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Preliminary Communications

Synthesis, evaluation and in *silico* molecular modeling of pyrroyl-1,3,4-thiadiazole inhibitors of InhA

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ABSTRACT

Enoyl acyl carrier protein reductase (ENR) is an essential type II fatty acid synthase (FAS-II) pathway enzyme that is an attractive target for designing novel antitubercular agents. Herein, we report sixty-eight novel pyrrolyl substituted aryloxy-1,3,4-thiadiazoles synthesized by three-step optimization processes. Three-dimensional quantitative structure–activity relationships (3D-QSAR) were established for pyrrolyl substituted aryloxy-1,3,4-thiadiazole series of InhA inhibitors using the comparative molecular field analysis (CoMFA). Docking analysis of the crystal structure of ENR performed by using Surflex-Dock in Sybyl-X 2.0 software indicates the occupation of pyrrolyl substituted aryloxy 1,3,4-thiadiazole into hydrophobic pocket of InhA enzyme. Based on docking and database alignment rules, two computational models were established to compare their statistical results. The analysis of 3D contour plots allowed us to investigate the effect of different substituent groups at different positions of the common scaffold. *In vitro* testing of ligands using biological assays substantiated the efficacy of ligands that were screened through in *silico* methods.

1. Introduction

Mycobacterium tuberculosis (*M. tuberculosis*) is mainly responsible for tuberculosis (TB), a deadly disease affecting one-third of the world population [1,2]. Occurrence of Human Immunodeficiency Virus (HIV), resulting in Acquired Immune Deficiency Syndrome (AIDS) has prompted the prevalence of TB, causing 50% of deaths among the HIV-infected patients due to co-infection with *M. tuberculosis*, thereby accelerating the collapse of immune system [3]. According to the World Health Organization (WHO), two million people die every year with at least nine million people getting infected. This provides greater opportunities to develop new active forms of TB [4].

The organization of bacterial fatty acid synthase type II system based on individual enzymes (FAS-II system) is quite different from the multifunctional fatty acid synthase type I system (FAS-I) found in eukaryotes, providing good prospects for its selective inhibition. One of the attractive antimicrobial drug targets is the bacterial fatty acid synthesis pathway (FAS-II) that has created interest in recent years for developing new chemical entities based on isoniazid (INH) [5–8]. Among the enzymes involved in FAS-II, the NADH-dependent enoyl acyl carrier protein reductase (ENR) encoded by *Mycobacterium* gene InhA is the key catalyst in mycolic acid biosynthesis. Studies in the past have established that InhA is the primary molecular target for INH [9], a frontline drug for over 40 years used in the treatment of TB. As a prodrug, INH must be first activated by KatG, a catalase-peroxidase that oxidizes INH to an acyl-radical, which binds covalently to NADH, the co-substrate for InhA [10]. The INH-NADH adduct functions as a potent inhibitor of InhA and other InhA inhibitors like diazaborines, triclosan, pyrazole derivatives and indole-5-amides have been reported earlier [11–15].

The 1,3,4-thiadiazoles and their analogs are the well known antimicrobial agents due to the presence of toxophoric —N=C—S moiety, which exhibits a broad spectrum of biological activities [16–23]. Based on our previous reports on pyrrole derivatives [24–27], we find it necessary to further develop pyrrolyl aryloxy thiadiazole derivatives as the effective anti-tubercular (anti-TB) agents.

One of the design strategies for new anti-TB compounds is based on hybridization. Azole derivatives have shown interesting anti-TB and antimicrobial activity, inhibiting the bacteria by blocking lipid biosynthesis and/or additional mechanisms. Thus,

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by hybridization between 1,3,4-thiadiazole and pyrrole moieties, new anti-TB agents are designed. In our ongoing efforts of designing new entities, herein we try to map pharmacophore from the structure of 1-cyclohexyl-*N*-(3,5-dichlorophenyl)-5-oxopyrrolidine-3-carboxamide (pyrrolidine carboxamide or 641) [15] which contains three hydrophobic moieties like 5-oxopyrrolidine, cyclohexyl and 3,5-dichlorophenyl, also contain carboxamide hydrogen bond acceptor and donor bridge between 5-oxopyrrolidine and 3,5-dichlorophenyl. Therefore, 5-oxopyrrolidine was mapped with 1,3,4-thiadiazole moiety, whereas carboxamide was mapped with methoxy group (—OCH_2), while hydrophobic moieties like cyclohexyl and 3,5-dichlorophenyl were mapped with pyrrole and *o*/*m*/*p* substituted phenyl derivatives as shown in Fig. 1. On the other hand, substitution in 2- and 5-positions of thiadiazole ring and the compounds obtained showed high lipophilicity, hypothesizing that this property could facilitate the passage of these compounds through *M. tuberculosis* bacterial membrane. In Fig. 2, some of the available antimicrobial and antimycobacterial agents (highlighted fragments) were considered for our intended new synthesis of various derivatives following the *Paal-Knorr* pyrrole synthesis. The computational strategies viz., three-dimensional quantitative structure–activity relationship (3D-QSAR) and molecular docking studies were used to correlate in *silico* results with *in vitro* analysis to find the ENR as the potential target of pyrrolyl aryloxy thiadiazole derivatives. Therefore, in this study, we have demonstrated Surflex-Docking and comparative molecular field analysis (CoMFA) analysis for activity prediction of pyrrolyl aryloxy thiadiazole derivatives that exhibit *in vitro* anti-TB activity.

2. Computational details

All calculations were performed with the commercially available SYBYL-X 2.0 software package (Tripos Associates, St. Louis, MO, USA) [28].

2.1. Data set and structures

In this study, 68 pyrrolyl thiadiazole anti-TB compounds that were synthesized have been considered for 3D-QSAR and docking analysis. The structures of pyrrolyl thiadiazole analogues were built using ChemDraw Ultra 11.0 and imported in SYBYL-X 2.0. All the hydrogen atoms were added, and structures were subsequently submitted to full energy minimization using the Conjugate Gradient energy minimization algorithm with a gradient of 0.01 kcal/mol Å. Tripos force field was used for minimization with nonbonded interactions cut-off of 8.0 Å using a distance dependent dielectric function with a dielectric constant value of 1. In the CoMFA study, 20 compounds were selected as the test set, which represented a range of antitubercular activities similar to that of a training set that was used to evaluate the predictive power of the resulting models. Biological activity of each compound was expressed as the minimal inhibitory concentrations (MIC) against *M. tuberculosis*, but −log(MIC) were used in 3D-QSAR analysis.

2.2. Alignment rule

The 3D structures of all the compounds in the training set and test set were built from SYBYL-X 2.0. Energy minimization was performed using Tripos force field [29] using the Powell optimization method [30] with a convergence gradient of 0.001 kcal/mol Å. Charges were calculated by MMFF94 (Merck Molecular Force Field) method and docked conformation of each compound in the active site of ENR was used as the pharmacophoric conformation for 3D-QSAR studies. Compound **9q**, which is one of the most active compounds, was used as the alignment template. Three reference atoms were used for alignment to identify pharmacophoric points: centroid of pyrrole ring, centroid of thiadiazole ring, and —CH_2 group attached to C_5 of thiadiazole ring (Fig. 3A and B).

2.3. CoMFA settings

The CoMFA descriptors [31], steric (Lennard-Jones 6–12 potential) and electrostatic (Coulombic potential) field energies were calculated using sp^3 carbon probe atom with +1 charge and van der Waals radius of 1.52 Å, with energy cut-off of 30 kcal/mol [32,33]. The CoMFA descriptor was used as independent variables, while the pMIC values were used as dependent variables in the partial least squares (PLS) regression analyses to derive the 3D-QSAR models using the standard implementation of the SYBYL package. In order to obtain the optimum number of principle components, the leave-one-out (LOO) cross-validation [34] method was utilized to measure how good the model fits the data in the training set. The cross-validated coefficient, q^2, was then calculated using Eq. (1):

$$q^2 = 1 - \frac{\sum_y (\mathbf{Y}_{pred} - \mathbf{Y}_{actual})^2}{\sum_y (\mathbf{Y}_{actual} - \mathbf{Y}_{mean})^2} \quad (1)$$

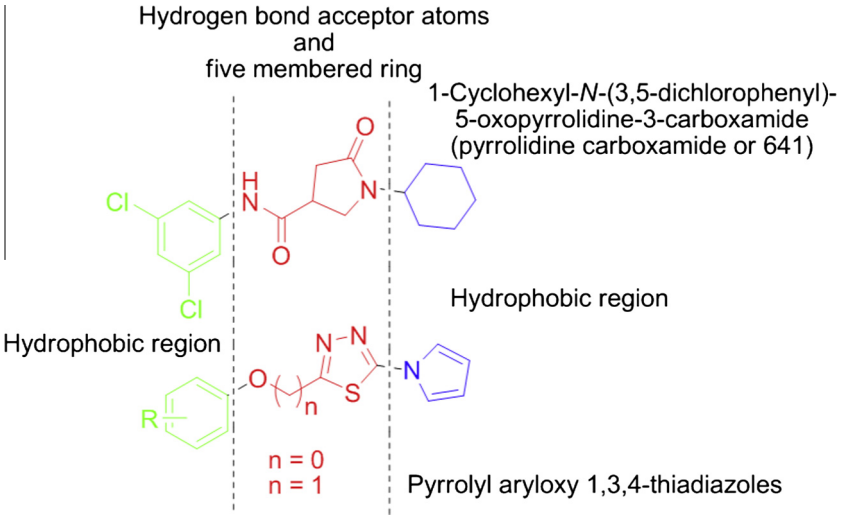


Fig. 1. Design concept for synthesized novel pyrrolyl aryloxy 1,3,4-thiadiazole derivatives.

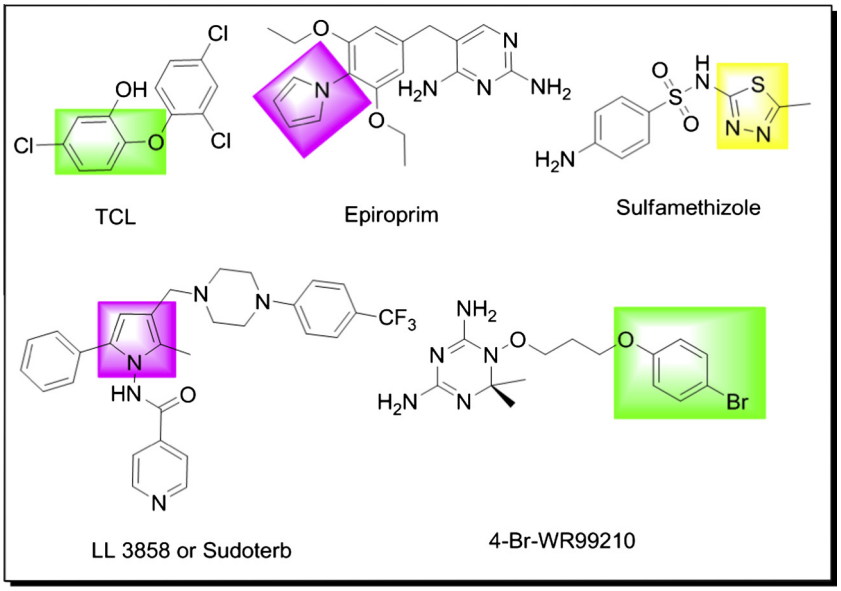


Fig. 2. Chemical structures of some available antimicrobial and antimycobacterial agents.

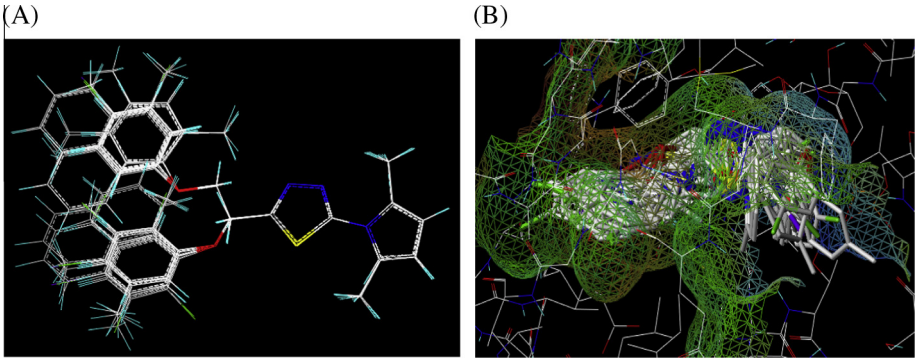


Fig. 3. (A) Database and (B) docking alignment for all 68 studied molecules.

where $\mathbf{Y}_{\text{predicted}}$, $\mathbf{Y}_{\text{observed}}$ and $\mathbf{Y}_{\text{mean}}$ are the predicted, actual (observed) and mean values of the target property (pMIC), respectively.

$$\text{SEE, SEP} = \sqrt{\frac{PRESS}{n-c-1}} \tag{2}$$

Here, n is the number of compounds, c is the number of components and *PRESS* is calculated as

$$PRESS = \sum y(\mathbf{Y}_{pred} - \mathbf{Y}_{actual})^2 \tag{3}$$

The noncrossvalidated form is determined through noncrossvalidated (conventional) r^2, the standard error of estimate (SEE), the F value (F-ratio) and the probability of r^2. To speed up the PLS analysis and to reduce the noise, a column filtering value of 2.00 kcal/mol was used. To further assess the robustness and statistical confidence of the derived model, the bootstrap analysis for 100 runs was performed [35]. Bootstrap analysis relies on the generation of many new sets of data from the original one, which were obtained by randomly choosing the samples from the original data set with repeated selection of the same sample being allowed. The statistical calculation was then performed on each of these bootstrap data sets. The difference between the parameters calculated from the original data set and the average of the parameters calculated from the many bootstrap data sets is a measure of the bias of the original calculations. In order to graphically interpret 3D-QSAR results in terms of field contributions, isocontour maps were generated using the field type "compound*coeff" and the contour levels were set to default values.

The 3D-QSAR model was assessed for its predictive ability using molecules of the test set that are not included in the construction of the model. External validation is the most acceptable validation method for the predictive ability of a QSAR model, since the molecules of the test set are not included in the training set. The external r^2_{pred} was calculated as:

$$r^2_{pred} = \frac{(SD - PRESS)}{SD} \tag{4}$$

where *SD* is the sum of squared deviations between biological activities of the test set and mean activity of the training set molecules, while *PRESS* is the sum of squared deviation between actual and predicted activities of the test set molecules as calculated by Eq. (3).

2.4. Preparation of InhA file

To explore the interaction and to illustrate accurate binding model for the active site of InhA with ligands, molecular docking was performed using Surflex-Dock module of another advanced version of SYBYL package (X 2.0). This docking approach aligns

the ligand to a "protomol" (also called idealized ligand) in the active site of the target. Surflex-Dock that adopted an empirical scoring function and a patented search engine [36,37] was employed for molecular docking study of the training set as well as the test set molecules into the active site of the monomeric unit "A" of the crystal structure of ENR catalytic core. The crystal structure of *M. tuberculosis* enoyl reductase (InhA) complexed with 1-cyclohexyl-*N*-(3,5-dichlorophenyl)-5-oxopyrrolidine-3-carboxamide (PDB ID 4TZK, 1.62 Å X-ray resolution) [15] was considered to examine the molecular docking study. All the hydrogen atoms of 4TZK were added by deleting all water molecules as well as united atom Amber7FF9902 were assigned for the protein and optimized for their geometry using Tripos force field.

3. Results and discussions

3.1. Chemistry

Compounds **6a–q**, **7a–q**, **8a–q** and **9a–q** were prepared as per Scheme 1. The aryloxy acetic/propionic acids (**2a–q/3a–q**) were prepared by condensing 2-chloro acetic acid/3-chloro propionic acid with phenol or substituted phenols. The 2-((phenoxy or substituted phenoxy)methyl/ethyl)-1,3,4-thiadiazol-5-amines **(4a–q and 5a–q)** were synthesized by the condensation of aryloxy acetic/propionic acids (**2a–q/3a–q**) with thiosemicarbazide in the presence of dehydrating agent ($POCl_3$). The *Paal-Knorr* pyrrole synthesis, which involves the reaction of 2,5-dimethoxy tetrahydrofuran or 2,5-hexanedione with amines, is among the most classical methods of heterocyclic pyrrole ring synthesis. Hence, the final derivatives viz., 2-((phenoxy or substituted phenoxy)methyl/ethyl)-5-(1*H*-pyrrol-1-yl)-1,3,4-thiadiazoles (**6a–q**, **8a–q**) and 2-((phenoxy or substituted phenoxy)methyl/ethyl)-5-(2,5-dimethyl-1*H*-pyrrol-1-yl)-1,3,4-thiadiazoles (**7a–q**, **9a–q**) were obtained by constructing pyrrole rings utilizing the free amino group at the 2-position of 1,3,4-thiadiazoles in the presence of dried glacial acetic acid.

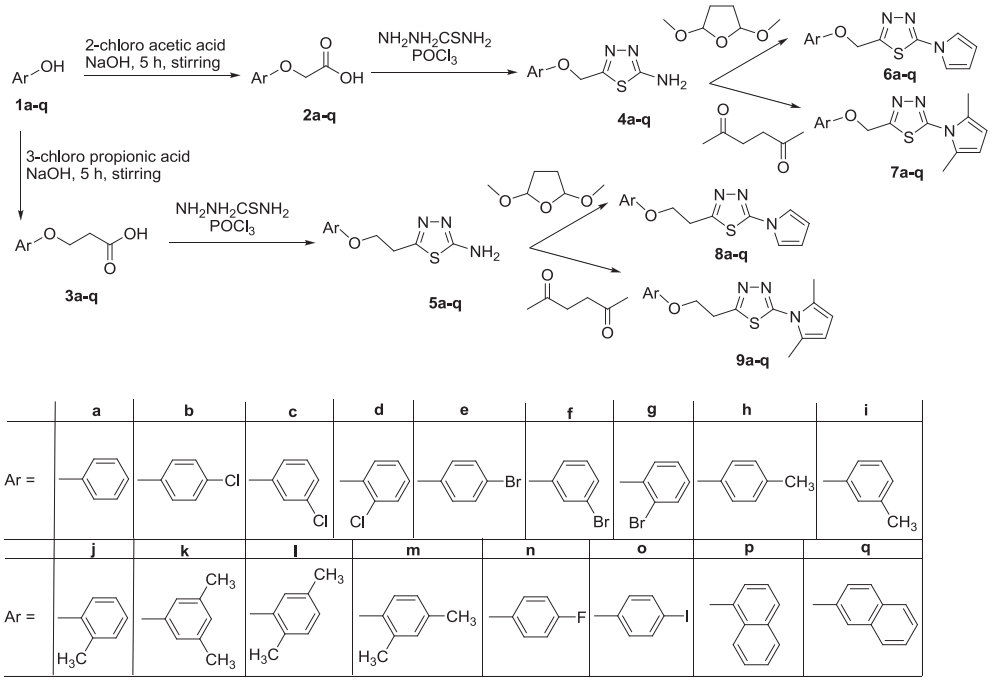
FTIR spectra showed two strong bands in the regions around 3171–2916 cm^{-1} and 1601–1507 cm^{-1}, corresponding to ν (Ar—H) and n ν (C═N), respectively. In the ^1H NMR spectra, resonance signals of —OCH_2 group (**6a–q**, **7a–q**), protons are seen as singlets in the range of δ 5.38–5.90 ppm, those of —OCH_2 and —CH_2 group (**8a–q**, **9a–q**) protons appear as triplets in the ranges of δ 5.41–4.30 ppm and δ 3.50–3.60 ppm, respectively. The pyrrole protons at the 3-H and 4-H positions appeared as singlet/triplet, ranging from δ 6.31 to 6.38 ppm for **6a–q**, **7a–q** and the observed singlet ranging from δ 5.84 to 5.94 ppm for **8a–q**, **9a–q**. The ^{13}C NMR spectra exhibited characteristic signals of 1,3,4-thiadiazole ring carbon atoms in the range of δ 168.59–161.14 ppm. The mass spectra showed an accurate molecular ion peak data for the respective compounds.

3.2. MIC

MIC values of the compounds against the selected *M. tuberculosis* $H_{37}Rv$ are given in Table 1. The tested compounds **(6a–q**, **7a–q**, **8a–q** and **9a–q)** showed activities against mycobacteria with the MIC values ranging from 3.125 to 100 μg/mL. Compound **9q** inhibited mycobacterial growth very effectively compared to others in the series with a MIC value of 3.125 μg/mL. One may expect an enhanced anti-TB activity of **7p**, **7q**, **9p**, **9q** molecules having bulky naphthoxy group with 2,5-dimethyl pyrrole. On the other hand, compounds **6n**, **7n**, **8n** and **9n** having halogen 4-F at the phenyl moiety, which is more electro-negative than other halogens like Cl, Br and I showed enhanced activity compared to other molecules in the series. The incorporation of bulky groups or halogen atoms increases the lipophilicity of the molecule, whereas mycobacterial cell wall is very lipophilic and contributions of these lipophilic substituents play an important role. Also, we have investigated the potential toxicity of eight selected pyrrolyl aryloxy 1,3,4-thiadiazole derivatives (**6n**, **7n**, **7p**, **7q**, **8n**, **9n**, **9p** and **9q**) towards mammalian Vero cell-lines and A_{549} (lung adenocarcinoma) cell-lines up to MIC values of 62.5 μg/mL. These compounds showed moderate cytotoxicity compared to standard INH (see Table 2).

3.3. Molecular modeling

After successful reproduction of crystallographic binding mode (Fig. 4) by Surflex-Dock, all the pyrrolyl aryloxy thiadiazole analogues were docked into the active site of InhA. Here, our aim



Scheme 1. Synthetic route for novel series of pyrrolyl aryloxy 1,3,4-thiadiazoles.

Table 1
Antitubercular data (MIC values in μg/mL) for the synthesized compounds (**6a–q**, **7a–q**, **8a–q** and **9a–q**).

Compd code	MIC value	Compd code	MIC value	Compd code	MIC value	Compd code	MIC value
6a	100	**7a**	50	**8a**	50	**9a**	25
6b	50	**7b**	25	**8b**	25	**9b**	25
6c	100	**7c**	50	**8c**	25	**9c**	25
6d	100	**7d**	25	**8d**	25	**9d**	25
6e	50	**7e**	25	**8e**	25	**9e**	25
6f	100	**7f**	50	**8f**	25	**9f**	25
6g	100	**7g**	50	**8g**	25	**9g**	25
6h	50	**7h**	25	**8h**	12.5	**9h**	25
6i	50	**7i**	50	**8i**	25	**9i**	50
6j	50	**7j**	50	**8j**	25	**9j**	50
6k	25	**7k**	12.5	**8k**	12.5	**9k**	12.5
6l	50	**7l**	50	**8l**	25	**9l**	25
6m	25	**7m**	12.5	**8m**	12.5	**9m**	12.5
6n	6.25	**7n**	6.25	**8n**	6.25	**9n**	6.25
6o	12.5	**7o**	12.5	**8o**	12.5	**9o**	12.5
6p	12.5	**7p**	6.25	**8p**	12.5	**9p**	6.25
6q	12.5	**7q**	6.25	**8q**	12.5	**9q**	3.125

Table 2
Cytotoxicity activity of selected pyrrolyl aryloxy 1,3,4-thiadiazole derivatives.

Compound	IC_{50} (μM)[a]	
	MV cell-lines[b]	A_{549}[c]
6n	187 ± 0.3	179 ± 0.3
7n	200 ± 0.3	201 ± 0.2
7p	213 ± 0.2	218 ± 0.2
7q	200 ± 0.3	213 ± 0.2
8n	208 ± 0.2	201 ± 0.2
9n	227 ± 0.2	225 ± 0.3
9p	218 ± 0.4	215 ± 0.3
9q	220 ± 0.3	225 ± 0.3
Isoniazid	>450	>450
Cisplatin	1.29	9.90

[a] Cytotoxicity is expressed as IC_{50}, which is the concentration of compound reduced by 50% of the optical density of treated cells with respect to untreated cells using MTT assay. Values are the means ± SEM of three independent experiments;
[b] Mammalian Vero cell-lines.
[c] A_{549} (lung adenocarcinoma) cell-lines.

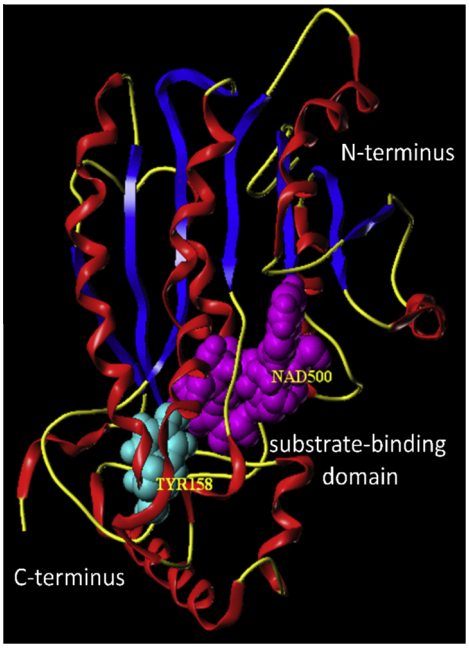


Fig. 4. Crystalline structure of 4TZK.

was to examine if all the analogues were docked into the active site of InhA in a similar way with the crystallographic binding modes. Docking studies with Surflex-Dock showed that compounds **9q** and **9n** occupy the same binding sites as that of pyrrolidine carboxamide (Fig. 5A and B). The pyrrolidine carboxamide showed two H-bonding interactions, the oxygen of carbonyl group on pyrrolidine makes H-bonds with active site amino acid Tyr158 (2.41 Å) and co-factor NAD^+ ribose (1.82 Å). Our attention was focused on the most characteristic receptor–ligand interactions of the most active analogue **9q**. As predicted by Surflex-Dock binding mode, compound **9q** showed that orientation and conformation is similar to that of the crystallographic ligand pyrrolidine carboxamide. The nitrogen atom at the 4-position of thiadiazole interact with the amino acid Try158 (2.01 Å) and co-factor NAD^+ (2.06 Å) (Fig. 6A). However, compound **9n** also binds in the same pattern compared to the crystallographic ligand pyrrolidine carboxamide. Here, each nitrogen of thiadiazole is making two H-bonds, one with amino acid Try158 (2.07, 2.66 Å) and another with co-factor NAD^+ (2.18, 2.27 Å) (Fig. 6B). In case of compounds viz., **6a**, **6b**, **6i**, **6k**, **8i**, **8k** and **8o**, instead of thiadiazole nitrogens, we found oxygen of bridge —OCH_2— making interaction at the substrate binding site. Representative docking conformations of compounds **6i** (—$CH_2O\cdots$HO-Tyr158, 2.50 Å and —$CH_2O\cdots$HO-NAD^+, 2.18 Å) and **8i** (—$CH_2O\cdots$HO-Tyr158, 2.13 Å and —$CH_2O\cdots$HO-NAD^+, 2.30 Å) are shown in Fig. 7A and B.

The docking scores viz., C-score, Crash, Polar, D_Score, PMF_Score, G_Score and Chem Score from Surflex dock are given in Table 3. Nine molecules were observed with better scores than the re-docked pyrrolidine carboxamide ligand. But the compound **9n** (C-score 10.19, MIC 6.25 μg/mL) showed the highest docking score compared to compound **9q** (C-score 9.11, MIC 3.125 μg/mL), instated of low lipophilicity than the **9q**. The hydrophobic and hydrophilic amino acids surrounded to aryloxy moiety (Gly192, Pro193, Ile194, Ala235, Thr236), bridge —OCH_2CH_2— (Phe149, Tyr158, Asn159, Ala191, Val238, Ala239, Lys240, Thr241), thiadiazole and pyrrole rings (Ser19, Ser20, Ile21, Ala22, Phe23, His24, Ile25, Ala26, Arg27, Gln30, Glu31, Gln32, Gln100, Thr101, Met103, Gly104, Asn106, Pro107, Thr196, Ala198) with the representative compound **9q** is depicted in Fig. 8A and B.

In general, it is observed that —CH_2O group and 1,3,4-thiadiazole moiety help to make H-bond with substrate binding site, while those of pyrrole and unsubstituted/substituted phenyl help to occupy or penetrate the molecules at the active sites.

3.4. CoMFA models

The pyrrolyl aryloxy 1,3,4-thiadiazole antitubercular agents were used to perform the CoMFA study. Two molecular superimposition maps were established: docked and minimized (database alignment) maps. The docked map was constructed by aligning the docking conformations directly. The minimized map was obtained by aligning the molecules after the docked ligands were energy-minimized. Chemical diversity method was used to split all the molecules into two sets, training and test set to perform the 3D-QSAR study on both the docked alignment and database alignment. The CoMFA model obtained from the training set showed good statistical significance (q^2 = 0.528/0.493, r^2 = 0.891/0.821, F_{ratio} = 189.879/145.273, and SEE = 0.120/0.153). However, linear regression of predicted activity and experimental activity data are shown in Fig. 9 for both the CoMFA models. The test set of 20 compounds were observed with the good predictive values, r^2_{pred} = 0.687/0.670 (Table 4). The actual, predicted pMIC values and residuals from the prediction of the training and test set compounds by the database and docking alignments for CoMFA models are shown in Table 5. The optimized maps are comparatively compact than the docked maps, while statistical indices are improved,

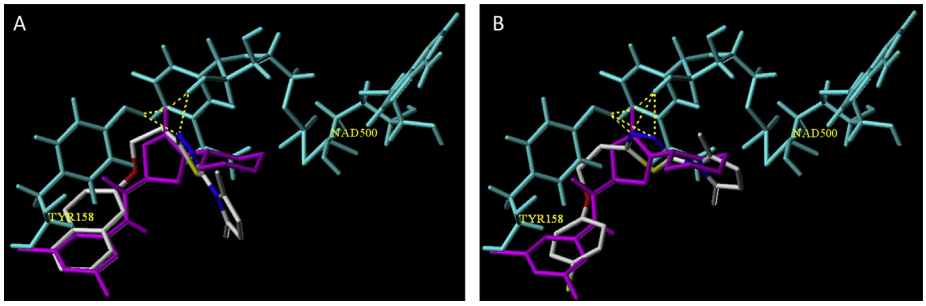


Fig. 5. Overlay of compound pyrrolidine carboxamide and **9q** (A) and **9n** (B) at active site.

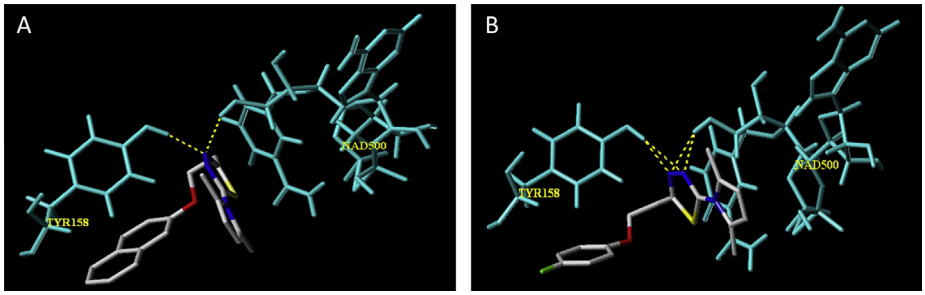


Fig. 6. Docking confirmation of compounds **9q** (A) and **9n** (B) at active site.

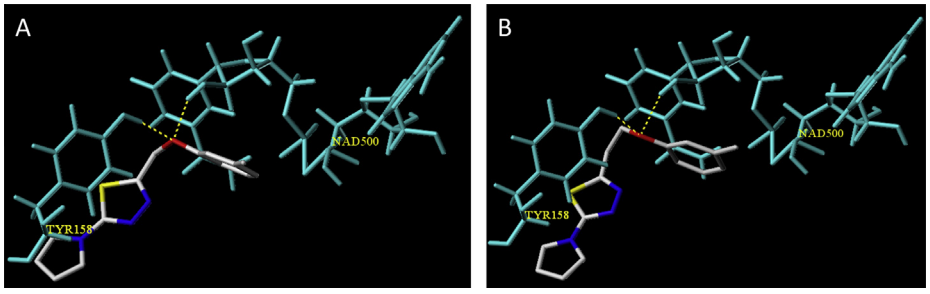


Fig. 7. Docking confirmation of compounds **6i** (A) and **8i** (B) at active site.

especially for crossvalidated q^2 from 0.493 to 0.528. The steric field contours are shown in yellow and green colors, while electrostatic field contours are shown in red and blue colors (Figs. 10 and 11). In the steric field graph, green contours represent the regions in which bulky groups confer an increase in activity, whereas yellow color represents regions where the bulky groups may lead to a decrease in activity. Similarly, in the map of electrostatic field, blue contours indicate regions where electropositive substitution increases the inhibitory activity, whereas red contours indicate the regions where electronegative substitution increases the activity. Compounds **9q** and **9n** of the training set are used as a template to depict the contour maps. It can be seen from the steric field contour for **9q** (Fig. 10A and B) that there is a large green polyhedron-like region around naphthyl, pyrrole and nitrogens of thiadiazole. The steric field contour for **9n** (Fig. 11A and B) shows a large green polyhedron-like region around benzene, pyrrole and nitrogens of thiadiazole. These observations suggest that substituent or hydrophobic or bulky groups near these green regions contribute positively to the activity of the compounds. Besides, yellow regions distribute less around the molecules.

Except for the influence of steric field, the potency of inhibitors is closely relevant to electrostatic characteristics of the substituent groups. The electrostatic contour maps (Figs. 10C, D and 11C, D) describe the relationship between electrical properties of substituents and bioactivities of the corresponding compounds. The potency is higher if more positive charge is near to the blue regions and more negative charge is near the red regions. The contribution of oxygen, nitrogen, fluorine to the activity due to electrostatic field in compounds **9q** and **9n** is eminent as shown in Figs. 10C–D and 11C–D.

4. Experimental section

Melting points were determined using the Shital-digital programmable melting point apparatus and are uncorrected. FTIR spectra in KBr pellets were recorded on a Bruker FTIR spectrophotometer. The ^{1}H and ^{13}C NMR spectra were recorded on a Bruker AVANCE II at 400 and 100/75 MHz, respectively; chemical shifts are expressed in parts per million (ppm) relative to TMS. The abbreviations used to describe the peak patterns are: (b) broad, (s) singlet, (d) doublet, (t) triplet, (q) quartet, and (m) multiplet. Mass spectra (MS) were recorded in a JEOL GCMATE II GC-Mass spectrometer and Schimadzu QP 20105 GC-Mass spectrometer. Analytical thin-layer chromatography (TLC) was performed on precoated TLC sheets of silica gel 60 F_{254} (Merck, Darmstadt, Germany) visualized by long- and short-wavelength ultraviolet (UV) lamps. Chromatographic purifications were performed on Merck aluminum oxide (70–230 mesh) and Merck silica gel (70–230 mesh).

Table 3
Surflex-Dock scores (kcal/mol) of pyrrolyl aryloxy 1,3,4-thiadiazole derivatives.

Compd code	C_Score	Crash	Polar	D_Score	PMF_ Score	G_Score	Chem Score
9n	10.19	−0.82	1.27	−158.572	−65.256	−298.689	−42.590
9q	9.11	−0.94	1.46	−149.208	−61.559	−273.175	−39.593
9m	9.08	−1.52	1.54	−164.068	−75.370	−297.962	−47.190
8m	9.02	−0.78	1.80	−149.171	−75.542	−270.589	−42.611
8q	8.95	−1.13	1.11	−144.725	−67.932	−280.222	−38.824
6p	8.95	−0.75	1.20	−148.555	−60.955	−270.336	−37.172
9j	8.88	−1.50	1.37	−151.599	−59.967	−281.051	−46.690
9h	8.87	−2.06	0.83	−163.739	−69.102	−319.692	−41.166
9p	8.75	−1.18	1.32	−158.721	−62.134	−305.232	−40.318
Pyrrolidine carboxamide	8.73	−1.39	1.18	−168.114	−49.191	−285.298	−37.478
9k	8.73	−1.19	0.89	−154.082	−59.500	−294.212	−36.214
9l	8.43	−0.76	1.26	−154.238	−63.701	−275.110	−39.256
9b	8.42	−0.64	1.24	−149.088	−64.222	−264.478	−37.599
9g	8.40	−1.23	1.13	−156.699	−56.602	−276.653	−37.729
8p	8.37	−1.56	1.67	−162.621	−70.322	−300.454	−42.876
8n	8.36	−0.80	1.03	−143.988	−57.534	−267.913	−35.074
9o	8.35	−1.05	1.14	−155.645	−66.897	−279.058	−41.066
7q	8.30	−1.44	1.19	−161.950	−72.903	−303.673	−42.697
8h	8.29	−1.04	1.78	−145.937	−62.226	−257.741	−36.927
9l	8.29	−0.84	1.24	−154.508	−69.417	−288.335	−42.442
9f	8.29	−1.29	1.19	−158.456	−55.011	−276.159	−40.146
9i	8.29	−1.26	1.32	−154.228	−74.728	−299.869	−41.631
9d	8.18	−1.36	0.54	−148.801	−50.483	−264.778	−33.726
8l	8.17	−0.90	1.77	−151.309	−69.147	−285.410	−40.983
7m	8.17	−1.04	1.09	−145.956	−68.894	−289.012	−36.836
8k	8.14	−1.03	1.84	−141.174	−55.937	−259.490	−38.331
7p	8.08	−1.51	1.09	−152.276	−71.372	−290.054	−41.200
9a	7.95	−0.80	1.17	−141.264	−73.224	−264.278	−37.821
7j	7.86	−1.53	1.36	−141.365	−66.044	−271.077	−36.008
7d	7.80	−1.25	1.32	−143.422	−59.110	−264.497	−35.999
9e	7.76	−1.34	1.31	−153.217	−62.741	−268.029	−40.437
6q	7.72	−1.66	1.80	−149.289	−74.087	−273.737	−40.557
7i	7.66	−1.33	1.31	−143.991	−70.217	−276.875	−40.212
8f	7.64	−0.91	1.86	−149.675	−73.234	−264.884	−39.993
8j	7.60	−1.15	0.00	−140.735	−56.320	−262.296	−30.376
8d	7.53	−0.75	1.63	−144.646	−70.040	−258.979	−36.595
7l	7.41	−1.63	1.11	−146.751	−68.825	−271.150	−37.705
7n	7.34	−0.72	0.59	−130.589	−63.900	−249.847	−29.532
6m	7.31	−1.54	1.81	−137.954	−70.301	−257.822	−36.796
7e	7.26	−0.69	1.22	−147.805	−74.195	−262.509	−36.966
7b	7.24	−0.69	1.13	−141.831	−72.767	−257.258	−36.937
8i	7.23	−1.06	1.84	−141.092	−69.715	−240.656	−37.691
8a	7.18	−0.66	1.71	−133.749	−71.744	−239.005	−34.521
6h	7.17	−0.71	0.71	−126.580	−56.893	−242.811	−29.570
7o	7.11	−1.06	1.24	−152.240	−72.786	−272.104	−38.796
7a	6.93	−0.60	1.20	−132.012	−72.776	−243.050	−37.751
8c	6.89	−0.51	1.57	−143.268	−71.228	−248.996	−37.046
7g	6.83	−1.31	1.08	−138.380	−61.439	−260.097	−34.565
6n	6.82	−0.73	0.00	−129.594	−44.117	−227.035	−27.013
6b	6.81	−0.81	0.49	−131.720	−47.419	−238.422	−28.332
8b	6.80	−0.64	1.51	−139.644	−77.150	−235.193	−37.744
7c	6.76	−0.56	1.26	−125.971	−53.267	−226.188	−36.741
8e	6.70	−1.68	1.80	−150.759	−71.505	−254.515	−40.067
6i	6.66	−0.90	1.17	−127.866	−60.934	−230.812	−30.956
6c	6.64	−1.07	1.83	−132.134	−70.092	−233.868	−35.716
8o	6.48	−0.67	1.21	−130.510	−64.371	−233.579	−34.324
6l	6.44	−0.68	1.05	−123.472	−40.233	−221.949	−28.618
6k	6.37	−0.87	0.73	−128.011	−59.272	−232.086	−31.005
8g	6.27	−1.48	1.53	−138.173	−32.533	−241.654	−30.672
7k	6.19	−1.65	0.91	−138.293	−51.645	−267.935	−37.893
6o	6.07	−1.56	1.73	−143.196	−74.085	−243.802	−37.073
6j	5.94	−0.80	0.00	−124.417	−44.544	−224.326	−26.623
6d	5.92	−0.64	0.84	−126.120	−50.924	−222.665	−26.394
6f	5.90	−1.61	0.90	−132.168	−51.247	−244.393	−29.708
7f	5.87	−1.02	1.14	−133.121	−57.891	−229.998	−35.262
6a	5.75	−1.20	1.39	−121.132	−58.970	−218.221	−30.424
6e	5.46	−0.53	0.73	−129.855	−45.093	−217.128	−27.518
6g	5.18	−0.63	0.59	−131.288	−42.197	−226.356	−26.712
7h	4.92	−0.59	0.00	−121.560	−54.044	−204.888	−26.325

*4.1. General procedure for the synthesis of aryloxy acetic/propionic acids (**2a–q** and **3a–q**)*

Equimolar quantities of 2-chloro acetic acid/3-chloro propionic acid (0.05 mol) and appropriate phenol (**1a–q**) (0.05 mol) were taken in a conical flask, to which aqueous solution of NaOH (0.12 mol in 25 mL water) was slowly added with constant stirring. The solution was stirred for 2 h until the solution turned clear, brown or yellow and then the reaction mixture was evaporated in a evaporating dish until the solid sodium salt was precipitated.

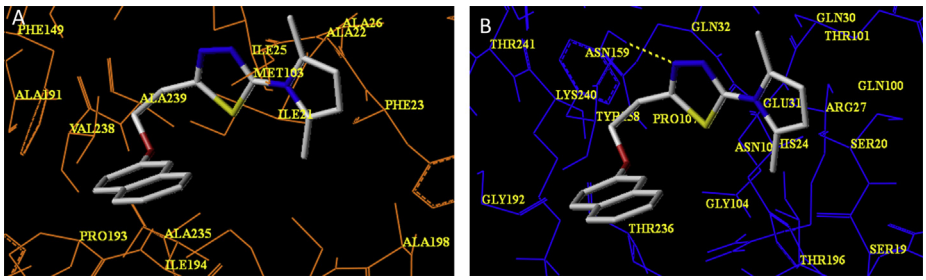


Fig. 8. Hydrophobic (A) and hydrophilic (B) amino acids surrounded to **9q**.

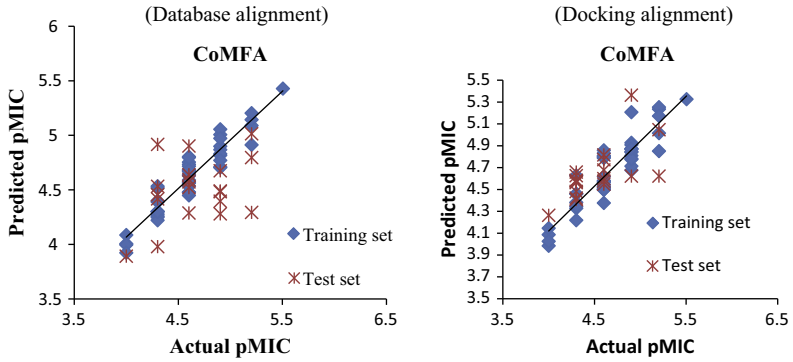


Fig. 9. Scatter plot diagrams for CoMFA analysis through database and docking alignment.

Table 4
PLS data summary.

Statistical parameters	CoMFA[a]	CoMFA[b]
q^2	0.528	0.493
No. of molecules in training set	48	48
No. of molecules in test set	20	20
ONC	05	04
SEE	0.120	0.153
r^2	0.891	0.821
F_{ratio}	189.879	145.273
r^2_{LOO}	0.888	0.866
r^2_{bs}	0.975	0.958
S.D.	0.004	0.011
r^2_{pred}	0.687	0.670
Fraction of field contributions		
Steric	0.303	0.240
Electrostatic	0.272	0.222

q^2, square of crossvalidated correlation coefficient; ONC, optimum number of components; SEE, standard error of estimate; S.D., standard deviation; r^2, square of non-crossvalidated correlation coefficient; F, $r^2/(1-r^2)$; r^2_{bs}, is mean r^2 of bootstrapping analysis (100 runs); SD_{bs}, is mean standard deviation by bootstrapping analysis; r^2_{pred}, predictive correlation coefficient;

[a] The training set of 48 molecules, database alignment is used in PLS analysis.

[b] The training set of 48 molecules, docked alignment is used in PLS analysis.

The salt was isolated, dried, dissolved in water and acidified by adding con. HCl. The precipitated aryloxy acetic/propionic acid was filtered and recrystallized from water or ethanol [38].

*4.2. General procedure for the preparation of 2-((phenoxy or substituted phenoxy)methyl/ethyl)-1,3,4-thiadiazol-5-amines (**4a–q** and **5a–q**) [23]*

A mixture of benzoic acid (50 mmol), *N*-aminothiourea (50 mmol) and $POCl_3$ (13 mL) was heated at 75 °C for 0.5 h. The mixture was cooled to which water (10 mL) was added and the reaction mixture was refluxed for 4 h. The mixture was cooled and pH was adjusted to 8.0 by adding 50% sodium hydroxide solution. The separated solid was filtered and recrystallized from ethanol to give desired compounds.

*4.3. General procedure for the preparation of 2-((phenoxy or substituted phenoxy)methyl/ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazoles (**6a–q** and **8a–q**)*

To a solution of 2-((phenoxy or substituted phenoxy)methyl/ethyl)-1,3,4-thiadiazol-5-amines (10 mmol) in 20 mL glacial acetic acid was added slowly 2,5-dimethoxytetrahydrofuran (15 mmol) at room temperature and the mixture was refluxed for 1 h (monitored by TLC). The reaction mixture was poured into ice cold water and basified with sodium bicarbonate solution. The separated solid was collected, washed with water and dried. The compounds were recrystallized from ethanol as a solvent.

*4.3.1. 2-(Phenoxymethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6a**)*

(Yield 69%). mp 79–81 °C; FTIR (KBr): 3147, 3124 (Ar—H), 1587 (C=N), 1244 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.42 (s, 2H, OCH_2), 6.37 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H), 6.99–7.05 (m, 4H, pyrrole-C_2, C_5—H and ph-C_2, C_6—H), 7.30–7.34 (m, 3H, ph-C_3, C_4, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.58, 162.60, 157.27, 129.76, 120.88, 114.84, 113.10, 64.84, MS (EI): m/z = found 257 [M^+]; calcd. 257.06.

*4.3.2. 2-((4-Chlorophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6b**)*

(Yield 65%). mp 115–117 °C; FTIR (KBr): 3098, 2926 (Ar—H), 1509 (C=N), 1226 (C—O—C), 832 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.39 (s, 2H, OCH_2), 6.37 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H,), 6.92, 6.96 (td, 2H, J = 2.28, 3.44, ph-C_3, C_5—H), 7.24–7.30 (m, 4H, pyrrole-C_2, C_5—H and ph-C_2, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.68, 161.86, 155.82, 129.66, 127.25, 120.89, 116.17, 113.22, 65.06; MS (EI): m/z = found 291 [M^+]; calcd. 291.02.

Table 5
Actual (Act) and predicted (Pred) pMIC values and residuals (Δ) of the database and docking aligned training set and test set molecules.

Comd	Database alignment			Comd	Database alignment			Comd	Docking alignment			Comd	Docking alignment		
	Act.	Pred.	Δ		Act.	Pred.	Δ		Act.	Pred.	Δ		Act.	Pred.	Δ
6a	4.000	4.086	−0.086	**8a**	4.301	4.518	−0.217	**6a**	4.000	4.263	−0.263	**8a**	4.301	4.581	−0.28
6b	4.301	4.303	−0.002	**8b**	4.602	4.804	−0.202	**6b**	4.301	4.572	−0.271	**8b**	4.602	4.832	−0.23
6c	4.000	4.006	−0.006	**8c**	4.602	4.684	−0.082	**6c**	4.000	4.086	−0.086	**8c**	4.602	4.564	0.038
6d*	4.000	3.892	0.108	**8d**	4.602	4.532	0.07	**6d***	4.000	3.984	0.016	**8d**	4.602	4.574	0.028
6e	4.301	4.277	0.024	**8e**	4.602	4.716	−0.114	**6e**	4.301	4.626	−0.325	**8e**	4.602	4.858	−0.256
6f	4.000	3.996	0.004	**8f***	4.602	4.639	−0.037	**6f**	4.000	4.145	−0.145	**8f***	4.602	4.529	0.073
6g	4.000	3.923	0.077	**8g**	4.602	4.575	0.027	**6g**	4.000	4.025	−0.025	**8g**	4.602	4.618	−0.016
6h	4.301	4.259	0.042	**8h**	4.903	4.710	0.193	**6h**	4.301	4.583	−0.282	**8h**	4.903	4.841	0.062
6i*	4.301	3.980	0.321	**8i***	4.602	4.594	0.008	**6i***	4.301	4.408	−0.107	**8i***	4.602	4.502	0.1
6j*	4.301	4.417	−0.116	**8j**	4.602	4.478	0.124	**6j***	4.301	4.218	0.083	**8j**	4.602	4.497	0.105
6k*	4.602	4.288	0.314	**8k**	4.903	4.899	0.004	**6k***	4.602	4.581	0.021	**8k**	4.903	4.846	0.057
6l	4.301	4.249	0.052	**8l***	4.602	4.521	0.081	**6l**	4.301	4.327	−0.026	**8l***	4.602	4.773	−0.171
6m	4.602	4.638	−0.036	**8m***	4.903	4.396	0.507	**6m**	4.602	4.513	0.089	**8m***	4.903	4.775	0.128
6n*	5.204	4.293	0.911	**8n**	5.204	5.143	0.061	**6n***	5.204	4.622	0.582	**8n**	5.204	5.047	0.157
6o*	4.903	4.280	0.623	**8o**	4.903	4.704	0.199	**6o***	4.903	4.676	0.227	**8o**	4.903	4.908	−0.005
6p	4.903	5.006	−0.103	**8p**	4.903	4.830	0.073	**6p**	4.903	4.868	0.035	**8p**	4.903	5.207	−0.304
6q	4.903	4.868	0.035	**8q**	4.903	5.055	−0.152	**6q**	4.903	4.927	−0.024	**8q**	4.903	5.363	−0.46
7a	4.301	4.390	−0.089	**9a**	4.602	4.569	0.033	**7a**	4.301	4.381	−0.08	**9a**	4.602	4.548	0.054
7b	4.602	4.754	−0.152	**9b***	4.602	4.902	−0.3	**7b**	4.602	4.787	−0.185	**9b***	4.602	4.794	−0.192
7c	4.301	4.221	0.08	**9c**	4.602	4.667	−0.065	**7c**	4.301	4.481	−0.18	**9c**	4.602	4.523	0.079
7d	4.602	4.449	0.153	**9d**	4.602	4.561	0.041	**7d**	4.602	4.376	0.226	**9d**	4.602	4.545	0.057
7e	4.602	4.733	−0.131	**9e**	4.602	4.796	−0.194	**7e**	4.602	4.808	−0.206	**9e**	4.602	4.816	−0.214
7f	4.301	4.299	0.002	**9f**	4.602	4.680	−0.078	**7f**	4.301	4.460	−0.159	**9f**	4.602	4.498	0.104
7g*	4.301	4.455	−0.154	**9g**	4.602	4.629	−0.027	**7g***	4.301	4.421	−0.12	**9g**	4.602	4.576	0.026
7h*	4.602	4.573	0.029	**9h**	4.602	4.590	0.012	**7h***	4.602	4.678	−0.076	**9h**	4.602	4.804	−0.202
7i	4.301	4.301	0	**9i***	4.301	4.916	−0.615	**7i**	4.301	4.628	−0.327	**9i***	4.301	4.460	−0.159
7j	4.301	4.533	−0.232	**9j***	4.301	4.532	−0.231	**7j**	4.301	4.563	−0.262	**9j***	4.301	4.345	−0.044
7k*	4.903	4.490	0.413	**9k**	4.903	4.972	−0.069	**7k***	4.903	4.713	0.19	**9k**	4.903	4.781	0.122
7l	4.301	4.400	−0.099	**9l**	4.602	4.479	0.123	**7l**	4.301	4.659	−0.358	**9l**	4.602	4.599	0.003
7m	4.903	4.816	0.087	**9m***	4.903	4.483	0.42	**7m**	4.903	4.810	0.093	**9m***	4.903	4.622	0.281
7n	5.204	4.912	0.292	**9n**	5.204	5.203	0.001	**7n**	5.204	4.851	0.353	**9n**	5.204	5.015	0.189
7o*	4.903	4.675	0.228	**9o**	4.903	4.770	0.133	**7o***	4.903	4.872	0.031	**9o**	4.903	4.872	0.031
7p	5.204	5.089	0.115	**9p***	5.204	4.796	0.408	**7p**	5.204	5.235	−0.031	**9p***	5.204	5.172	0.032
7q*	5.204	5.014	0.19	**9q**	5.505	5.428	0.077	**7q***	5.204	5.253	−0.049	**9q**	5.505	5.326	0.179

* Indicates test set compounds.

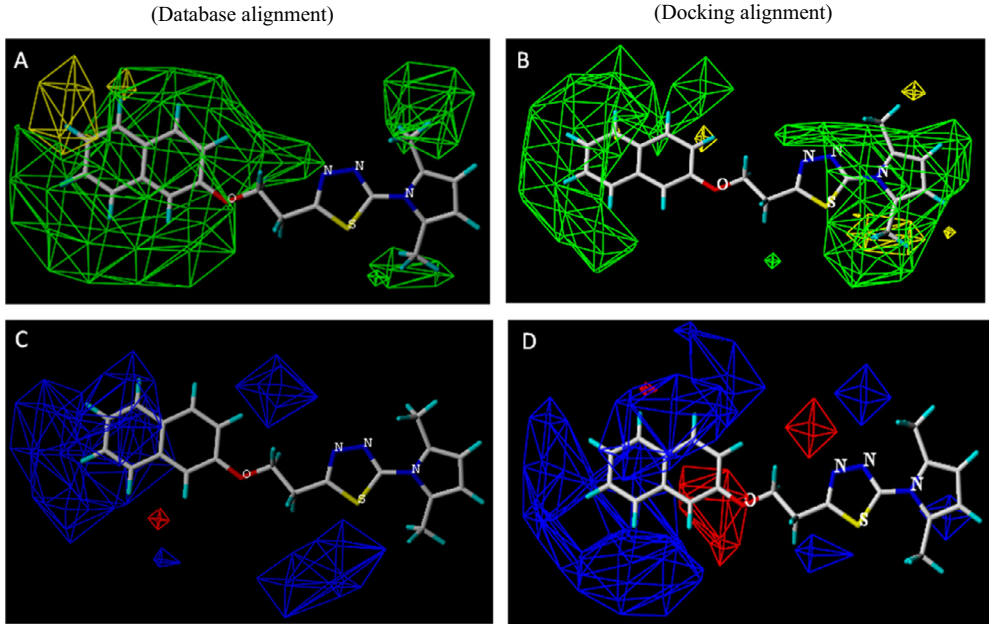


Fig. 10. CoMFA (A, B) steric and (C, D) electrostatic contour maps. The most active molecule, **9q** is displayed in the background.

*4.3.3. 2-((3-Chlorophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6c**)*

(Yield 65%). mp 118–120 °C; FTIR (KBr): 3120, 3028 (Ar—H), 1535 (C=N), 1242 (C—O—C), 723 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.42 (s, 2H, OCH_2), 6.32 (t, 2H, J = 2.18, pyrrole-C_3, C_4—H), 6.88 (s, 1H, Ph-C_6), 7.03 (s, 1H, ph-C_4), 7.21–7.30 (m, 4H, ph-C_2, C_5—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 162.57, 161.62, 153.93, 130.15, 127.09, 121.03, 120.33,

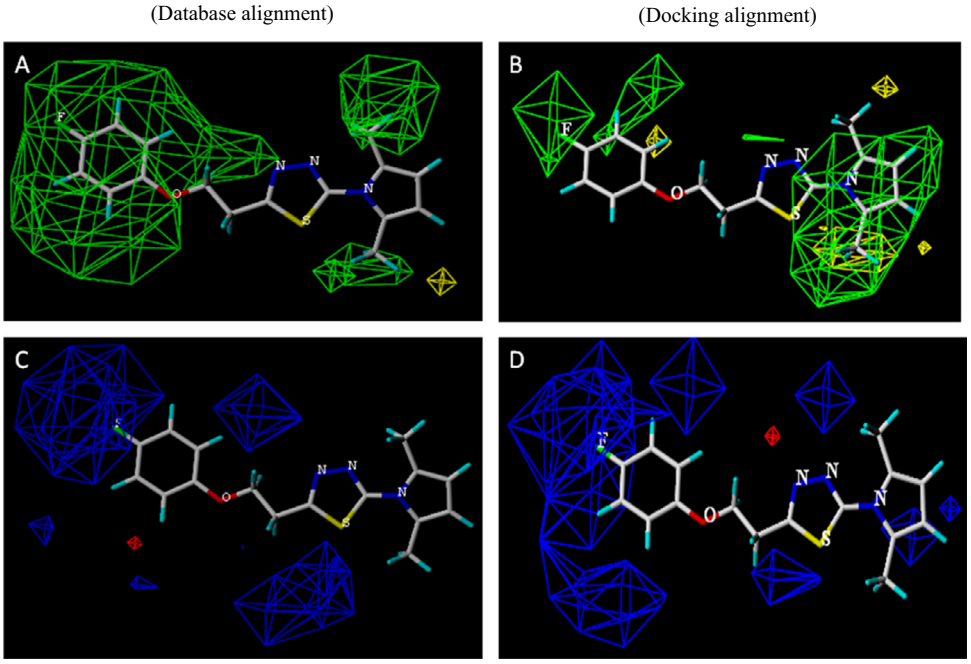


Fig. 11. CoMFA (A, B) steric and (C, D) electrostatic contour maps. The moderately active molecule, **9n** is displayed in the background.

117.21, 112.75, 110.42, 64.71; MS (EI): *m/z* = found 291 [M^+]; calcd. 291.02.

*4.3.4. 2-((2-Chlorophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6d**)*

(Yield 62%). mp 83–85 °C; FTIR (KBr): 3107, 3066 (Ar—H), 1590 (C=N), 1253 (C—O—C), 698 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.49 (s, 2H, OCH_2), 6.38 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H), 6.97–7.62 (m, 6H, ph-C_3, C_4, C_5, C_6—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 162.57, 161.62, 153.93, 130.15, 127.09, 121.03, 120.33, 117.21, 112.75, 110.42, 64.71; MS (EI): *m/z* = found 291 [M^+]; calcd. 291.02.

*4.3.5. 2-((4-Bromophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6e**)*

(Yield 67%). mp 119–121 °C; FTIR (KBr): 3099 (Ar—H), 1510 (C=N), 1288 (C—O—C), 830 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.40 (s, 2H, OCH_2), 6.38 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H), 6.89, 6.90 (dd, 2H, J = 2.16, 2.20, ph-C_2—H, C_6—H), 7.30 (t, 2H, J = 2.20, pyrrole-C_2, C_5—H), 7.41, 7.42 (dd, 2H, J = 2.20, 2.20, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.66, 161.81, 156.34, 132.62, 120.90, 116.67, 113.22, 64.99; MS (ESI): *m/z* = found 337.98 [M^++1], 338.98 [M^++2]; calcd. 336.21.

*4.3.6. 2-((3-Bromophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6f**)*

(Yield 65%). mp 111–113 °C; FTIR (KBr): 3127, 3065 (Ar—H), 1529 (C=N), 1252 (C—O—C), 825 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.41 (s, 2H, OCH_2), 6.33 (t, 2H, J = 2.08, pyrrole-C_3, C_4—H), 6.82 (s, 1H, ph-C_6—H), 7.02 (s, 1H, ph-C_4—H), 7.18–7.29 (m, 4H, ph-C_2, C_5—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.12, 162.29, 161.75, 130.16, 123.25, 121.37, 120.65, 116.92, 113.22, 111.61, 65.13; MS (ESI): *m/z* = found 337.98 [M^++1]; calcd. 336.21.

*4.3.7. 2-((2-Bromophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6g**)*

(Yield 65%). mp 100–102 °C; FTIR (KBr): 3162, 3055 (Ar—H), 1552 (C=N), 1289 (C—O—C), 827 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.45 (s, 2H, OCH_2), 6.33 (t, 2H, J = 2.12, pyrrole-C_3, C_4—H), 6.87–6.90 (m, 2H, ph-C_4, C_6—H), 7.20–7.27 (m, 3H, pyrrole-C_2, C_5—H and ph-C_5—H), 7.42–7.45 (m, 1H, ph-C_3—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 162.89, 161.59, 156.12, 131.29, 127.32, 121.01, 120.12, 114.07, 112.29, 111.15, 64.56; MS (ESI): *m/z* = found 337.98 [M^++1]; calcd. 336.21.

*4.3.8. 2-(1H-Pyrrol-1-yl)-5-(4-tolyloxymethyl)-1,3,4-thiadiazole (**6h**)*

(Yield 70%). mp 98–100 °C; FTIR (KBr): 3145, 3099 (Ar—H), 1588 (C=N), 1218, 1247 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.23 (s, 3H, CH_3), 5.38 (s, 2H, OCH_2), 6.36 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H), 6.88–6.91 (m, 2H, ph-C_2, C_6—H), 7.11 (m, 2H, J = 8.68, ph-C_3, C_5—H), 7.29 (t, 2H, J = 2.28, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.51, 162.87, 155.15, 131.59, 130.16, 120.15, 114.66, 113.33, 65.00, 20.45; MS (EI): *m/z* = found 271 [M^+]; calcd. 271.08.

*4.3.9. 2-(1H-Pyrrol-1-yl)-5-(3-tolyloxymethyl)-1,3,4-thiadiazole (**6i**)*

(Yield 61%). mp 68–70 °C; FTIR (KBr): 3128, 3057 (Ar—H), 1597 (C=N), 1217 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.35 (s, 3H, CH_3), 5.41 (s, 2H,OCH_2), 6.37 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H), 6.79–6.85 (m, 3H, ph-C_2, C_4, C_6—H), 7.19 (t, 1H, J = 7.76, ph-C_5—H), 7.30 (t, 2H, J = 2.20, pyrrole-C_2—H, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.50, 162.78, 157.26, 139.93, 129.95, 123.02, 120.85, 115.66, 113.05, 111.61, 64.77, 21.45; MS (EI): *m/z* = found 271 [M^+]; calcd. 271.08.

*4.3.10. 2-(1H-Pyrrol-1-yl)-5-(2-tolyloxymethyl)-1,3,4-thiadiazole (**6j**)*

(Yield 62%). mp 89–91 °C; FTIR (KBr): 2919 (Ar—H), 1510 (C=N), 1240 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.28 (s, 3H, CH_3), 5.42 (s, 2H, OCH_2), 6.38 (t, 2H, J = 2.20, pyrrole-C_3, C_4—H), 6.96 (m, 2H, ph-C_4, C_5—H), 7.17 (t, 2H, J = 7.28, ph-C_3, C_6—H), 7.31 (t, 2H, J = 2.20, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.49, 163.03, 155.46, 131.13, 127.05,

121.91, 120.90, 113.10, 111.28, 64.92, 16.25; MS (EI): *m/z* = found 271 [M^+]; calcd. 271.08.

*4.3.11. 2-((3,5-Dimethylphenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6k**)*

(Yield 60%). mp 103–105 °C; FTIR (KBr): 3112, 3016 (Ar—H), 1553 (C=N), 1247 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.34, (s, 6H, $2CH_3$), 5.41 (s, 2H, OCH_2), 6.33 (t, 2H, *J* = 2.12, pyrrole-C_3, C_4—H), 6.62 (m, 2H, ph-C_3, C_6—H), 6.90–6.93 (m, 1H, ph-C_4—H), 7.30 (t, 2H, *J* = 2.18, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.32, 161.26, 160.03, 140.94, 123.77, 120.26, 113.08, 111.65, 64.57, 21.49; MS (EI): *m/z* = found 285 [M^+]; calcd. 285.09.

*4.3.12. 2-((2,5-Dimethylphenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6l**)*

(Yield 60%). mp 92–94 °C; FTIR (KBr): 3108, 3026 (Ar—H), 1548 (C=N), 1235 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.20 (s, 3H, CH_3), 2.34 (s, 3H, CH_3), 5.39 (s, 2H, OCH_2), 6.34 (t, 2H, *J* = 2.24, pyrrole-C_3, C_4—H), 6.65 (s, 1H, ph-C_4—H), 6.90–6.92 (m, 1H, ph-C_6—H), 7.03–7.06 (m, 1H, ph-C_3—H), 7.28 (t, 2H, *J* = 2.24, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.13, 161.29, 156.89, 136.58, 130.06, 123.57, 120.27, 120.08, 117.16, 113.10, 65.12, 21.45, 16.01; MS (EI): *m/z* = found 285 [M^+]; calcd. 285.09.

*4.3.13. 2-((2,4-Dimethylphenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6m**)*

(Yield 60%). mp 118–120 °C; FTIR (KBr): 3123, 3007 (Ar—H), 1519 (C=N), 1255 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.21 (s, 3H, CH_3), 2.33 (s, 3H, CH_3), 5.41 (s, 2H, OCH_2), 6.33 (t, 2H, *J* = 2.08, pyrrole-C_3, C_4—H), 6.73–7.00 (m, 3H, ph-C_3, C_5, C_6—H), 7.29 (t, 2H, *J* = 2.10, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.19, 161.25, 155.73, 131.95, 130.79, 127.06, 126.88, 120.11, 113.09, 111.56, 65.34, 21.43, 16.00; MS (EI): *m/z* = found 285 [M^+]; calcd. 285.09.

*4.3.14. 2-((4-Fluorophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6n**)*

(Yield 62%). mp 80–82 °C; FTIR (KBr): 3143, 3075 (Ar—H), 1571 (C=N), 1250 (C—O—C), 756 (C—F); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.39 (s, 2H, OCH_2), 6.38 (t, 2H, *J* = 2.24, pyrrole-C_3, C_4—H), 6.93–7.03 (m, 4H, ph-C_2, C_3, C_5, C_6—H), 7.30 (t, 2H, *J* = 2.24, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.63, 161.92, 155.86, 154.60, 120.85, 116.59, 115.82, 112.29, 65.21; MS (EI): *m/z* = found 275 [M^+]; calcd. 275.05.

*4.3.15. 2-((4-Iodophenoxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6o**)*

(Yield 60%). mp 115–117 °C; FTIR (KBr): 3140, 3064 (Ar—H), 1558 (C=N), 1255 (C—O—C), 719 (C—I); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.41 (s, 2H, OCH_2), 6.33 (t, 2H, *J* = 2.12, pyrrole-C_3—H, C_4—H), 6.82–6.85 (m, 2H, ph-C_2, C_6—H), 7.29 (t, 2H, *J* = 2.20, pyrrole-C_2, C_5—H), 7.53–7.56 (m, 2H, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.59, 161.85, 156.19, 138.62, 120.77, 116.07, 113.06, 83.59, 65.19; MS (EI): *m/z* = found 382 [M^+]; calcd. 382.96.

*4.3.16. 2-((Naphthalen-1-yloxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6p**)*

(Yield 58%). mp 88–90 °C; FTIR (KBr): 3051, 2917 (Ar—H), 1535 (C=N), 1240 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.62 (s, 2H, OCH_2), 6.38 (t, 2H, *J* = 2.12, pyrrole-C_3, C_4—H), 6.77–8.29 (m, 9H, naphthyl-C_2, C_3, C_4, C_5, C_6, C_7, C_8—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.61, 162.42, 155.22, 135.10, 134.18, 129.93, 129.50, 127.64, 127.06, 126.71, 124.38, 120.87, 118.32, 118.27, 113.31, 113.12, 108.75, 64.92; MS (ESI): *m/z* = found 308.09 [M^++1]; calcd. 307.08.

*4.3.17. 2-((Naphthalen-2-yloxy)methyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**6q**)*

(Yield 58%). mp 120–122 °C; FTIR (KBr): 2954 (Ar—H), 1507 (C=N), 1241 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 5.50 (s, OCH_2), 6.35 (t, *J* = 2.16, pyrrole-C_3, C_4—H), 6.73–7.79 (m, 7H, naphthyl-C_1, C_3, C_4, C_5, C_6, C_7, C_9—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.59, 162.40, 156.02, 131.26, 130.59, 129.87, 127.82, 126.89, 126.73, 124.11, 120.80, 118.83, 113.22, 105.94, 64.76; MS (EI): *m/z* = found 308.09 [M^++1]; calcd. 307.08.

*4.3.18. 2-(2-Phenoxyethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8a**)*

(Yield 67%). mp 90–92 °C; FTIR (KBr): 3124, 2924 (Ar—H), 1592 (C=N), 1294 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.52 (t, 2H, *J* = 5.84, CH_2), 4.31 (t, 2H, *J* = 5.80, OCH_2), 6.34 (t, 2H, *J* = 2.28, pyrrole-C_3, C_4—H), 6.91–7.00 (m, 3H, ph-C_2, C_4, C_6—H), 7.27–7.33 (m, 4H, pyrrole-C_2, C_5—H and ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.22, 161.93, 158.01, 129.69, 121.58, 120.84, 114.60, 112.82, 65.50, 30.99; MS (EI): *m/z* = found 271 [M^+]; calcd. 271.08.

*4.3.19. 2-(2-(4-Chlorophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8b**)*

(Yield 62%). mp 85–87 °C; FTIR (KBr): 3107, 2923 (Ar—H), 1545 (C=N), 1254 (C—O—C), 820 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.50 (t, 2H, *J* = 5.82, CH_2), 4.30 (t, 2H, *J* = 5.80, OCH_2), 6.33 (t, 2H, *J* = 2.20, pyrrole-C_3, C_4—H), 7.01 (d, 2H, *J* = 7.75, ph-C_2, C_6—H), 7.26 (t, 2H, *J* = 2.22, pyrrole-C_2, C_5—H), 7.37 (d, 2H, *J* = 7.76, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.22, 161.93, 158.01, 129.69, 121.58, 120.84, 114.60, 112.82, 65.50, 30.99; MS (EI): *m/z* = found 305 [M^+]; calcd. 305.04.

*4.3.20. 2-(2-(3-Chlorophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8c**)*

(Yield 60%). mp 80–82 °C; FTIR (KBr): 3118, 2968 (Ar—H), 1557 (C=N), 1235 (C—O—C), 727 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.52 (t, 2H, *J* = 5.82, CH_2), 4.33 (t, 2H, *J* = 5.84, OCH_2), 6.34 (t, 2H, *J* = 2.18, pyrrole-C_3, C_4—H), 6.89 (s, 1H, Ph-C_6), 7.07 (s, 1H, Ph-C_4), 7.20–7.32 (m, 4H, ph-C_2, C_5—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.09, 161.81, 153.24, 130.58, 126.99, 121.26, 120.39, 117.09, 112.63, 110.21, 65.42, 31.29; MS (EI): *m/z* = found 305 [M^+]; calcd. 305.04.

*4.3.21. 2-(2-(2-Chlorophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8d**)*

(Yield 62%). mp 79–81 °C; FTIR (KBr): 3125, 3016 (Ar—H), 1588 (C=N), 1265 (C—O—C), 725 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.55 (t, 2H, *J* = 5.84, CH_2), 4.36 (t, 2H, *J* = 5.84, OCH_2), 6.31 (t, 2H, *J* = 2.20, pyrrole-C_3, C_4—H), 6.93 (s, 1H, ph-C_4), 7.11–7.58 (m, 5H, ph-C_3, C_5, C_6—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.11, 161.72, 153.58, 130.29, 127.10, 121.18, 120.52, 117.13, 112.70, 110.36, 65.14, 31.13; MS (EI): *m/z* = found 305 [M^+]; calcd. 305.04.

*4.3.22. 2-(2-(4-Bromophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8e**)*

(Yield 64%). mp 92–94 °C; FTIR (KBr): 3121, 3051 (Ar—H), 1548 (C=N), 1259 (C—O—C), 829 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.53 (t, 2H, *J* = 5.88, CH_2), 4.33 (t, 2H, *J* = 5.84, OCH_2), 6.32 (t, 2H, *J* = 2.16, pyrrole-C_3, C_4—H), 6.83 (d, 2H, *J* = 5.20, ph-C_2, C_6—H), 7.29 (t, 2H, *J* = 2.20, pyrrole-C_2, C_5—H), 7.39 (d, 2H, *J* = 5.12, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.58, 161.90, 156.13, 131.91, 121.03, 117.19, 112.88, 65.19, 31.08; MS (ESI): *m/z* = found 351.93 [M^++1]; calcd. 350.23.

*4.3.23. 2-(2-(3-Bromophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8f**)*

(Yield 60%). mp 98–100 °C; FTIR (KBr): 3159, 3038 (Ar—H), 1571 (C=N), 1258 (C—O—C), 820 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.50 (t, 2H, J = 5.86, CH_2), 4.37 (t, 2H, J = 5.84, OCH_2), 6.34 (t, 2H, J = 2.18, pyrrole-C_3, C_4—H), 6.89 (s, 1H, ph-C_6—H), 7.11 (s, 1H, ph-C_4—H), 7.23–7.37 (m, 4H, ph-C_2, C_5—H and pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.41, 161.98, 161.52, 131.36, 123.18, 121.06, 120.54, 117.03, 112.56, 112.95, 65.42, 31.19; MS (ESI): m/z = found 351.93 [M^++1]; calcd. 350.23.

*4.3.24. 2-(2-(2-Bromophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8g**)*

(Yield 60%). mp 78–80 °C; FTIR (KBr): 3109, 2924 (Ar—H), 1525 (C=N), 1246 (C—O—C), 830 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.51 (t, 2H, J = 5.80, CH_2), 4.28 (t, 2H, J = 5.84, OCH_2), 6.35 (t, 2H, J = 2.22, pyrrole-C_3, C_4—H), 6.80, 6.82 (td, 2H, J = 2.22, 1.96, pyrrole-C_2, C_6—H), 7.28 (d, 2H, J = 8.23, ph-C_2, C_5—H), 7.37, 7.39 (td, 2H, J = 2.22, 1.96, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.15, 161.51, 157.14, 132.49, 120.84, 116.37, 113.81, 112.84, 65.86, 30.84; MS (ESI): m/z = found 349.88 [M^+-1]; calcd. 350.23.

*4.3.25. 2-(1H-Pyrrol-1-yl)-5-(2-(4-tolyloxy)ethyl)-1,3,4-thiadiazole (**8h**)*

(Yield 65%). mp 70–72 °C; FTIR (KBr): 3098, 2926 (Ar—H), 1588 (C=N), 1234 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.29 (s, 3H, CH_3), 3.58 (t, 1H, J = 5.80, CH_2), 4.29 (t, 2H, J = 5.80, OCH_2), 6.35 (t, 2H, J = 2.20, pyrrole-C_3, C_4—H), 6.82–6.85 (d, 2H, ph-C_2, C_5—H), 7.11 (d, 2H, J = 8.40, ph-C_2, C_5—H), 7.30 (t, 2H, J = 2.20, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.22, 162.05, 156.20, 130.88, 120.83, 112.84, 108.32, 65.67, 31.03, 20.54; MS (EI): m/z = found 285 [M^+]; calcd. 285.09.

*4.3.26. 2-(1H-Pyrrol-1-yl)-5-(2-(3-tolyloxy)ethyl)-1,3,4-thiadiazole (**8i**)*

(Yield 59%). mp 90–92 °C; FTIR (KBr): 3033, 2926 (Ar—H), 1591 (C=N), 1261 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.33 (s, 3H, CH_3), 3.53 (t, 2H, J = 5.80, CH_2), 4.30 (t, 2H, J = 5.76, OCH_2), 6.35 (t, 2H, J = 2.12, pyrrole-C_3, C_4—H), 6.76 (t, 1H, J = 3.76, ph-C_4—H), 6.81 (d, 1H, J = 7.48, ph-C_2—H), 7.18 (d, 1H, J = 7.80, ph-C_6—H), 7.24 (s, 1H, ph-C_5—H), 7.30 (t, 2H, J = 2.12, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.29, 162.57, 157.22, 139.90, 130.01, 123.00, 120.83, 115.51, 113.12, 111.60, 65.12, 31.07, 21.50; MS (EI): m/z = found 285 [M^+]; calcd. 285.09.

*4.3.27. 2-(1H-Pyrrol-1-yl)-5-(2-(2-tolyloxy)ethyl)-1,3,4-thiadiazole (**8j**)*

(Yield 60%). mp 76–78 °C; FTIR (KBr): 3021, 2923 (Ar—H), 1592 (C=N), 1296 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.28 (s, 3H, CH_3), 3.56 (t, 2H, J = 5.76, CH_2), 4.31 (t, 2H, J = 5.76, OCH_2), 6.35 (t, 2H, J = 2.16, pyrrole-C_3, C_4—H), 6.82 (d, 1H, J = 8.40, ph-C_5—H), 6.89 (t, 2H, J = 7.36, ph-C_4), 7.15 (t, 2H, J = 7.24, ph-C_3, C_6—H), 7.28 (t, 2H, J = 2.16, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.13, 162.04, 156.20, 130.98, 126.97, 121.17, 120.97, 112.84, 110.85, 65.57, 31.10, 16.57; MS (EI): m/z = found 285 [M^+]; calcd. 285.09.

*4.3.28. 2-(2-(3,5-Dimethylphenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8k**)*

(Yield 55%). mp 97–99 °C; FTIR (KBr): 3122, 3008 (Ar—H), 1562 (C=N), 1236 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.30 (s, 6H, $2CH_3$), 3.53 (t, 2H, J = 5.80, CH_2), 5.44 (t, 2H, J = 5.80, OCH_2), 6.32 (t, 2H, J = 2.18, pyrrole-C_3, C_4—H), 6.66 (m, 2H, ph-C_3, C_6—H), 6.92–6.95 (m, 1H, ph-C_4—H), 7.33 (t, 2H, J = 2.22, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.28, 161.19, 160.00, 140.97, 123.65, 120.21, 113.02, 111.61, 65.39, 21.47; MS (EI): m/z = found 299 [M^+]; calcd. 299.11.

*4.3.29. 2-(2-(2,5-Dimethylphenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8l**)*

(Yield 55%). mp 100–102 °C; FTIR (KBr): 3171, 2920 (Ar—H), 1571 (C=N), 1261 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.14 (s, 3H, CH_3), 2.26 (s, 3H, CH_3), 3.57 (t, 2H, J = 5.78, CH_2), 4.31 (t, 2H, J = 5.80, OCH_2), 6.35 (t, 2H, J = 2.18, pyrrole-C_3, C_4—H), 6.64 (d, 1H, J = 6.59, ph-C_4—H), 6.72 (d, 1H, J = 7.03, ph-C_6—H), 7.04 (d, 1H, J = 7.03, ph-C_3—H), 7.28 (t, 2H, J = 2.24, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.11, 161.09, 156.72, 136.29, 130.67, 123.45, 121.78, 116.06, 111.90, 65.51, 31.13, 21.09, 16.09; MS (EI): m/z = found 299 [M^+]; calcd. 299.11.

*4.3.30. 2-(2-(2,4-Dimethylphenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8m**)*

(Yield 55%). mp 93–95 °C; FTIR (KBr): 3118, 3020 (Ar—H), 1535 (C=N), 1239 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.22 (s, 3H, CH_3), 2.33 (s, 3H, CH_3), 3.56 (t, 2H, J = 5.82, CH_2), 5.37 (t, 2H, J = 5.78, OCH_2), 6.31 (t, 2H, J = 2.10, pyrrole-C_3, C_4—H), 6.69–6.98 (m, 3H, ph-C_3, C_5, C_6—H), 7.36 (t, 2H, J = 2.10, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.22, 161.11, 155.62, 131.83, 130.14, 127.11, 126.93, 120.10, 113.13, 111.49, 65.55, 31.21, 21.59, 16.03; MS (EI): m/z = found 299 [M^+]; calcd. 299.11.

*4.3.31. 2-(2-(4-Fluorophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8n**)*

(Yield 59%). mp 98–100 °C; FTIR (KBr): 3140, 3072 (Ar—H), 1565 (C=N), 1253 (C—O—C), 753 (C—F); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 3.59 (t, 2H, J = 5.80, CH_2), 5.41 (t, 2H, J = 5.84, OCH_2), 6.35 (t, 2H, J = 2.12, pyrrole-C_3, C_4—H), 6.94–7.05 (m, 4H, ph-C_2, C_3, C_5, C_6—H), 7.32 (t, 2H, J = 2.18, pyrrole-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.60, 161.89, 155.85, 154.62, 121.01, 116.60, 115.89, 112.33, 65.37, 30.88; MS (EI): m/z = found 289 [M^+]; calcd. 289.07.

*4.3.32. 2-(2-(4-Iodophenoxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8o**)*

(Yield 55%). mp 102–104 °C; FTIR (KBr): 3138, 3052 (Ar—H), 1565 (C=N), 1249 (C—O—C), 721 (C—I); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.95 (t, 2H, J = 5.74, CH_2), 4.53 (t, 2H, J = 5.76, OCH_2), 6.34 (t, 2H, J = 2.18, pyrrole-C_3, C_4—H), 6.85 (d, 2H, J = 7.76, ph-C_2, C_6—H), 7.31 (t, 2H, J = 2.22, pyrrole-C_2, C_5—H), 7.55 (d, 2H, J = 7.76, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.55, 161.69, 156.20, 138.60, 120.79, 116.08, 113.09, 83.56, 65.30, 31.01; MS (EI): m/z = found 396 [M^+]; calcd. 396.97.

*4.3.33. 2-(2-(Naphthalen-1-yloxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8p**)*

(Yield 58%). mp 80–82 °C; FTIR (KBr): 3056, 2925 (Ar—H), 1597 (C=N), 1264 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.92 (t, 2H, J = 5.70, CH_2), 4.57 (t, 2H, J = 5.74, OCH_2), 6.35 (s, 2H, pyrrole-C_2, C_3—H), 6.86 (s, 2H, ph-C_2—H), 7.28 (s, 2H, pyrrole-C_3, C_4—H), 7.54–8.32 (m, 7H, naphthyl-C_2, C_3, C_4, C_5, C_6, C_7, C_8—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.61, 162.42, 155.22, 135.1, 134.18, 129.93, 129.50, 127.64, 127.00, 126.71, 126.82, 123.83, 118.32, 118.27, 113.31, 113.12, 106.91, 63.06, 34.31; MS (EI): m/z = found 321 [M^+]; calcd. 321.09.

*4.3.34. 2-(2-(Naphthalen-2-yloxy)ethyl)-5-(1H-pyrrol-1-yl)-1,3,4-thiadiazole (**8q**)*

(Yield 55%). mp 86–88 °C; FTIR (KBr): 3140, 2950 (Ar—H), 1565 (C=N), 1235 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.95 (t, 2H, J = 5.74, CH_2), 4.59 (t, 2H, J = 5.74, OCH_2), 6.33 (t, 2H, J = 2.24, pyrrole-C_3, C_4—H), 6.70–7.80 (m, 7H, naphthyl-C_1, C_3, C_4, C_5, C_6,

C_7, C_9—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 163.63, 162.48, 155.99, 131.18, 130.57, 129.95, 127.96, 126.75, 126.69, 124.12, 120.83, 118.89, 113.20, 105.93, 64.12, 33.87; MS (EI): *m/z* = found 321 [M^+]; calcd. 321.09.

*4.4. General procedure for the preparation of 2-((substituted phenoxy) methyl/ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazoles (**7a–q** and **9a–q**)*

To a solution of 2-((phenoxy or substituted phenoxy)methyl/ethyl)-1,3,4-thiadiazol-5-amines (10 mmol) in 20 mL glacial acetic acid was added slowly acetonyl acetone (15 mmol) at room temperature and the mixture was refluxed for 1 h (monitored by TLC). The reaction mixture was poured into ice cold water and basified with sodium bicarbonate solution. The separated solid was collected, washed with water and dried. The compounds were recrystallized with *n*-hexane as a solvent.

*4.4.1. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(phenoxymethyl)-1,3, 4-thiadiazole (**7a**)*

(Yield 63%). mp 59–61 °C; FTIR (KBr): 2951 (Ar—H), 1588 (C=N), 1250 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 5.48 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 7.01–7.06 (m, 3H, ph-C_3, C_4, C_5—H), 7.32–7.36 (m, 2H, ph-C_2, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.50, 161.81, 157.42, 130.19, 129.82, 129.71, 122.32, 114.90, 109.20, 65.35, 13.45; MS (EI): *m/z* = found 285 [M^+]; calcd. 285.09.

*4.4.2. 2-((4-Chlorophenoxy)methyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**7b**)*

(Yield 60%). mp 98–100 °C; FTIR (KBr): 3070, 2955 (Ar—H), 1582 (C=N), 1249 (C—O—C), 823 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.26 (s, 6H, $2CH_3$), 5.45 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 6.95, 6.97 (dd, 2H, *J* = 2.22, 1.98, ph-C_2, C_6—H), 7.28–7.29 (dd, 2H, *J* = 2.20, 2.20, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.38, 161.14, 156.16, 129.29, 129.12, 125.85, 116.45, 108.89, 64.59, 12.97; MS (EI): *m/z* = found 319 [M^+]; calcd. 319.05.

*4.4.3. 2-((3-Chlorophenoxy)methyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**7c**)*

(Yield 60%). mp 103–105 °C; FTIR (KBr): 3118, 3056 (Ar—H), 1574 (C=N), 1247 (C—O—C), 729 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.23 (s, 6H, $2CH_3$), 5.47 (s, 2H, OCH_2), 5.91 (s, 2H, pyrrole-C_3, C_4—H), 6.89 (s, 1H, ph-C_6), 7.07 (s, 1H, ph-C_4), 7.25–7.29 (m, 2H, ph-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 166.89, 161.09, 153.77, 130.00, 127.59, 127.12, 120.17, 119.81, 117.05, 109.18, 110.42, 65.23, 13.41; MS (EI): *m/z* = found 319 [M^+]; calcd. 319.05.

*4.4.4. 2-((2-Chlorophenoxy)methyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**7d**)*

(Yield 60%). mp 85–87 °C; FTIR (KBr): 3066, 2920 (Ar—H), 1540 (C=N), 1244 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.26 (s, 6H, $2CH_3$), 5.54 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 6.99, 7.01, 7.03 (dt, 1H, *J* = 1.28, 1.24, 1.36, ph-C_4—H), 7.08, 7.10 (dd, 1H, *J* = 1.24, 1.32, ph-C_6—H), 7.24, 7.26, 7.28 (dt, 1H, *J* = 1.52, 3.08, 1.56, ph-C_4—H), 7.40–7.42 (dd, 1H, *J* = 1.60, 1.56, ph-C_3—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 166.96, 162.06, 153.08, 130.74, 130.18, 128.01, 123.59, 123.00, 114.58, 109.25, 66.56, 13.45; MS (EI): *m/z* = found 319 [M^+]; calcd. 319.05.

*4.4.5. 2-((4-Bromophenoxy)methyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**7e**)*

(Yield 62%). mp 101–103 °C; FTIR (KBr): 3113, 3053 (Ar—H), 1552 (C=N), 1262 (C—O—C), 835 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.27 (s, 6H, $2CH_3$), 5.51 (s, 2H, OCH_2), 5.90 (s, 2H, pyrrole-C_3, C_4—H), 6.93–6.96 (m, 2H, ph-C_2, C_6—H), 7.40–7.45 (m, 2H, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.60, 162.52, 157.01, 132.53, 127.25, 116.50, 109.35, 65.01, 13.42; MS (ESI): *m/z* = found 365.95 [M^++1], 366.95 [M^++2]; calcd. 364.26.

*4.4.6. 2-((3-Bromophenoxy)methyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**7f**)*

(Yield 60%). mp 89–91 °C; FTIR (KBr): 3112, 3052 (Ar—H), 1549 (C=N), 1248 (C—O—C), 821 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.22 (s, 6H, $2CH_3$), 5.55 (s, 2H, OCH_2), 5.89 (s, 2H, pyrrole-C_3, C_4—H), 6.87 (s, 1H, ph-C_6—H), 7.10 (s, 1H, ph-C_4—H), 7.22–7.25 (m, 2H, ph-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.52, 161.69, 160.17, 130.28, 127.32, 123.20, 120.53, 117.00, 111.60, 109.19, 65.17, 13.48; MS (ESI): *m/z* = found 365.95 [M^++1], 366.95 [M^++2]; calcd. 364.26.

*4.4.7. 2-((2-Bromophenoxy)methyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**7g**)*

(Yield 60%). mp 86–88 °C; FTIR (KBr): 3159, 3042 (Ar—H), 1545 (C=N), 1234 (C—O—C), 833 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 5.59 (s, 2H, OCH_2), 5.92 (s, 2H, pyrrole-C_3, C_4—H), 6.89–6.93 (m, 2H, ph-C_4, C_6—H), 7.18–7.21 (m, 1H, ph-C_5—H), 7.41–7.45 (m, 1H, ph-C_3—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.49, 161.92, 156.34, 131.45, 127.59, 127.30, 121.17, 112.49, 111.73, 109.50, 65.14, 13.41; MS (ESI): *m/z* = found 365.95 [M^++1], 366.95 [M^++2]; calcd. 364.26.

*4.4.8. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(4-tolyloxymethyl)-1,3, 4-thiadiazole (**7h**)*

(Yield 68%). mp 65–67 °C; FTIR (KBr): 3105, 3069 (Ar—H), 1613 (C=N), 1250 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 2.30 (s, 3H CH_3), 5.44 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 6.91, 6.92 (dd, 2H, *J* = 2.04, 1.96, ph-C_2, C_6—H) 7.13 (d, 2H, *J* = 8.36, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.76, 161.76, 155.35, 131.72, 130.23, 130.18, 114.79, 109.15, 65.58, 20.52, 13.44; MS (EI): *m/z* = found 299 [M^+]; calcd. 299.11.

*4.4.9. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(3-tolyloxymethyl)-1,3 ,4-thiadiazole (**7i**)*

(Yield 63%). mp 66–68 °C; FTIR (KBr): 3053, 2917 (Ar—H), 1595 (C=N), 1222 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 2.35 (s, 3H, CH_3), 5.46 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 6.81–6.87 (m, 3H, ph-C_2, C_4, C_6—H), 7.21 (t, 1H, *J* = 7.76, ph-C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.67, 161.77, 157.47, 140.01, 130.18, 129.54, 123.15, 115.82, 111.69, 109.20, 65.34, 21.52, 13.46; MS (EI): *m/z* = found 299 [M^+]; calcd. 299.11.

*4.4.10. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-tolyloxymethyl)-1,3, 4-thiadiazole (**7j**)*

(Yield 64%). mp 90–92 °C; FTIR (KBr): 3057, 2955 (Ar—H), 1600 (C=N), 1221 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.28 (s, 9H, $3CH_3$), 5.47 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 6.93–6.97 (m, 2H, ph-C_4, C_5—H) 7.18–7.25 (m, 2H, ph-C_3, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.03, 161.71, 155.65, 131.19, 130.16, 127.13, 127.03, 122.04, 111.52, 109.23, 65.53, 16.26, 13.46; MS (EI): *m/z* = found 299 [M^+]; calcd. 299.11.

*4.4.11. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((3,5-dimethylphenoxy)methyl)-1,3,4-thiadiazole (**7k**)*

(Yield 59%). mp 69–71 °C; FTIR (KBr): 3062, 2916 (Ar—H), 1583 (C=N), 1245 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.21 (s, 6H, $2CH_3$), 2.34 (s, 6H, $2CH_3$), 5.41 (s, 2H, OCH_2), 5.91 (s, 2H, pyrrole-C_3, C_4—H), 6.60–6.63 (m, 2H, ph-C_2, C_6—H), 6.95–6.97 (m,

1H, ph-C_4—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.63, 161.77, 160.42, 141.26, 127.13, 123.16, 110.48, 109.71, 65.29, 21.46, 13.40; MS (EI): *m/z* = found 313 [M^+]; calcd. 313.12.

*4.4.12. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((2,5-dimethylphenoxy) methyl)-1,3,4-thiadiazole (**7l**)*

(Yield 59%). mp 66–68 °C; FTIR (KBr): 3116, 3013 (Ar—H), 1536 (C═N), 1241 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.19 (s, 6H, $2CH_3$), 2.23 (s, 3H, CH_3), 2.35 (s, 3H, CH_3), 5.48 (s, 2H, OCH_2), 5.90 (s, 2H, pyrrole-C_3, C_4—H), 6.67 (s, 1H, ph-C_4—H), 6.91–6.94 (m, 1H, ph-C_6—H), 7.00–7.04 (m, 1H, ph-C_3—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.36, 161.96, 156.78, 136.63, 130.14, 127.26, 123.59, 120.15, 117.36, 109.49, 65.55, 21.56, 16.09, 13.48; MS (EI): *m/z* = found 313 [M^+]; calcd. 313.12.

*4.4.13. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((2,4-dimethylphenoxy) methyl)-1,3,4-thiadiazole (**7m**)*

(Yield 57%). mp 60–62 °C; FTIR (KBr): 3126, 3012 (Ar—H), 1543 (C═N), 1259 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.20 (s, 6H, $2CH_3$), 2.22 (s, 3H, CH_3), 2.32 (s, 3H, CH_3), 5.43 (s, 2H, OCH_2), 5.92 (s, 2H, pyrrole-C_3, C_4—H), 6.75–7.03 (m, 3H, ph-C_3, C_5, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.15, 161.79, 155.86, 131.99, 130.84, 127.29, 127.09, 126.82, 111.45, 109.48, 65.30, 21.40, 16.02, 13.40; MS (EI): *m/z* = found 313 [M^+]; calcd. 313.12.

*4.4.14. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((4-fluorophenoxy) methyl)-1,3,4-thiadiazole (**7n**)*

(Yield 62%). mp 80–82 °C; FTIR (KBr): 3113, 3075 (Ar—H), 1536 (C═N), 1251 (C—O—C), 758 (C—F); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.26 (s, 6H, $2CH_3$), 5.44 (s, 2H, OCH_2), 5.95 (s, 2H, pyrrole-C_3, C_4—H), 6.93–7.03 (m, 4H, ph-C_2, C_3, C_5, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.23, 161.52, 156.45, 153.49, 127.33, 117.06, 116.99, 108.15, 65.16, 13.27; MS (EI): *m/z* = found 303 [M^+]; calcd. 303.08.

*4.4.15. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((4-iodophenoxy)methyl)-1,3,4-thiadiazole (**7o**)*

(Yield 58%). mp 79–81 °C; FTIR (KBr): 3147, 3044 (Ar—H), 1542 (C═N), 1251 (C—O—C), 698 (C—I); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.22 (s, 6H, $2CH_3$), 5.42 (s, 2H, OCH_2), 5.93 (s, 2H, pyrrole-C_3, C_4—H), 6.68–6.71 (m, 2H, ph-C_2, C_6—H), 7.49–7.53 (m, 2H, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.08, 161.45, 156.83, 138.74, 127.12, 115.61, 108.35, 65.08, 13.23; MS (EI): *m/z* = found 410 [M^+]; calcd. 410.99.

*4.4.16. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((naphthalen-1-yloxy) methyl)-1,3,4-thiadiazole (**7p**)*

(Yield 60%). mp 78–80 °C; FTIR (KBr): 3060 (Ar—H), 1581 (C═N), 1245 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.27 (s, 6H, $2CH_3$), 5.67 (s, 2H, OCH_2), 5.95 (s, 2H, pyrrole-C_3, C_4—H), 6.99 (d, 1H, *J* = 7.68, naphthyl-C_2—H), 7.41 (t, 1H, *J* = 6.76, naphthyl-C_3—H), 7.50–7.54 (m, 3H, naphthyl-C_4, C_6, C_7—H), 7.84 (t, 1H, *J* = 6.76, naphthyl-C_5—H), 8.24 (t, 1H, *J* = 6.64, naphthyl-C_8—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.29, 162.77, 155.32, 135.18, 134.63, 128.00, 127.70, 125.79, 124.48, 123.09, 120.97, 109.26, 105.58, 65.21, 13.50; MS (EI): *m/z* = found 335 [M^+]; calcd. 335.11.

*4.4.17. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-((naphthalen-2-yloxy) methyl)-1,3,4-thiadiazole (**7q**)*

(Yield 58%). mp 84–86 °C; FTIR (KBr): 3055, 2919 (Ar—H), 1601 (C═N), 1223 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.26 (s, 6H, $2CH_3$), 5.60 (s, 2H, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 7.22–7.81 (m, 7H, naphthyl-C_2, C_3, C_4, C_5, C_6, C_7, C_9—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.18, 161.88, 156.20, 130.52, 129.19, 128.84, 127.76, 127.29, 126.93, 126.53, 124.03, 118.91, 109.12, 105.87, 65.18, 13.47; MS (EI): *m/z* = found 335 [M^+]; calcd. 335.11.

*4.4.18. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-phenoxyethyl)-1,3,4-thiadiazole (**9a**)*

(Yield 66%). mp 98–100 °C; FTIR (KBr): 3027, 2924 (Ar—H), 1591 (C═N). 1299 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.22 (s, 6H, $2CH_3$), 3.60 (t, 2H, *J* = 5.88, CH_2), 4.37 (t, 2H, *J* = 5.88, OCH_2), 5.92 (s, 2H, pyrrole-C_3, C_4—H), 6.92, 6.94 (dd, 2H, *J* = 7.76, 7.80, ph-C_3, C_5—H), 6.98 (d, 1H, *J* = 7.36, ph-C_4—H), 7.28, 7.32 (dd, 2H, *J* = 7.44, 7.50, ph-C_2, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.81, 162.15, 158.12, 130.10, 129.59, 121.58, 114.59, 109.00, 65.78, 31.38, 13.36; MS (EI): *m/z* = found 299 [M^+]; calcd. 299.11.

*4.4.19. 2-(2-(4-Chlorophenoxy)ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**9b**)*

(Yield 62%). mp 102–104 °C; FTIR (KBr): 3065, 2928 (Ar—H), 1551 (C═N), 1240 (C—O—C), 829 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.21 (s, 6H, $2CH_3$), 3.61 (t, 2H, *J* = 5.80, CH_2), 4.35 (t, 2H, *J* = 5.80, OCH_2), 5.95 (s, 2H, pyrrole-C_3, C_4—H), 6.90–6.95 (m, 2H, ph-C_2, C_6—H), 7.25–7.28 (m, 2H, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.49, 161.27, 156.18, 129.35, 129.16, 125.83, 116.52, 108.90, 65.17, 31.42, 13.03; MS (EI): *m/z* = found 333 [M^+]; calcd. 333.07.

*4.4.20. 2-(2-(3-Chlorophenoxy)ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**9c**)*

(Yield 58%). mp 95–97 °C; FTIR (KBr): 3109, 2955 (Ar—H), 1562 (C═N), 1235 (C—O—C), 812 (C—Cl); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.22 (s, 6H, $2CH_3$), 3.59 (t, 2H, *J* = 5.70, CH_2), 4.45 (t, 2H, 5.72, OCH_2), 5.94 (s, 2H, pyrrole-C_3, C_4—H), 6.87 (s, 1H, ph-C_6), 7.08 (s, 1H, ph-C_4), 7.27–7.31 (m, 2H, ph-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.13, 161.52, 153.52, 130.13, 127.51, 127.35, 120.19, 119.79, 117.06, 110.49, 109.25, 65.03, 31.40, 13.38; MS (EI): *m/z* = found 333 [M^+]; calcd. 333.07.

*4.4.21. 2-(2-(2-Chlorophenoxy)ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**9d**)*

(Yield 58%). mp 82–84 °C; FTIR (KBr): 3105, 2956 (Ar—H), 1539 (C═N), 1251 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.24 (s, 6H, $2CH_3$), 3.51 (t, 2H, *J* = 5.76, CH_2), 4.49 (t, 2H, *J* = 5.80, OCH_2), 5.92 (s, 2H, pyrrole-C_3, C_4—H), 6.98–7.02 (m, 1H, ph-C_4—H), 7.11–7.14 (m, 1H, ph-C_6—H), 7.27–7.31 (m, 1H, ph-C_4—H), 7.41–7.42 (m, 1H, ph-C_3—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.01, 161.98, 152.85, 130.56, 130.19, 128.03, 123.35, 123.01, 114.57, 109.53, 64.98, 31.59, 13.39; MS (EI): *m/z* = found 333 [M^+]; calcd. 333.07.

*4.4.22. 2-(2-(4-Bromophenoxy)ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**9e**)*

(Yield 64%). mp 103–105 °C; FTIR (KBr): 3105, 3098 (Ar—H), 1565 (C═N), 1259 (C—O—C), 831 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 3.59 (t, 2H, *J* = 5.80, CH_2), 4.52 (t, 2H, *J* = 5.80, OCH_2), 5.93 (s, 2H, pyrrole-C_3, C_4—H), 6.91–6.95 (m, 2H, ph-C_2, C_6—H), 7.41–7.45 (m, 2H, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.62, 162.63, 157.09, 132.52, 127.29, 116.42, 65.36, 31.48, 13.40; MS (EI): *m/z* = found 378 [M^+]; calcd. 378.29.

*4.4.23. 2-(2-(3-Bromophenoxy)ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**9f**)*

(Yield 60%). mp 107–109 °C; FTIR (KBr): 3128, 3050 (Ar—H), 1552 (C═N), 1239 (C—O—C), 820 (C—Br); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.24 (s, 6H, $2CH_3$), 3.53 (t, 2H, *J* = 5.84, CH_2), 4.59 (t, 2H, *J* = 5.86, OCH_2), 5.95 (s, 2H, pyrrole-C_3, C_4—H), 6.85 (s, 1H,

ph-C_6—H), 7.19 (s, 1H, ph-C_4—H), 7.24–7.29 (m, 2H, ph-C_2, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.88, 161.72, 160.09, 130.59, 127.55, 123.16, 120.47, 117.05, 109.49, 111.52, 65.09, 31.44, 13.43; MS (EI): *m/z* = found 378 [M^+]; calcd. 378.29.

*4.4.24. 2-(2-(2-Bromophenoxy)ethyl)-5-(2,5-dimethyl-1H-pyrrol-1-yl)-1,3,4-thiadiazole (**9g**)*

(Yield 65%). mp 88–90 °C; FTIR (KBr): 3026, 2921 (Ar—H), 1564 (C=N), 1299 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.23 (s, 6H, $2CH_3$), 2.43 (s, 3H, CH_3), 3.65 (t, 2H, *J* = 5.78, CH_2), 4.38 (t, 2H, *J* = 5.78, OCH_2), 5.93 (s, 2H, pyrrole-C_3, C_4—H), 6.84 (d, 1H, *J* = 6.23, ph-C_6—H), 6.91 (d, 1H, *J* = 6.48, ph-C_5—H), 7.18–7.25 (m, 2H, ph-C_3, C_4—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.17, 162.15, 155.26, 130.10, 129.34, 113.90, 108.20, 65.48, 30.71, 12.67; MS (EI): *m/z* = found 378 [M^+]; calcd. 378.29.

*4.4.25. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(4-tolyloxy)ethyl)-1,3,4-thiadiazole (**9h**)*

(Yield 67%). mp 65–67 °C; FTIR (KBr): 3019, 2966 (A—H), 1584 (C=N), 1294 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.23 (s, 6H, $2CH_3$), 2.44 (s, 3H, CH_3), 3.60 (t, 2H, *J* = 5.80, CH_2), 4.37 (t, 2H, *J* = 5.82, OCH_2), 5.92 (s, 2H, pyrrole-C_3, C_4—H), 6.72–7.02 (m, 2H, ph-C_2, C_6—H), 7.09–7.12 (m, 2H, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.89, 161.83, 155.27, 131.55, 130.20, 130.09, 114.54, 109.78, 65.52, 31.41, 20.48, 13.40; MS (EI): *m/z* = found 313 [M^+]; calcd. 313.12.

*4.4.26. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(3-tolyloxy)ethyl)-1,3,4-thiadiazole (**9i**)*

(Yield 65%). mp 90–92 °C; FTIR (KBr): 3028, 2922 (Ar—H), 1564 (C=N), 1297 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.19 (s, 6H, $2CH_3$), 2.28 (s, 3H, CH_3), 3.60 (t, 2H, *J* = 5.76, CH_2), 4.29 (t, 2H, *J* = 5.76, OCH_2), 5.93 (s, 2H, pyrrole-C_3, C_4—H), 6.84 (d, 2H, *J* = 8.25, ph-C_2, C_6—H), 7.10 (d, 2H, *J* = 8.54, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.78, 162.03, 157.35, 151.98, 132.10, 130.4, 115.78, 113.14, 108.36, 64.52, 30.05, 21.58, 13.00; MS (EI): *m/z* = found 313 [M^+]; calcd. 313.12.

*4.4.27. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(2-tolyloxy)ethyl)-1,3,4-thiadiazole (**9j**)*

(Yield 65%). mp 85–87 °C; FTIR (KBr): 3027, 2924 (Ar—H), 1581 (C=N), 1297 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.14 (s, 6H, $2CH_3$), 2.26 (s, 3H, CH_3), 3.54 (t, 2H, *J* = 5.80, CH_2), 4.41 (t, 2H, *J* = 5.78, OCH_2), 5.84 (s, 2H, pyrrole-C_3, C_4—H), 6.69, 6.71 (dd, 2H, *J* = 2.20, 1.98, ph-C_2, C_6—H), 7.10, 7.12 (dd, 2H, *J* = 2.22, 1.98, ph-C_4, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 168.59, 162.56, 155.38, 131.40, 130.10, 126.25, 120.29, 112.14, 108.46, 63.52, 29.05, 13.75, 12.81; MS (EI): *m/z* = found 313 [M^+]; calcd. 313.12.

*4.4.28. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(3,5-dimethylphenoxy)ethyl)-1,3,4-thiadiazole (**9k**)*

(Yield 52%). mp 91–93 °C; FTIR (KBr): 3052, 2928 (Ar—H), 1575 (C=N), 1252 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.24 (s, 6H, $2CH_3$), 2.33 (s, 6H, $2CH_3$), 3.52 (t, 2H, *J* = 5.70, CH_2), 4.49 (t, 2H, *J* = 5.76, OCH_2), 5.95 (s, 2H, pyrrole-C_3, C_4—H), 6.60–6.64 (m, 2H, ph-C_2, C_6—H), 6.93–6.95 (m, 1H, ph-C_4—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.55, 161.69, 160.38, 141.21, 127.09, 123.56, 110.29, 109.06, 65.20, 31.03, 21.42, 13.39; MS (EI): *m/z* = found 327 [M^+]; calcd. 327.14.

*4.4.29. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(2,5-dimethylphenoxy)ethyl)-1,3,4-thiadiazole (**9l**)*

(Yield 52%). mp 87–89 °C; FTIR (KBr): 3132, 3056 (Ar—H), 1580 (C=N), 1254 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.20 (s, 6H, $2CH_3$), 2.24 (s, 3H, CH_3), 2.37 (s, 3H, CH_3), 3.50 (t, 2H, *J* = 5.84, CH_2), 4.52 (t, 2H, *J* = 5.84, OCH_2), 5.93 (s, 2H, pyrrole-C_3, C_4—H), 6.69 (s, 1H, ph-C_4—H), 6.93–6.95 (m, 1H, ph-C_6—H), 7.11–7.12 (m, 1H, ph-C_3—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.45, 161.58, 156.39, 136.60, 130.17, 127.35, 123.05, 120.59, 117.07, 109.11, 65.41, 31.50, 21.50, 16.17, 13.40; MS (ESI): *m/z* = found 329.04 [M^++2]; calcd. 327.14.

*4.4.30. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(2,4-dimethylphenoxy)ethyl)-1,3,4-thiadiazole (**9m**)*

(Yield 52%). mp 99–101 °C; FTIR (KBr): 3155, 3049 (Ar—H), 1562 (C=N), 1238 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.22 (s, 6H, $2CH_3$), 2.24 (s, 3H, CH_3), 2.31 (s, 3H, CH_3), 3.48 (t, 2H, *J* = 5.80, CH_2), 4.57 (t, 2H, *J* = 5.82, OCH_2), 5.90 (s, 2H, pyrrole-C_3, C_4—H), 6.75–7.05 (m, 3H, ph-C_3, C_5, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.39, 161.65, 155.80, 131.34, 130.19, 127.55, 127.12, 126.47, 111.37, 109.18, 65.24, 31.38, 21.42, 16.08, 13.42; MS (EI): *m/z* = found 327 [M^+]; calcd. 327.14.

*4.4.31. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(4-fluorophenoxy)ethyl)-1,3,4-thiadiazole (**9n**)*

(Yield 57%). mp 102–104 °C; FTIR (KBr): 3105, 3088 (Ar—H), 1559 (C=N), 1250 (C—O—C), 759 (C—F); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 3.53 (t, 2H, *J* = 5.76, CH_2), 4.59 (t, 2H, *J* = 5.76, OCH_2), 5.93 (s, 2H, pyrrole-C_3, C_4—H), 6.95–7.13 (m, 4H, ph-C_2, C_3, C_5, C_6—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.51, 161.43, 156.38, 153.27, 127.30, 117.05, 117.01, 108.23, 65.24, 31.38, 13.22; MS (EI): *m/z* = found 317 [M^+]; calcd. 317.10.

*4.4.32. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(4-iodophenoxy)ethyl)-1,3,4-thiadiazole (**9o**)*

(Yield 52%). mp 95–97 °C; FTIR (KBr): 3135, 3050 (Ar—H), 1562 (C=N), 1256 (C—O—C), 699 (C—I); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.21 (s, 6H, $2CH_3$), 3.50 (t, 2H, *J* = 5.80, CH_2), 4.57 (t, 2H, *J* = 5.84, OCH_2), 5.91 (s, 2H, pyrrole-C_3, C_4—H), 6.70 (d, 2H, *J* = 8.26, ph-C_2, C_6—H), 7.50 (d, 2H, *J* = 8.40, ph-C_3, C_5—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.03, 161.17, 156.79, 138.62, 127.56, 115.78, 108.19, 65.11, 31.50, 13.18; MS (EI): *m/z* = found 425 [M^+]; calcd. 425.01.

*4.4.33. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(naphthalen-1-yloxy)ethyl)-1,3,4-thiadiazole (**9p**)*

(Yield 55%). mp 76–78 °C; FTIR (KBr): 3142, 3016 (Ar—H), 1578 (C=N), 1255 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.25 (s, 6H, $2CH_3$), 3.38 (t, 2H, *J* = 5.76, CH_2), 4.47 (t, 2H, *J* = 5.78, OCH_2), 5.91 (s, 2H, pyrrole-C_3, C_4—H), 6.99 (d, 1H, *J* = 6.29, naphthyl-C_2—H), 7.40 (t, 1H, *J* = 2.37, naphthyl-C_3—H), 7.49–7.54 (m, 3H, naphthyl-C_4, C_6, C_7—H), 7.85 (t, 1H, *J* = 2.30, naphthyl-C_5—H), 8.20 (t, 1H, *J* = 2.28, naphthyl-C_8—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.56, 162.93, 155.30, 135.37, 134.56, 127.98, 127.39, 125.51, 124.36, 123.12, 120.74, 109.02, 105.87, 65.35, 31.50, 13.49; MS (EI): *m/z* = found 349 [M^+]; calcd. 349.12.

*4.4.34. 2-(2,5-Dimethyl-1H-pyrrol-1-yl)-5-(2-(naphthalen-2-yloxy)ethyl)-1,3,4-thiadiazole (**9q**)*

(Yield 55%). mp 97–99 °C; FTIR (KBr): 3078, 2923 (Ar—H), 1559 (C=N), 1235 (C—O—C); ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm: 2.24 (s, 6H, $2CH_3$), 3.42 (t, 2H, *J* = 5.84, CH_2), 4.51 (t, 2H, *J* = 5.86, OCH_2), 5.95 (s, 2H, pyrrole-C_3, C_4—H), 7.20–7.82 (m, 7H, naphthyl-C_2, C_3, C_4, C_5, C_6, C_7, C_9—H); ^{13}C NMR (100 MHz, $CDCl_3$) δ ppm: 167.35, 161.92, 156.08, 130.58, 129.36, 128.77, 127.84, 127.26, 126.67, 126.52, 124.16, 118.87, 109.38, 105.75, 65.32, 31.45, 13.15; MS (EI): *m/z* = found 349 [M^+]; calcd. 349.12.

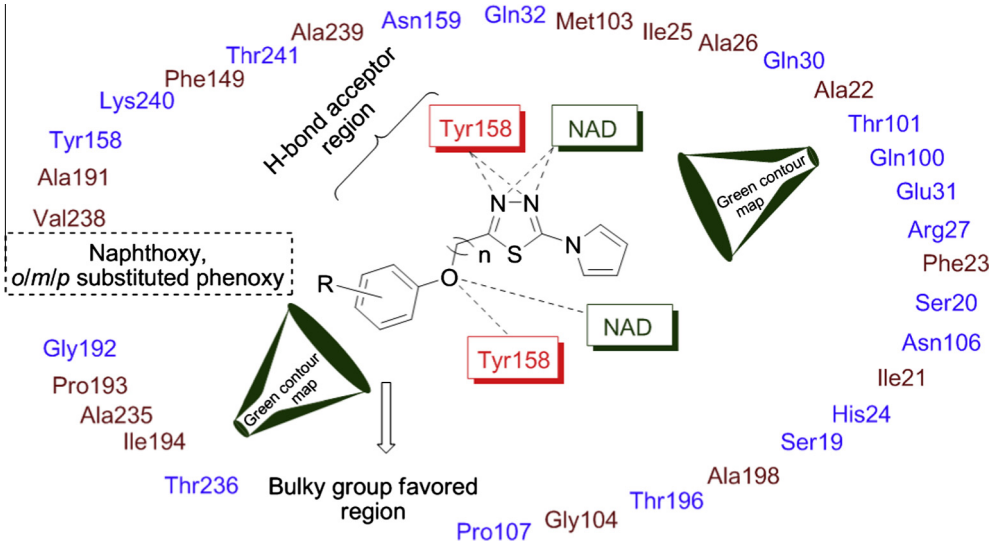


Fig. 12. Docked region and structure–activity relationship for pyrrole aryloxy 1,3,4-thiadiazoles. Brown colored residue represents hydrophobic amino acids and blue colored residues represent hydrophilic amino acids.

5. Biological activity

5.1. Antitubercular activity

MIC values were determined for pyrrolyl aryloxy 1,3,4-thiadiazoles against *M. tuberculosis* strain $H_{37}Rv$ by the Microplate Alamar Blue assay (MABA) [39] using INH as the standard drug. The 96 wells plate received 100 μL of Middlebrook 7H9 broth and serial dilution of compounds were made directly on the plate with drug concentrations of 0.2, 0.4, 0.8, 1.6, 3.125, 6.25, 12.5, 25, 50 and 100 μg/mL. Plates were covered, sealed with parafilm and incubated at 37 °C for 5 days. Then, 25 μL of freshly prepared 1:1 mixture of almar blue reagent and 10% Tween 80 were added to the plate and incubated for 24 h. A blue color in the well was interpreted as no bacterial growth and pink color was scored as the growth. The MIC was defined as the lowest drug concentration, which prevented the color change from blue to pink. Table 1 reveals antitubercular activity (MIC) data.

5.2. MTT-based cytotoxic activity

The cellular conversion of MTT [3-(4,5-dimethylthiazo-2-yl)-2,5-diphenyl-tetrazolium bromide] into a formazan product [40] was used to evaluate cytotoxic activity (IC_{50}) of some synthesized compounds against mammalian Vero cell-lines and A_{549} (lung adenocarcinoma) cell-lines up to concentrations of 62.5 μg/mL using the Promega Cell Titer 96 non-radioactive cell proliferation assay [41] using cisplatin as the positive control. The IC_{50} values presented in Table 2 are the averages ± SEM of three independent experiments.

6. Structure–activity relationships (SARs)

The structure–activity relationship relates to the features of a chemical structure to a property, effect, or biological activity associated with those of chemical structures (Fig. 12). The structure–activity relationship discussed is based on results obtained from *in vitro* anti-TB and in *silico* analyses of synthesized pyrrolyl-1,3,4-thiadiazoles, not compared with compounds listed in Fig. 2. Azoles are important pharmacophore in drug discovery, so a series of compounds were obtained by hybridization between thiadiazole and a pyrrole ring. In doing so, we found that thiadiazole contains two nitrogen atoms (H-bond acceptor atom), which are making key interactions with the amino acid Tyr158 and co-factor NAD+(substrate binding site), but in few cases, instead of thiadiazole nitrogens, we found that oxygen of bridge —OCH_2— making interaction at the substrate binding site i.e. amino acid Tyr158 and co-factor NAD+(**6a**, **6b**, **6i**, **6k**, **8i**, **8k** and **8o**). Since these compounds making H-bond with substrate binding site, fails to retain the desired biological activity, due to the presence of CH_3, Cl and I at the phenyl moiety. In addition, CoMFA green contour maps (steric field) over aryloxy, thiadiazole and pyrrole groups revealed that bulky and H-bond acceptor atoms help to achieve the desired antitubercular activity (**9q** = 3.125 μg/mL, specially naphthoxy bulky group).

Hydrophobic amino acid residues were more observed towards aryloxy moiety than pyrrole. However, along with the naphthoxy bulky group, only fluorine substituted derivatives (**6n**, **7n**, **8n** and **9n** = 6.25 μg/mL) achieved better antitubercular activity compared to other electron withdrawing substituents (Cl, Br and I); such type of increased activity is attributed to presence of fluorine atoms (highly electro negative) in the molecule which increases the lipophilicity and affects the partitioning of a molecule into membranes, but electron donating CH_3 substituted derivatives were found to be moderately active. However, the substitution of pyrrole ring with two methyl groups at 2-CH_3 and 5-CH_3 positions significantly improved the antitubercular activity (compounds **7a–h**, **7k**, **7m**, **7p** and **7q** more active than **6a–h**, **6k**, **6m**, **6p** and **6q**, that of compounds **9a**, **9p** and **9q** are more active than **8a**, **8p** and **8q**, respectively), this is supported by CoMFA analysis a green contours on pyrrole methyl groups. Since we have used single (—OCH_2—) or double (—OCH_2CH_2—) carbon linkages to connect aryloxy and thiadiazole moieties, we found that as the distance between these two moieties increased, antitubercular activity also increased.

7. Conclusions

In this study, drug design approach was employed to generate new structures based on the combination of molecular docking and 3D-QSAR studies performed on the novel derivatives of pyrrole containing aryloxy thiadiazole that are explored as novel anti-TB agents. Among the compounds investigated, **9q** and **9n** displayed significant activities (3.125 and 6.25 μg/mL) against *M. tuberculosis* H37Rv strain. Docking simulation studies have shown that these

compounds are bound mainly with the substrate binding site of InhA and the scoring function for all the compounds is similar or higher than that of the reference inhibitor. Designed structures have shown interactions with the substrate binding site of InhA, confirming their high inhibitory potency, depending on the type of aryl ring modification. The CoMFA models showed high correlative and predictive abilities. A high bootstrapped r^2 value with small standard deviations suggest similar relationships between the compounds. For comparative purpose, two different alignment rules including docked alignment and database alignment were used to obtain 3D-QSAR models. Model generated from the database alignment showed better correlation with the anti-TB activities having improved predictability. Though the knowledge of receptor structure is not a prerequisite for 3D-QSAR analysis, the present study clearly showed the importance of crystal structure (or 3D model) for a receptor to facilitate the structure alignment to provide a model that is statistically a more reliable approach.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bioorg.2015.03.001. These data include MOL files and InChiKeys of the most important compounds described in this article.

References

[1] Y. Zhang, K. Post-Martens, S. Denkin, Drug Discov. Today 11 (2006) 21–27.
[2] S. Hasan, S. Daugelat, P.S. Rao, M. Schreiber, PLoS Comput. Biol. 2 (2006), http://dx.doi.org/10.1371/journal.pcbi.0020061.
[3] S.T. Cole, P.M. Alzari, Biochem. Soc. Trans. 35 (2007) 1321–1324.
[4] WHO Report 2008: The Stop TB Strategy, Case Reports, Treatment Outcomes and Estimates of TB Burden. <http://www.who.int/tb/publications/global_report/2008/annex_3/en/index.html>.
[5] J.W. Campbell, J.E. Cronan Jr., Annu. Rev. Microbiol. 55 (2001) 305–332.
[6] R.J. Heath, C.O. Rock, Curr. Opin. Investig. Drugs 5 (2004) 146–153.
[7] S.W. White, J. Zheng, Y.M. Zhang, C.O. Rock, Annu. Rev. Biochem. 74 (2005) 791–831.
[8] Y.M. Zhang, Y.J. Lu, C.O. Rock, Lipids 39 (2004) 1055–1060.
[9] A. Banerjee, E. Dubnau, A. Quemard, V. Balasubramanian, K.S. Um, T. Wilson, D. Collins, G. de Lisle, W.R. Jacobs Jr., Science 263 (1994) 227–230.
[10] Y. Zhang, B. Heym, B. Allen, D. Young, S. Cole, Nature 358 (1992) 591–593.
[11] P. Pan, P.J. Tonge, Curr. Top. Med. Chem. 12 (2012) 672–693.
[12] H. Lu, P.J. Tonge, Acc. Chem. Res. 41 (2008) 11–20.
[13] R.J. Heath, Y.T. Yu, M.A. Shapiro, E. Olson, C.O. Rock, J. Biol. Chem. 273 (1998) 30316–30320.
[14] M.R. Kuo, H.R. Morbidoni, D. Alland, S.F. Sneddon, B.B. Gourlie, M.M. Staveski, M. Leonard, J.S. Gregory, A.D. Janjigian, C. Yee, J.M. Musser, B. Kreiswirth, H. Iwamoto, R. Perozzo, W.R. Jacobs Jr., J.C. Sacchettini, D.A. Fidock, J. Biol. Chem. 278 (2003) 20851–20859.
[15] X. He, A. Alian, R. Stroud, R. Paul, O. de Montellano, J. Med. Chem. 49 (2006) 6308–6323.
[16] A.R. Bhat, A. Azam, I. Choi, F. Athar, Eur. J. Med. Chem. 46 (2011) 3158–3166.
[17] A. Foroumadi, Z. Kargar, A. Sakhteman, Z. Sharifzadeh, R. Feyzmohammadi, M. Kazemi, A. Shafiee, Bioorg. Med. Chem. Lett. 16 (2006) 1164–1167.
[18] A. Foroumadi, F. Soltani, H. Moallemzadeh-Haghighi, A. Shafiee, Arch. Pharm. (Weinheim) 33 (2005) 112–116.
[19] S.J. Gilani, S.A. Khan, N. Siddiqui, Bioorg. Med. Chem. Lett. 20 (2010) 4762–4765.
[20] B.S. Holla, K.N. Poojary, B.S. Rao, M.K. Shivananda, Eur. J. Med. Chem. 37 (2002) 511–517.
[21] M.G. Mamolo, V. Falagiani, D. Zampieri, L. Vio, E. Banfi, G. Scialino, Il Farmaco 58 (2003) 631–637.
[22] E.E. Oruç, S. Rollas, F. Kandemirli, N. Shvets, A.S. Dimoglo, J. Med. Chem. 30 (2004) 6760–6767.
[23] G. Tu, S. Li, H. Huang, G. Li, F. Xiong, X. Mai, H. Zhu, B. Kuang, W.F. Xu, Bioorg. Med. Chem. 16 (2008) 6663–6668.
[24] S.D. Joshi, U.A. More, K. Pansuriya, T.M. Aminabhavi, A.K. Gadad, J. Saudi Chem. Soc. (2013), http://dx.doi.org/10.1016/j.jscs.2013.09.002.
[25] S.D. Joshi, H.M. Vagdevi, V.P. Vaidya, G.S. Gadaginamath, Eur. J. Med. Chem. 43 (2008) 1989–1996.
[26] S.D. Joshi, U.A. More, T.M. Aminabhavi, A.M. Badiger, Med. Chem. Res. 23 (2014) 107–126.
[27] S.D. Joshi, U.A. More, S.R. Dixit, H.H. Korat, T.M. Aminabhavi, A.M. Badiger, Med. Chem. Res. 23 (2014) 1123–1147.
[28] Tripos International Sybyl-X 2.0 Tripos International, St. Louis, MO, USA (2012).
[29] M. Clark, R.D. Cramer III, N. Van Opdenbosch, J. Comput. Chem. 10 (1989) 982–1012.
[30] M.J.D. Powell, Math. Program. 12 (1977) 241–254.
[31] S. Eswaran, A.V. Adhikari, N.K. Pal, I.H. Chowdhury, Bioorg. Med. Chem. Lett. 20 (2010) 1040–1044.
[32] G. Klebe, U. Abraham, J. Comput.-Aided Mol. Des. 13 (1999) 1–10.
[33] G. Klebe, U. Abraham, T. Mietzner, J. Med. Chem. 37 (1994) 4130–4146.
[34] R.D. Cramer, J.D. Bunce, D.E. Patterson, QSAR 7 (1988) 18–25.
[35] R.D. Cramer III, J.D. Bunce, D.E. Patterson, I.E. Frank, Quant. Struct. Act. Relat. 7 (1988) 18–25.
[36] A.N. Jain, J. Comput.-Aided Mol. Des. 10 (1996) 427–440.
[37] A.N. Jain, J. Med. Chem. 46 (2003) 499–511.
[38] U.A. More, S.D. Joshi, T.M. Aminabhavi, A.K. Gadad, M.N. Nadagouda, V.H. Kulkarni, Eur. J. Med. Chem. 71 (2014) 199–218.
[39] S.G. Franzblau, R.S. Witzig, J.C. McLaughlin, P. Torres, G. Madico, A. Hernandez, M.T. Degnan, M.B. Cook, V.K. Quenzer, R.M. Ferguson, R.H. Gilman, J. Clin. Microbiol. 36 (1998) 362–366.
[40] T. Mosmann, J. Immunol. Methods 65 (1983) 55–63.
[41] L.L. Gundersen, J. Nissen-Meyer, B. Spilsberg, J. Med. Chem. 45 (2002) 1383–1386.