: 217, 309/3.65. IR ($CHCl_3$) cm^{-1}: 2958, 2907, 2870 (C–H), 1624, 1606, 1569 (C=N), 1482 (C=C), 1392, 1363 (CH_3), 1063 (C–O–C), 868, 843 (Ar–H). MS: the peak 308.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.84–8.79 (m, 2H, Ar), 8.10–8.06 (m, 2H, Ar), 7.68 (d, 1H, J = 1.79 Hz, Ar), 7.38 (d, 1H, J = 1.79 Hz, Ar), 1.55 (s, 9H, CH_3), 1.40 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 160.0, 150.7, 148.4, 147.1, 142.0, 134.6, 134.1, 120.8, 114.7, 35.1, 34.5, 31.7, 30.0.

4.1.4.56. 5,7-Di-*tert*-butyl-2-(6-methylpyridin-2-yl)-benzoxazole (8f). Method A. Yield 37%. Anal. Calcd for $C_{21}H_{26}N_2O$ (322.44): 78.22% C, 8.13% H, 8.69% N. Found: 78.30% C, 8.19% H, 8.57% N. Mp 58–60 °C. HPLC purity 98.75%. TLC: R_f (S_1) 0.13, R_f (S_2) 0.48. UV (nm), λ_{max}/log ε: 208, 239, 309/3.73. IR ($CHCl_3$) cm^{-1}: 2961, 2908, 2870 (C–H), 1623, 1553 (C=N), 1457 (C=C), 1393, 1364 (CH_3), 1082 (C–O–C), 866, 804 (Ar–H). MS: the peak 322.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.04 (d, 1H, J = 7.69 Hz, Ar), 7.75 (t, 1H, J = 7.69 Hz, Ar), 7.71 (d, 1H, J = 1.92 Hz, Ar), 7.34 (d, 1H, J = 1.92 Hz, Ar), 7.28 (d, 1H, J = 7.69 Hz, Ar), 2.71 (s, 3H, CH_3), 1.55 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.3, 159.7, 147.9, 147.1, 145.6, 142.2, 136.9, 133.9, 125.0, 120.2, 120.1, 115.0, 35.0, 34.5, 31.8, 30.0, 24.7.

4.1.4.57. 5,7-Di-*tert*-butyl-2-(1*H*-indol-3-yl)-benzoxazole (8g). Method A. Yield 51%. Anal. Calcd for $C_{23}H_{26}N_2O$ (346.47): 79.73% C, 7.56% H, 8.09% N. Found: 79.81% C, 7.63% H, 7.92% N. Mp 209–211 °C. HPLC purity 98.08%. TLC: R_f (S_2) 0.20. UV (nm), λ_{max}/log ε: 221/3.76, 267, 318. IR (KBr) cm^{-1}: 2961, 2907, 2869 (C–H), 1632 (C=N), 1482, 1458 (C=C), 1403, 1364 (CH_3), 1041 (C–O–C), 865, 743 (Ar–H). MS: the peak 346.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 9.46 (br s, 1H, NH), 8.45 (d, 1H,

J = 7.69 Hz, Ar), 8.21–8.14 (m, 1H, Ar), 7.67-7.64 (m, 1H, Ar), 7.53–7.44 (m, 1H, Ar), 7.41–7.21 (m, 3H, Ar), 1.60 (s, 9H, CH_3), 1.41 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 160.9, 147.6, 146.0, 141.5, 136.4, 133.3, 127.9, 124.9, 123.4, 122.0, 121.2, 118.6, 113.1, 111.9, 105.1, 35.1, 34.4, 31.9, 30.1.

4.1.4.58. 5,7-Di-*tert*-butyl-2-(furan-2-yl)-benzoxazole (8h). Method A. Yield 95%. Anal. Calcd for $C_{19}H_{23}NO_2$ (297.39): 76.73% C, 7.80% H, 4.71% N. Found: 76.84% C, 7.89% H, 4.63% N. Viscous oil. HPLC purity 98.06%. TLC: R_f (S_1) 0.51, R_f (S_2) 0.77. UV (nm), λ_{max}/log ε: 203, 248, 309/3.69. IR (KBr) cm^{-1}: 2961, 2907, 2870 (C–H), 1637, 1600 (C=N), 1541, 1460 (C=C), 1393, 1364 (CH_3), 1087 (C–O–C), 896, 867 (Ar–H). MS: the peak 297.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 8.18 (dd, 1H, J = 3.02 Hz, J = 1.10 Hz, Ar), 7.80 (dd, 1H, J = 4.94 Hz, J = 1.10 Hz, Ar), 7.63 (d, 1H, J = 1.79 Hz, Ar), 7.45 (dd, 1H, J = 4.94 Hz, J = 3.02 Hz, Ar), 7.30 (d, 1H, J = 1.79 Hz, Ar), 1.54 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 159.2, 147.7, 146.5, 142.1, 133.6, 129.6, 127.4, 126.9, 126.6, 119.5, 114.1, 35.1, 34.5, 31.8, 30.0.

4.1.4.59. 5,7-Di-*tert*-butyl-2-(5-methylfuran-2-yl)-benzoxazole (8i). Method A. Yield 98%. Anal. Calcd for $C_{20}H_{25}NO_2$ (311.42): 77.14% C, 8.09% H, 4.50% N. Found: 77.20% C, 8.16% H, 4.41% N. Mp 98–100 °C. HPLC purity 98.83%. TLC: R_f (S_1) 0.35, R_f (S_2) 0.76. UV (nm), λ_{max}/log ε: 204, 249, 315/3.64. IR (KBr) cm^{-1}: 2967, 2917, 2868 (C–H), 1639, 1574 (C=N), 1482 (C=C), 1392, 1362 (CH_3), 1021 (C–O–C), 864, 837, 782 (Ar–H). MS: the peak 311.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 7.60 (d, 1H, J = 1.79 Hz, Ar), 7.27 (d, 1H, J = 1.79 Hz, Ar), 7.12 (d, 1H, J = 3.29 Hz, Ar), 6.21–6.18 (m, 1H, Ar), 2.45 (br s, 3H, CH_3), 1.51 (s, 9H, CH_3), 1.38 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 156.2, 155.1, 147.8, 146.2, 142.0, 141.3, 133.5, 119.4, 114.8, 114.1, 108.4, 35.0, 34.4, 31.8, 30.0, 13.9.

4.1.4.60. 5,7-Di-*tert*-butyl-2-(5-ethylfuran-2-yl)-benzoxazole (8j). Method A. Yield 40%. Anal. Calcd for $C_{21}H_{27}NO_2$ (325.44): 77.50% C, 8.36% H, 4.30% N. Found: 77.61% C, 8.43% H, 4.22% N. Viscous oil. HPLC purity 98.64%. TLC: R_f (S_1) 0.51, R_f (S_2) 0.75. UV (nm), λ_{max}/log ε: 202, 316/3.64. IR ($CHCl_3$) cm^{-1}: 2968, 2907, 2872 (C–H), 1648, 1567 (C=N), 1481 (C=C), 1392, 1365 (CH_3). MS: the peak 325.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 7.60 (d, 1H, J = 1.92 Hz, Ar), 7.27 (d, 1H, J = 1.92 Hz, Ar), 7.14 (d, 1H, J = 3.57 Hz, Ar), 6.22–6.19 (m, 1H, Ar), 2.80 (q, 2H, J = 7.42 Hz, CH_2), 1.51 (s, 9H, CH_3), 1.38 (s, 9H, CH_3), 1.32 (t, 3H, J = 7.42 Hz, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 161.8, 155.2, 147.8, 146.2, 142.0, 141.1, 133.5, 119.3, 114.7, 114.1, 106.8, 35.0, 34.4, 31.8, 29.9, 21.7, 11.9.

4.1.4.61. 5,7-Di-*tert*-butyl-2-(thiophen-2-yl)-benzoxazole (8k). Method A. Yield 54%. Anal. Calcd for $C_{19}H_{23}NOS$ (313.46): 72.80% C, 7.40% H, 4.47% N. Found: 72.73% C, 7.48% H, 4.38% N. Mp 72–73 °C. HPLC purity 99.08%. TLC: R_F (S_1) 0.43, R_f (S_2) 0.86. UV (nm), λ_{max}/log ε: 206, 251, 314/3.72. IR ($CHCl_3$) cm^{-1}: 2958, 2907, 2869 (C=H), 1624, 1581 (C=N), 1481 (C=C), 1392, 1363 (CH_3), 1012 (C–O–C), 865, 858 (Ar–H). MS: the peak 313.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 7.90 (dd, 1H, J = 3.84 Hz, J = 1.24 Hz, Ar), 7.62 (d, 1H, J = 1.79 Hz, Ar), 7.53 (dd, 1H, J = 4.95 Hz, J = 1.24 Hz, Ar), 7.29 (d, 1H, J = 1.79 Hz, Ar), 7.18 (dd, 1H, J = 4.95 Hz, J = 3.84 Hz, Ar), 1.53 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 158.5, 147.9, 146.6, 142.1, 133.6, 130.1, 129.7, 129.3, 128.1, 119.5, 114.0, 35.1, 34.4, 31.8, 30.0.

4.1.4.62. 5,7-Di-*tert*-butyl-2-(thiophen-3-yl)-benzoxazole (8l). Method A. Yield 42%. Anal. Calcd for $C_{19}H_{23}NOS$ (313.46): 72.80% C, 7.40% H, 4.47% N. Found: 72.86% C, 7.51% H, 4.35% N. Viscous oil. HPLC purity 98.95%. TLC: R_F (S_1) 0.52, R_f (S_2) 0.78. UV (nm), λ_{max}/log ε: 211/3.65, 301. IR (KBr) cm^{-1}: 2961, 2907, 2869 (C–H), 1627, 1582 (C=N), 1481 (C=C), 1399, 1364 (CH_3), 1064 (C–O–C), 865, 796 (Ar–H). MS: the peak 313.6 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 7.66 (dd, 1H, J = 1.92 Hz, J = 0.82 Hz, Ar), 7.62 (d, 1H, J = 1.92 Hz, Ar), 7.30 (d, 1H, J = 1.92 Hz, Ar), 7.25 (dd, 1H, J = 3.57 Hz, J = 0.82 Hz, Ar), 6.60 (dd, 1H, J = 3.57 Hz, J = 1.64 Hz, Ar), 1.52 (s, 9H, CH_3), 1.39 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 154.9, 148.0, 146.3, 145.3, 143.0, 141.8, 133.7, 126.9, 119.7, 114.3, 113.5, 35.0, 34.4, 31.8, 30.0.

4.1.4.63. (5,7-Di-*tert*-butyl-benzoxazol-2-yl)-methanol (8m). Method C. Yield 63%. Anal. Calcd for $C_{16}H_{23}NO_2$ (261.36): 73.53% C, 8.87% H, 5.36% N. Found: 73.66% C, 8.98% H, 5.22% N. Mp 111–112 °C. HPLC purity 98.74%. UV (nm), λ_{max}/log ε: 210, 238, 276/3.61. IR (KBr) cm^{-1}: 2965, 2906, 2870 (C–H), 1610, 1578 (C=N), 1483 (C=C), 1392, 1363 (CH_3), 1099 (C–O–C), 875, 839 (Ar–H). MS: the peak 261.5 was found. ^{1}H NMR (300 MHz, $CDCl_3$), δ: 7.55 (d, 1H, J = 1.92 Hz, Ar), 7.28 (d, 1H, J = 1.92 Hz, Ar), 4.94 (s, 2H, CH_2), 4.06 (br s, 1H, OH), 1.46 (s, 9H, CH_3), 1.37 (s, 9H, CH_3). ^{13}C NMR (75 MHz, $CDCl_3$), δ: 165.1, 147.8, 147.1, 140.6, 134.0, 119.6, 113.9, 58.0, 35.0, 34.4, 31.8, 29.9.

4.2. Lipophilicity HPLC determination (capacity factor *K*/calculated log *K*)

The HPLC separation module Waters Alliance 2695 XE and Waters Photodiode Array Detector 2996 (Waters Corp., Milford, MA, USA) were used. The chromatographic column Symmetry® C_{18} 5 μm, 4.6 × 250 mm, Part No. WAT054275, (Waters Corp., Milford, MA, USA) was used. The HPLC separation process was monitored by Millennium32® Chromatography Manager Software, Waters 2004 (Waters Corp., Milford, MA, USA). The mixture of MeOH p.a. (90.0%) and H_2O-HPLC—Mili-Q Grade (10.0%) was used as a mobile phase. The total flow of the column was 1.0 mL/min, injection 30 μL, column temperature 45 °C and sample temperature 10 °C. The detection wavelength 210 nm

was chosen. The KI methanolic solution was used for the dead time (T_D) determination. Retention times (T_R) were measured in minutes.

The capacity factors K were calculated using the Millennium32® Chromatography Manager Software according to the formula $K = (T_R - T_D)/T_D$, where T_R is the retention time of the solute, whereas T_D denotes the dead time obtained via an unretained analyte. Log K, calculated from the capacity factor K, is used as the lipophilicity index converted to log P scale. The log K values of the individual compounds are shown in Tables 1 and 2.

4.3. Lipophilicity calculations

Log P, that is the logarithm of the partition coefficient for *n*-octanol/water, was calculated using the programs CS ChemOffice Ultra ver. 7.0 (CambridgeSoft, Cambridge, MA, USA) and ACD/Log P ver. 1.0 (Advanced Chemistry Development Inc., Toronto, Canada). C log P values (the logarithm of *n*-octanol/water partition coefficient based on established chemical interactions) were generated by means of CS ChemOffice Ultra ver. 7.0 (CambridgeSoft, Cambridge, MA, U.S.A.) software. The results are shown in Table 1 and 2.

4.4. Biological methods

4.4.1. Antimycobacterial evaluation. Primary screening was conducted at 6.25 μg/mL against *M. tuberculosis* $H_{37}Rv$ (ATCC27294) in BACTEC 12B medium using both microdilution assay and the Microplate Almar Blue Assay (MABA).[30] Compounds demonstrating at least 90% inhibition in the primary screen were tested at lower concentration against *M. tuberculosis* $H_{37}Rv$ to determine the MIC testing by MABA. The MIC is defined as the lowest concentration effecting a reduction in fluorescence of 99% relative to controls.[30] Antituberculosis assays for atypical strains were evaluated against *M. kansasii* CNCTC My 235/80, *M. avium* My 80/72 and *M. avium* My 152/74. The cultures were 10 days old. The assay was carried out in the semisynthetic Sula medium (Sevac, Prague, Czech Republic) at pH 6.0 and 37 °C. Used method was the microdilution broth method. The ability of the compounds to inhibit mycobacterial growth was determined after 7, 14 and 21 days.

4.4.2. Cytotoxicity assay. The cytotoxic effect of the compounds was tested at concentrations equal to and greater than the MIC for *M. tuberculosis* by MTT assay on human intestinal cell line HCT-8.[31] The cells were grown in RPMI 1640 medium supplemented with 10% horse serum and 2.0 mmol sodium pyruvate at 37 °C in a humidified atmosphere of 5% CO_2. For the experiments, cells were harvested with trypsin, resuspended in fresh medium to a final concentration of 5×10^4 cells/mL and seeded in aliquots (100 μL) onto 96-well Nunclon® tissue culture plates (Nunc GmbH & Co. KG, Germany). The medium was removed after 72 h of cell incubation and replaced with RPMI culture medium containing tested compounds dissolved in DMSO (1%). In control wells, the cells were incubated in medium containing DMSO without tested compound (positive control for cell viability) and in the medium containing 5% DMSO (positive control for cytotoxic effect). The ability of the compounds to inhibit cellular growth was determined after 72 h by adding 10 μL MTT (5.5 mg/mL) solution (Sigma-Aldrich, USA) to each well. After incubation for 4 h, the dark blue formazan crystal product was dissolved in 100 μL lysis solution (10% SDS with 0.01 mol HCl). The absorbance was read at 590 nm using multiplate spectrofluorimeter GENios PlusTM (Tecan, Switzerland). Each concentration of the compounds was tested in triplicate; the assays were repeated three times in separate experiments.

Acknowledgments

This work was financially supported by the Ministry of Health of the Czech Republic (No. 1A8238-3) and by the Ministry of Education of the Czech Republic (MSM 0021620822). Antimycobacterial data were provided by the Tuberculosis Antimicrobial Acquisition and Coordinating Facility (TAACF) through a research and development contract with the U.S. National Institute of Allergy and Infectious Diseases.

References and notes

1. Vinsova, J. *Cesk. Slov. Farm.* **2003**, *52*, 282.
2. Ramalinghan, C.; Balasubramanian, S.; Kabilan, S.; Vasudevan, M. *J. Eur. Med. Chem.* **2004**, *39*, 527.
3. Turan-Zitouni, G.; Demirayak, S.; Ozdemir, A.; Kaplacikli, Z. A.; Yildiz, M. T. *Eur. Med. Chem.* **2003**, *39*, 267.
4. Temiz, O.; Oren, I.; Sener, E. A.; Yalcin, I.; Ucartuerk, N. *Farmaco* **1998**, *53*, 337.
5. Oren, I.; Temiz, O.; Yalcin, I.; Sener, E. A.; Altanlar, N. *Eur. J. Pharm. Sci.* **1998**, *7*, 153.
6. Rida, S. M.; Ashour, F. A.; El-Hawash, S. A. M.; El-Semary, M. M.; Badr, M. H.; Shalaby, M. A. *Eur. Med. Chem.* **2005**, *20*, 949.
7. Kumar, D.; Jacob, M. R.; Reynolds, M. B.; Kerwin, S. M. *Bioorg. Med. Chem.* **2002**, *10*, 3997.
8. Kim, J. S.; Sun, Q.; Gatto, B.; Yu, C.; Liu, A.; Liu, L. F.; LaVoie, E. *J. Bioorg. Med. Chem.* **1996**, *4*, 621.
9. Hoffman, J. M.; Smith, A. M.; Rooney, C. S.; Fisher, T. E.; Wai, J. S.; Thomas, C. M.; Bamberger, D. L.; Barne, J. L.; William, T. M. *J. Med. Chem.* **1993**, *36*, 953.
10. Perrin, L.; Rakik, A.; Yearly, S.; Baumberger, C.; Kinloch-deLoies, S.; Pechiere, M.; Hirschel, B. *AIDS* **1996**, *10*, 1233.
11. Kozich, V.; Drezer, J.; Vodchits, A.; Wernere, W. *Chem. Phys. Lett.* **2005**, *415*, 121.
12. Holler, M. G.; Campo, L. F.; Brandelli, A.; Stefani, V. *J. Photochem. Photobiol. A: Chem.* **2002**, *149*, 217.
13. Laber, B.; Usunow, G.; Wiecko, E.; Franke, W.; Franke, H.; Koehn, A. *Pesticide Biochem. Physiol.* **1999**, *63*, 173.
14. Dunwell, D. W.; Evans, D. *J. Med. Chem.* **1977**, *20*, 797.
15. Vinsova, J.; Horak, V.; Buchta, V.; Kaustova, J. *Molecules* **2005**, *10*, 760.
16. Topliss, J. G. *J. Med. Chem.* **1977**, *20*, 463.
17. Hartmann, T.; Schmitt, J. *Drug Disc. Today Technol.* **2004**, *1*, 431.
18. Dolezal, M.; Palek, L.; Vinsova, J.; Buchta, V.; Jampilek, J.; Kralova, K. *Molecules* **2006**, *11*, 242.

19. Musiol, R.; Jampilek, J.; Buchta, V.; Silva, L.; Niedbala, H.; Podeszwa, B.; Palka, A.; Majerz-Maniecka, K. A.; Oleksyn, B.; Polanski, J. *Bioorg. Med. Chem.* **2006**, *14*, 3592.
20. Jampilek, J.; Dolezal, M.; Palek, L.; Vinsova, J.; Buchta, V. *Bioorg. Med. Chem.* **2006**, submitted.
21. Terasnima, M.; Ishii, M.; Kanaoka, Y. *Synthesis* **1982**, 484.
22. Pottorf, R. S.; Chacha, N. K.; Katkevics, M.; Ozila, V.; Suna, E.; Ghane, H.; Redgberg, T.; Player, M. R. *Tetrahedron Lett.* **2003**, *44*, 175.
23. Jimenez, P. V. M.; Camacho, C. C.; Gueizado, R. M.; Noech, H.; Contreras, R. *J. Organomet. Chem.* **2000**, *614*, 283.
24. Voleva, V. B.; Prokofeva, T. I.; Prokofev, A. I.; Belostotskaya, I. S.; Komisarova, N. L.; Ershov, V. V. *Russ. Chem. Bull.* **1995**, *44*, 1720.
25. Bowman, D. F.; Hewgill, F. R. *J. Chem. Soc. C* **1971**, 1777.
26. Hart, H.; Cassis, F. A. *J. Am. Chem. Soc.* **1951**, *73*, 3179.
27. Lodyato, V. I.; Yurkova, I. L.; Sorokin, V. L.; Shadyro, O. I.; Dolgopalets, V. I.; Kisel, M. A. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 1179.
28. Stegmann, H. B. *Justus Liebigs Ann. Chem.* **1972**, *755*, 17.
29. Khachaturiyan, G. H.; Shif, A. I.; Olekhnovich, L. P.; Revinskii, Y. V.; Bren, V. A.; Ivakhnenko, E. P. *Russ. J. Org. Chem.* **1995**, *31*, 645.
30. Collins, L.; Franzblau, S. G. *Antimicrob. Agents Chemother.* **1997**, *41*, 1004.
31. Mosmann, T. *J. Immunol. Methods* **1983**, *65*, 55.