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Evaluation of selective human MAO inhibitory activities of some novel pyrazoline derivatives

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Abstract A series of 1-[2-((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetyl]-3,5-diaryl-4,5-dihydro-1*H*-pyrazole derivatives were prepared by reacting 2-((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetylhydrazine with appropriate chalcones. The chemical structures of all compounds were confirmed by elemental analyses, IR, ^{1}H NMR and ESI–MS. All the compounds were investigated for their ability to selectively inhibit monoamine oxidase (MAO) by in vitro tests. MAO activities of the compounds were compared with moclobemide and selegiline and all the compounds were found to inhibit human MAO-A selectively. The inhibition profile was found to be competitive and reversible for all compounds by in vitro tests. Among the compounds examined, compounds **5ae, 5af** and **5ag** were more selective than moclobemide, with respect to the K_i values experimentally found. In addition, the compound **5bg** showed MAO-A inhibitor activity as well as moclobemide. A series of experimentally tested compounds (**5ae–5ch**) were docked computationally to the active site of the MAO-A and MAO-B isoenzyme. The AUTODOCK 4.01 program was employed to perform automated molecular docking.

Keywords 2-Pyrazoline · 2-Benzoxazolinone · Chalcone · Monoamine oxidase inhibitory activity · Molecular docking

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Introduction

Human monoamine oxidases A and B (MAO-A and B) are the most intensively investigated flavin-dependent amine oxidases and play an important role in the control of intracellular concentration of monoaminergic neurotransmitters. The development of human MAO inhibitors led to important breakthroughs in the therapy of several neuropsychiatric disorders. MAO-A inhibitors are prescribed for the treatment of mental depression and anxiety (Yamada and Yasuhara 2004). MAO-B inhibitors are used with L-DOPA and/or dopamine agonists in the symptomatic treatment of Parkinson's disease (Drukarch and van Muiswinkel 2000; Schapira 2007).

Most current monoamine oxidase inhibitors lead to side effects by a lack of affinity and selectivity toward one of the isoforms. So, it remains fundamental to design new more potent, reversible and selective inhibitors of MAO-A and MAO-B.

Different families of heterocycles containing 2 or 4 nitrogen atoms have been used as scaffolds for synthesizing selective monoamine oxidase inhibitors, but the early period of the MAO-inhibitors started with hydrazine derivatives. Pyrazole, pyrazoline, and pyrazolidine derivatives can be considered as a cyclic hydrazine moiety. This scaffold also displayed promising antidepressant and anticonvulsant properties as demonstrated by different and

established animal models. Diversely substituted pyrazoles, embedded with a variety of functional groups, are important biological agents and a significant amount of research activity has been directed toward this chemical class (Secci et al. 2011). On the basis of this observation, in previous communications, it was reported that N_1-acetyl, N_1-thiocarbamoyl, 1,3,5-triphenyl and 1-quinazolinone-3,5-diphenylpyrazolines exhibited high potency along with good selectivity due to their synthetic accessibility permitted a number of chemical changes (Bilgin et al. 1993; Palaska et al. 2001, 2008; Manna et al. 2002; Gökhan et al. 2003; Chimenti et al. 2004, 2005, 2006a, b, 2007, 2008a; Gökhan-Kelekçi et al. 2007, 2009; Özdemir et al. 2007, 2008). These observations motivated us to link different heterocyclic moieties to synthesize a new series of pyrazoline derivatives by combining the benzoxazolinone moiety at the first position in order to evaluate the effect of this substitution on monoamine oxidase inhibitory effects.

Materials and methods

Chemistry

All chemicals and solvents used in the present study were purchased from Merck A.G., Aldrich Chemical. Melting points of the compounds were determined with a Thomas Hoover Capillary Melting Point Apparatus and were uncorrected. Infrared (IR) spectra were obtained with a Perkin Elmer SpctrumOne, Nicolet 520 FT-IR spectrometer and the results were expressed in wave number (cm^{-1}). ^1H NMR spectrums were recorded on a Bruker 400 MHz UltraShield spectrometer using dimethylsulfoxide (DMSO-d_6) with chemical shifts reported as δ (ppm) from TMS. Mass spectrums were undertaken using Waters 2695 Alliance Micromass ZQ LC/MS spectrometer in methanol according to the EI technique. Elemental analyses (C, H, N) were performed on an LECO CHNS 932 analyzer at the laboratory of Ankara University. The purity of the compounds was assessed by TLC on silicagel $HF_{254+366}$ (E.Merck, Darmstadt, Germany).

General procedure for the preparation of 1,3-diaryl-2-propen-1-ones (4e–h) (chalcones)

Chalcone derivatives were synthesized by condensing acetophenone (10 mmol) and appropriate benzaldehydes (10 mmol) in the presence of sodium hydroxide (12.5 mmol) in water and ethanol (5/3 mL) at 0 °C for 1 h. The solid mass separated out was filtered, dried and crystallized from methanol (Dawey and Tivey 1958). 4e: m.p. 58–58.5 °C (Irie and Watanabe 1980; Lipson et al. 2005), 4f: m.p. 60–62 °C (Irie and Watanabe 1980), 4g: m.p. 75–76 °C (Ueno et al. 1983; Dong et al. 2008), 4h: m.p.: 135–137 °C (Sarabhai and Mathur 1963; Kubota et al. 2006).

General procedure for the preparation of 1-[2-((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetyl]-3,5-diaryl-4,5-dihydro-1H-pyrazoles (5)

2-((5-Methyl/chloro)-2-benzoxazolinone-3-yl)acetylhydrazine (1 mmol) was dissolved in 2 mL of DMF and 20 mL of *n*-propanol. 1,3-Diaryl-2-propen-1-one (1 mmol) and eight drops of hydrochloric acid was added to this solution and was refluxed for approximately 120 h (Gökhan-Kelekçi et al. 2009). The reaction mixture was then cooled and the solid precipitated was recrystallized. If solid was not precipitated, the solution was purified by chromatography on a silica gel column.

Biochemistry

Chemicals

hMAO-A (recombinant, expressed in baculovirus infected BTI insect cells), hMAO-B (recombinant, expressed in baculovirus infected BTI insect cells), *R*-(–)-deprenyl hydrochloride, resorufin, dimethyl sulfoxide, and other chemicals were purchased from Sigma-Aldrich TM (Germany). Moclobemide was donated (Roche Pharmaceuticals, Germany). The Amplex®-Red MAO Assay Kit (Molecular Probes, USA) contained benzylamine, *p*-tyramine, Clorgyline (MAO-A inhibitor), Pargyline (MAO-B inhibitor), and horseradish peroxidase.

Determination of inhibitory activities of the compounds on human MAO-A and -B

The activity of hMAO-A and hMAO-B (using *p*-tyramine as common substrate for both isoforms) was found to be 185.60 ± 9.50 pmol/mg/min ($n = 3$). The interactions of the synthesized compounds with hMAO isoforms were determined by a fluorimetric method described and modified previously (Anderson et al. 1993; Yáñez et al. 2006; Chimenti et al. 2008b). The production of H_2O_2 catalyzed by MAO isoforms was detected using 10-acetyl-3,7-dihydroxyphenoxazine (Amplex®-Red reagent), a non-fluorescent, highly sensitive, and stable probe that reacts with H_2O_2 in the presence of horseradish peroxidase to produce the fluorescent product resorufin. The reaction was started by adding (final concentrations) 200 μM Amplex Red reagent, 1 U/mL horseradish peroxidase, and *p*-tyramine (concentration range 0.1–1 mM).

Control experiments were carried out simultaneously by replacing the test drugs (novel pyrazoline derivatives and

reference inhibitors) with appropriate dilutions of the vehicles. In addition, the possible capacity of novel compounds to modify the fluorescence generated in the reaction mixture due to non-enzymatic inhibition (e.g., for directly reacting with Amplex Red reagent) was determined by adding these compounds to solutions containing only the Amplex Red reagent in a sodium phosphate buffer.

Kinetic experiments

Newly synthesized compounds were dissolved in dimethyl sulfoxide, with a maximum concentration of 1 %, and used in the final concentration range of 0.1–1,000 nM. Kinetic data for interaction of the enzyme with the compounds were determined using the Microsoft Excel package program. The slopes of the Lineweaver–Burk plots were plotted versus the inhibitor concentration and the K_i values were determined from the x axis intercept as $-K_i$. Each K_i value is the representative of single determination where the correlation coefficient (R^2) of the replot of the slopes versus the inhibitor concentrations was at least 0.98. SI (K_i(MAO-A)/K_i(MAO-B)) was also calculated. The protein was determined according to the Bradford method (Bradford 1976), in which bovine serum albumin was used as a standard.

Reversibility experiments

Reversibility of the MAO inhibition with novel derivatives was evaluated by a centrifugation-ultrafiltration method (Chimenti et al. 2010). In brief, adequate amounts of the recombinant hMAO-A or B were incubated together with a single concentration of the newly synthesized compounds or the reference inhibitors in a sodium phosphate buffer (0.05 M, pH 7.4) for 15 min at 37 °C. After this incubation period, an aliquot was stored at 4 °C and used for the measurement of MAO-A and -B activity. The remaining incubated sample was placed in an Ultrafree-0.5 centrifugal tube (Millipore, USA) with a 30 kDa Biomax membrane in the middle of the tube and centrifuged at 9,000×g for 20 min at 4 °C. The enzyme retained in the 30 kDa membrane was resuspended in a sodium phosphate buffer at 4 °C and centrifuged again two successive times. After the third centrifugation, the enzyme retained in the membrane was resuspended in sodium phosphate buffer (300 mL) and an aliquot of this suspension was used for MAO-A and -B activity determination.

Control experiments were performed simultaneously (to define 100 % MAO activity) by replacing the test drugs with appropriate dilutions of the vehicles. The corresponding values of percent (%) MAO isoform inhibition were separately calculated for samples with and without repeated washing.

Molecular docking studies

The crystal structures of MAO-A and MAO-B were extracted from the protein data bank (PDB) [http://www.rcsb.org). (for MAO-A pdb code: 2Z5X; human monoamine oxidase in complex with harmine, resolution 2.2 Å (Son et al. 2008) and for MAO-B pdb code: 2V5Z; human MAO-B in complex with inhibitor safinamide, resolution 1.6 Å (Binda et al. 2007)]. Each structure was cleaned of all water molecules and inhibitors as well as all non-interacting ions before being used in the docking studies. The initial oxidized form of the FAD was used in all docking studies. For MAO-A and MAO-B, one of the two subunits was taken as the target structure. Using a fast Dreiding-like force field, each protein's geometry was first optimized and then submitted to the "Clean Geometry" toolkit of Discovery Studio (Accelrys, Inc.) for a more complete check. Missing hydrogen atoms were added based on the protonation state of the titratable residues at a pH of 7.4. Ionic strength was set to 0.145 and the dielectric constant was set to 10. The ADT (V. 1.5.4) (ADT) (Morris et al. 2009) graphical user interface program was employed to setup the enzymes for molecular docking.

Ligand setups

The 3D structures of ligand molecules were built, optimized at (PM3) level and saved in pdb format. The ADT package was also employed here to generate the docking input files of ligands. AutoDock 4.2 was used for all dockings; the detailed docking procedure has been given elsewhere (Yelekçi et al. 2007).

Results and discussion

Chemistry

A novel series of 1-[2-((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetyl]-3,5-diaryl-4,5-dihydro-1*H*-pyrazole derivatives were synthesized and investigated for the ability to inhibit the activity of the A and B isoforms of human MAO. The synthesis pathway of the compounds was given in Scheme 1. 5-Methyl-2-benzoxazolinone **1b**, was synthesized as per the methods in the literature using 4-methyl-2-aminophenol and urea (Close et al. 1949). Treatment of (5-methyl/chloro)-2-benzoxazolinone with ethyl chloroacetate in K_2CO_3/acetone gave the N-alkylated product ethyl ((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetate **2a–2c** (Milcent et al. 1996; Potts et al. 1980; Ünlü et al. 1992). The acid hydrazides **3a–3c** were prepared by the reaction of ethyl ((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetate and hydrazine hydrate in ethanol

(Çakır et al. 2001; Gökçe et al. 2001; Önkol et al. 2008; Salgın-Gökşen et al. 2007). On the other hand, α,β-unsaturated carbonyl compounds (chalcones) **4e–4h** were prepared by reacting appropriate aldehydes and acetophenone derivatives under basic condition according to the Claisen–Schmidt condensation (Dawey and Tivey 1958). The reaction of hydrazides **3a–3c** with chalcones **4e–4h** in *n*-propanol under acidic condition gave compounds 1-[2-((5-methyl/chloro)-2-benzoxazolinone-3-yl)acetyl]-3,5-diaryl-4,5-dihydro-1*H*-pyrazoles **5ae–5ch**.

The purity of the synthesized compounds was checked by elemental analyses and the results were within ±0.4 % of the theoretical values. The structures of the synthesized compounds were determined on the basis of spectral data analysis; such as IR, ^{1}H NMR and ESI–MS (Table 1).

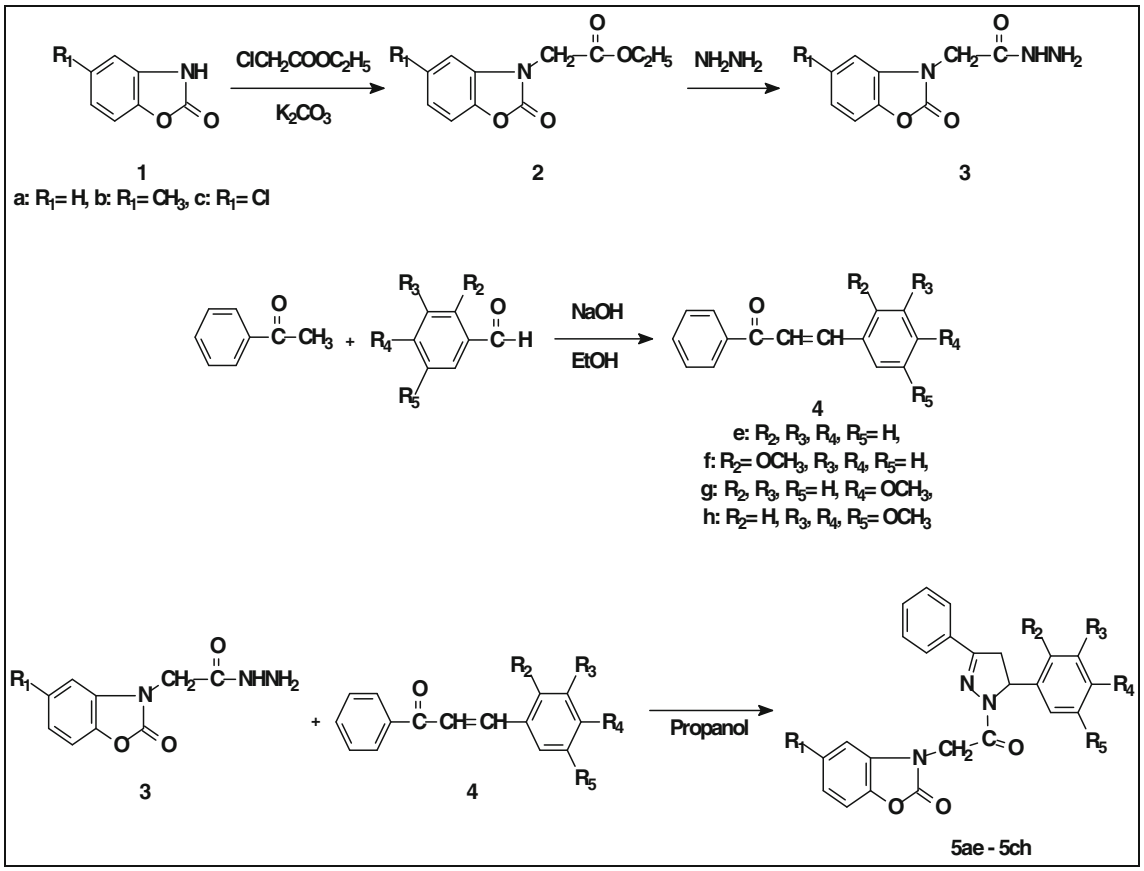
Two C=O stretching bands viewed at 1,789–1,753 cm^{-1} and 1,679–1,664 cm^{-1} in the IR spectra of compounds **5ae–5ch**. The IR spectra of all the compounds showed C=C and C=N stretching bands at 1,609–1,440 cm^{-1}.

In the ^{1}H NMR spectrum of the compounds **5ae–5ch**, it was observed three distinct doublet of doublets of the ABX system at δ 5.71–3.09 ppm due to pyrazoline ring (Shekarchi et al. 2008). The CH (H_X) proton appeared between δ 5.71 and 5.52 ppm due to vicinal coupling with the two magnetically non-equivalent protons of the methylene group at position 4 of the pyrazoline ring. The signals of H_A and H_B of pyrazoline ring were observed as doublet of doublets in the regions 3.95–3.88 ppm (H_B) and 3.26–3.09 ppm (H_A). The CH_2 protons between the benzoxazolinone and pyrazoline ring resonated as a pair of doublet of doublets between δ 5.37–5.15 and 5.12–5.03 ppm. The signals for methoxy and methyl appeared at δ 3.80–3.59 ppm and δ 2.31–2.27 ppm, respectively (Holla et al. 2000; Chen et al. 2011).

The characteristic peaks were observed in the mass spectra of the compounds. The ions produced under ESI showed a characteristic $[M + Na]^+$ ion peak as the base signal for all compounds. Characteristic $[M + Na + 2]^+$ isotope peaks were observed in the mass spectra of the compounds having chloride ion (compounds **5ce, 5cf, 5cg, 5ch**).

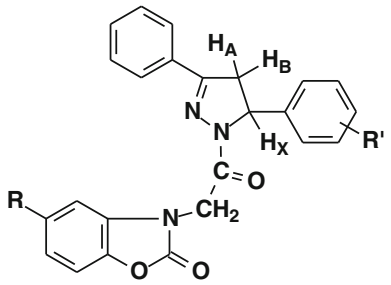
Biochemistry

MAO-A and MAO-B inhibitory activities of newly synthesized pyrazoline derivatives were determined using hMAO isoforms by a fluorimetric method. All the tested compounds were found to inhibit MAO-A selectively and



Scheme 1 Synthesis of the compounds

Table 1 Some characteristic and spectroscopic data of the synthesized compounds (**5ae–5ch**)



Compounds	Melting point (°C)	IR ν (cm^{-1})	^{1}H NMR (DMSO-d_6) δ ppm (*J in Hz)*	Mass *m/z*
5ae	192–194	3,057, 2,934 (C–H), 1,763, 1,673 (C=O), 1,486, 1,440 (C=C, C=N)	3.23 (dd, 1H, H_A, J_{AB}:18.4 Hz, J_{AX}:4.5 Hz), 3.95 (dd, 1H, H_B, J_{AB}:18.6 Hz, J_{BX}:11.8 Hz), 5.11 (d, 1H, N–CH_1H$_2$–CO, J:17.8 Hz), 5.25 (d, 1H, N–CH$_1$$H_2$-CO, J:17.7 Hz), 5.61 (dd, 1H, H_X, J_{BX}:11.6 Hz, J_{AX}:4.6 Hz), 7.13 (t, 1H, 2-benzox.–H_5), 7.18 (t, 1H, 2-benzox.–H_6), 7.24–7.28 (m, 4H, 2-benzox.–H_4 ve phenyl-3H), 7.32–7.37 (m, 3H, 2-benzox.–H_7 ve phenyl-2H), 7.51–7.52 (m, 3H, phenyl-3H), 7.87–7.88 (m, 2H, phenyl-2H)	436, 421, 420 (100 %), 398
5af	198–200	2,934, 2,838 (C–H), 1,777, 1,673 (C=O), 1,599, 1,489, 1,440 (C=C, C=N)	3.09 (dd, 1H, H_A, J_{AB}:18.0 Hz, J_{AX}:4.6 Hz), 3.80 (s, 3H, –OCH_3), 3.90 (dd, 1H, H_B, J_{AB}:18.0 Hz, J_{BX}:11.8 Hz), 5.10 (d, 1H, N–CH_1H$_2$–CO, J:17.7 Hz), 5.28 (d, 1H, N–CH$_1$$H_2$-CO, J:17.7 Hz), 5.71 (dd, 1H, H_X, J_{BX}:11.8 Hz, J_{AX}:4.6 Hz), 6.89 (t, 1H, phenyl-H), 7.04 (t, 2H, phenyl-2H), 7.13 (t, 1H, 2-benzox.–H_5), 7.19 (t, 1H, 2-benzox.–H_6), 7.25 (d, 1H, 2-benzox.–H_4, J:7.7 Hz), 7.28 (d, 1H, phenyl-H, J:7.6 Hz), 7.36 (d, 1H, 2-benzox.–H_7, J:7.8 Hz), 7.47–7.51 (m, 3H, phenyl-3H), 7.84–7.86 (m, 2H, phenyl-2H)	466, 451, 450 (100 %), 428
5ag	214–215	2,957, 2,941, 2,828 (C–H), 1,779, 1,669 (C=O), 1,603, 1,516, 1,490, 1,447 (C=C, C=N)	3.22 (dd, 1H, H_A, J_{AB}:18.4 Hz, J_{AX}:4.8 Hz), 3.72 (s, 3H, –OCH_3), 3.90 (dd, 1H, H_B, J_{AB}:18.4 Hz, J_{BX}:11.6 Hz), 5.08 (d, 1H, N–CH_1H$_2$–CO, J:18.0 Hz), 5.21 (d, 1H, N–CH$_1$$H_2$–CO, J:17.6 Hz), 5.54 (dd, 1H, H_X, J_{BX}:11.6 Hz, J_{AX}:4.8 Hz), 6.88 (d, 2H, 4-methoxyphenyl-2H, J:8.8 Hz), 7.10–7.19 (m, 4H, 2-benzox.–H_5, 2-benzox.–H_6 ve 4-methoxyphenyl-2H), 7.25 (d, 1H, 2-benzox.–H_4, J:7.6 Hz), 7.35 (d, 1H, 2-benzox.–H_7, J:7.2 Hz), 7.50–7.52 (m, 3H, phenyl-3H), 7.86–7.88 (m, 2H, phenyl-2H)	466, 451, 450 (100 %), 428
5ah	236.5-237.5	2,997, 2,941, 2,825 (C–H), 1,766, 1,679 (C=O), 1,590, 1,457, 1,443 (C=C, C=N)	3.26 (dd, 1H, H_A, J_{AB}:18.2 Hz, J_{AX}:5.2 Hz), 3.62 (s, 3H, –OCH_3), 3.75 (s, 6H, –OCH_3), 3.91 (dd, 1H, H_B, J_{AB}:18.3 Hz, J_{BX}:11.9 Hz), 5.12 (d, 1H, N–CH_1H$_2$-CO, J:17.6 Hz), 5.34 (d, 1H, N–CH$_1$$H_2$-CO, J:17.7 Hz), 5.55 (dd, 1H, H_X, J_{BX}:11.8 Hz, J_{AX}:5.1 Hz), 6.52 (s, 2H, 3,4,5-trimethoxyphenyl-2H), 7.14 (t, 1H, 2-benzox.–H_5), 7.19 (t, 1H, 2-benzox.-H_6), 7.32 (d, 1H, 2-benzox.–H_4, J:7.6 Hz), 7.37 (d, 1H, 2-benzox.–H_7, J:7.8 Hz), 7.51–7.52 (m, 3H, phenyl-3H), 7.85–7.87 (m, 2H, phenyl-2H)	526, 511, 510 (100 %), 488, 320, 176

Table 1 continued

Compounds	Melting point (°C)	IR ν (cm^{-1})	^{1}H NMR (DMSO-d_6) δ ppm (*J in Hz)*	Mass *m/z*
5be	201.5–202.5	3,063, 2,925 (C–H), 1,755, 1,677 (C=O), 1,499, 1,441 (C=C, C=N)	2.30 (s, 3H, –CH_3), 3.23 (dd, 1H, H_A, J_{AB}:18.2 Hz, J_{AX}:4.8 Hz), 3.94 (dd, 1H, H_B, J_{AB}:18.0 Hz, J_{BX}:11.6 Hz), 5.05 (d, 1H, N–CH_1H$_2$–CO, J:18.0 Hz), 5.19 (d, 1H, N–CH$_1$$H_2$–CO, J:17.6 Hz), 5.60 (dd, 1H, H_X, J_{BX}:11.8 Hz, J_{AX}:4.8 Hz), 6.92 (d, 1H, 2-benzox.–H_6, J_{67}:8.4 Hz), 7.06 (s, 1H, 2-benzox.–H_4), 7.21 (d, 1H, 2-benzox.–H_7, J_{67}:8.0 Hz), 7.23–7.28 (m, 3H, phenyl-3H), 7.32–7.35 (m, 2H, phenyl-2H), 7.49–7.52 (m, 3H, phenyl-3H), 7.86–7.88 (m, 2H, phenyl-2H)	450, 435, 434 (100 %), 412
5bf	206–207	3,472 (O–H), 3,055, 2,913, 2,834 (C–H), 1,753, 1,675 (C=O), 1,597, 1,499, 1,443 (C=C, C=N)	2.31 (s, 3H, –CH_3), 3.09 (dd, 1H, H_A, J_{AB}:18.0 Hz, J_{AX}:4.7 Hz), 3.79 (s, 3H, –OCH_3), 3.89 (dd, 1H, H_B, J_{AB}:18.0 Hz, J_{BX}:11.8 Hz), 5.04 (d, 1H, N–CH_1H$_2$–CO, J:17.6 Hz), 5.27 (d, 1H, N–CH$_1$$H_2$–CO, J:17.6 Hz), 5.71 (dd, 1H, H_X, J_{BX}:11.7 Hz, J_{AX}:4.6 Hz), 6.89 (t, 1H, phenyl-H), 6.93 (d, 1H, 2-benzox.–H_6, J_{67}:8.16 Hz), 7.03–7.06 (m, 2H, phenyl-2H), 7.08 (s, 1H, 2-benzox.–H_4), 7.22 (d, 1H, 2-benzox.–H_7, J_{67}:8.12 Hz), 7.26 (t, 1H, phenyl-H), 7.47–7.51 (m, 3H, phenyl-3H), 7.84–7.86 (m, 2H, phenyl-2H)	481, 480, 465, 464 (100 %), 443, 442
5bg	172–173	3,074, 2,952, 2,925, 2,830 (C–H), 1,776, 1,674 (C=O), 1,515, 1,495, 1,441 (C=C, C=N)	2.30 (s, 3H, –CH_3), 3.22 (dd, 1H, H_A, J_{AB}:18.4 Hz, J_{AX}:4.8 Hz), 3.72 (s, 3H, –OCH_3), 3.90 (dd, 1H, H_B, J_{AB}:18.0 Hz, J_{BX}:11.6 Hz), 5.03 (d, 1H, N–CH_1H$_2$–CO, J:18.0 Hz), 5.15 (d, 1H, N–CH$_1$$H_2$–CO, J:17.6 Hz), 5.54 (dd, 1H, H_X, J_{BX}:11.4 Hz, J_{AX}:4.8 Hz), 6.88 (d, 2H, 4-methoxyphenyl-2H, J:8.8 Hz), 6.92 (d, 1H, 2-benzox.-H_6, J_{67}:8.4 Hz), 7.04 (s, 1H, 2-benzox.–H_4), 7.16 (d, 2H, 4-methoxyphenyl -2H, J:8.8 Hz), 7.21 (d, 1H, 2-benzox.–H_7, J_{67}:8.4 Hz), 7.50–7.52 (m, 3H, phenyl-3H), 7.86–7.88 (m, 2H, phenyl-2H)	480, 465, 464 (100 %), 442
5bh	237–238	2,929, 2,834 (C–H), 1,766, 1,673 (C=O), 1,609, 1,503, 1,436 (C=C, C=N)	2.27 (s, 3H, –CH_3), 3.23 (dd, 1H, H_A, J_{AB}:18.6 Hz, J_{AX}:5.2 Hz), 3.59 (s, 3H, –OCH_3), 3.72 (s, 6H, –OCH_3), 3.88 (dd, 1H, H_B, J_{AB}:18.2 Hz, J_{BX}:11.6 Hz), 5.03 (d, 1H, N–CH_1H$_2$–CO, J:17.6 Hz), 5.27 (d, 1H, N–CH$_1$$H_2$–CO, J:18.0 Hz), 5.52 (dd, 1H, H_X, J_{BX}:11.6 Hz, J_{AX}:5.2 Hz), 6.49 (s, 2H, 3,4,5- trimethoxyphenyl-2H), 6.91 (d, 1H, 2-benzox.–H_6, J_{67}:8.0 Hz), 7.09 (s, 1H, 2-benzox.–H_4), 7.20 (d, 1H, 2-benzox.–H_7, J_{67}:8.4 Hz), 7.47–7.49 (m, 3H, phenyl-3H), 7.82–7.85 (m, 2H, phenyl-2H)	540, 525, 524 (100 %), 502
5ce	146–148	3,055, 2,929 (C–H), 1,755, 1,668 (C=O), 1,487, 1,440 (C=C, C=N)	3.24 (dd, 1H, H_A, J_{AB}:18.4 Hz, J_{AX}:4.8 Hz), 3.95 (dd, 1H, H_B, J_{AB}:18.2 Hz, J_{BX}:11.6 Hz), 5.12 (d, 1H, N–CH_1H$_2$–CO, J:18.0 Hz), 5.27 (d, 1H, N–CH$_1$$H_2$–CO, J:17.6 Hz), 5.61 (dd, 1H, H_X, J_{BX}:11.8 Hz, J_{AX}:4.8 Hz), 7.18 (dd, 1H, chlorzox.-H_6, J_{67}:8.6 Hz, J_{46}:2.0 Hz), 7.25–7.29 (m, 3H, phenyl-3H), 7.33–7.36 (m, 2H, phenyl-2H), 7.40 (d, 1H, chlorzox.–H_7, J_{67}:8.4 Hz), 7.51–7.53 (m, 4H, chlorzox.–H_4 ve phenyl-3H), 7.86–7.89 (m, 2H, phenyl-2H)	470, 457, 456, 455, 454 (100 %), 434, 432

Table 1 continued

Compounds	Melting point (°C)	IR ν (cm^{-1})	^{1}H NMR (DMSO-d_6) δ ppm (*J in Hz*)	Mass *m/z*
5cf	176–177	3,059, 2,948, 2,842 (C–H), 1,767, 1,672 (C=O), 1,487, 1,455, 1,440 (C=C, C=N)	3.09 (dd, 1H, H_A, J_{AB}:18.1 Hz, J_{AX}:4.6 Hz), 3.80 (s, 3H, –OCH_3), 3.89 (dd, 1H, H_B, J_{AB}:17.9 Hz, J_{BX}:11.7 Hz), 5.10 (d, 1H, N–CH_1H$_2$–CO, J:17.7 Hz), 5.29 (d, 1H, N–CH$_1$$H_2$–CO, J:17.7 Hz), 5.71 (dd, 1H, H_X, J_{BX}:11.7 Hz, J_{AX}:4.6 Hz), 6.89 (t, 1H, phenyl-H), 7.05 (t, 2H, phenyl-2H), 7.18 (dd, 1H, chlorzox.–H_6, J_{67}:8.5 Hz, J_{46}:2.1 Hz), 7.26 (t, 1H, phenyl-1H), 7.39 (d, 1H, chlorzox.–H_7, J_{67}:8.5 Hz), 7.49–7.50 (m, 4H, chlorzox.–H_4 ve phenyl-3H), 7.84–7.86 (m, 2H, phenyl-2H)	487, 486, 485, 484 (100 %), 462, 354
5cg[a]	–	–	–	–
5ch	262–263	3,063, 2,944, 2,822 (C–H), 1,771, 1,664 (C=O), 1,593, 1,491, 1,440 (C=C, C=N)	3.26 (dd, 1H, H_A, J_{AB}:18.2 Hz, J_{AX}:5.2 Hz), 3.61 (s, 3H, –OCH_3), 3.75 (s, 6H, –OCH_3), 3.91 (dd, 1H, H_B, J_{AB}:18.2 Hz, J_{BX}:11.6 Hz), 5.11 (d, 1H, N–CH_1H$_2$–CO, J:17.6 Hz), 5.37 (d, 1H, N–CH$_1$$H_2$–CO, J:18.0 Hz), 5.55 (dd, 1H, H_X, J_{BX}:11.8 Hz, J_{AX}:5.2 Hz), 6.53 (s, 2H, 3,4,5-trimethoxyphenyl-2H), 7.19 (dd, 1H, chlorzox.–H_6, J_{67}:8.4 Hz, J_{46}:2.0 Hz), 7.41 (d, 1H, chlorzox.–H_7, J_{67}:8.0 Hz), 7.50–7.52 (m, 3H, chlorzox.–H_4 ve phenyl-2H), 7.55 (d, 1H, phenyl-H, J:2.4 Hz), 7.85–7.87 (m, 2H, phenyl-2H)	562, 560, 547, 546, 545, 544 (100 %), 522, 182

[a] Şahin et al. 2011

competitively (Table 2). These novel compounds were reversible inhibitors of hMAO-A since the enzyme activity was restored after centrifugation-ultrafiltration steps (Table 2).

Except compounds with **h** substitution (trimethoxy) in phenyl ring, all the compounds were found to be a potent MAO-A inhibitors with K_i values in nM range and with $SI_{MAO\text{-}A}$ in the magnitude of 10^3–10^4. Compounds **5ae**, which is unsubstituted, and **5af**, which has a methoxy substitution on R_2 position were appeared as the most potent MAO-A inhibitors within this series with K_i values of 0.003 ± 10^{-5} and 0.010 ± 10^{-3} μM, respectively. Docking results given in Table 2 are in agreement with the biochemical evaluations. The high inhibitory potency and selectivity of **5ae** through hMAO-A were discussed in detail in the next part according to the computational data obtained.

Compound **5bg,** which carries a methyl group at R_1 position of benzoxazolinone ring and a methoxy group at para position of phenyl ring inhibited hMAO-A with K_i value of 0.090 ± 10^{-3} (Table 2). Experimental selectivity index for this compound was found as 0.004, which is satisfactory and comparable with $SI_{MAO\text{-}A}$ of known MAO-A inhibitor; moclobemide (0.004).

It was suggested that in case that the benzoxazolinone ring is unsubstituted or substituted with methyl group (**a** or **b** substitution), MAO-A inhibitory activity is better compared to chloride substitution (**c** substitution), except in the case of compound **5be**. Furthermore, trimethoxy substitution in phenyl ring (**h** substitution) has been found unfavorable in terms of MAO-A inhibitory activity. Among compounds that benzoxazolinone ring is unsubstituted or substituted with chloride group (**a** or **c** substitution), compounds carrying **e** substitution was found the most potent MAO-A inhibitor among the substitutions of e, f and g.

In the present study, we have successfully identified new compounds which are reversible and selective inhibitor of hMAO-A. It was suggested that unsubstituted benzoxazolinone ring favors MAO-A inhibitory activity whereas methoxy substitutions of phenyl ring at meta and para positions reveals a significant decrease in MAO-A inhibition activity. Results of this study will provide a useful information for designing a new series of potent, selective and reversible MAO-A inhibitors in future.

Molecular docking studies

To figure out the detailed interactions of the docked poses of the inhibitors, compound **5ae** was selected for visualization. The binding modes for inhibitor **5ea** (Fig. 1) in the MAO-A and MAO-B active site cavities are shown in below images. A careful analysis of the binding mode of the compound **5ea** in the MAO-A cavity revealed that the benzoxazolinone ring of this compound inserted into the hydrophobic pocket lined with the TYR444, TYR407 and FAD cofactor. Two phenyl rings of inhibitor **5ae** make two

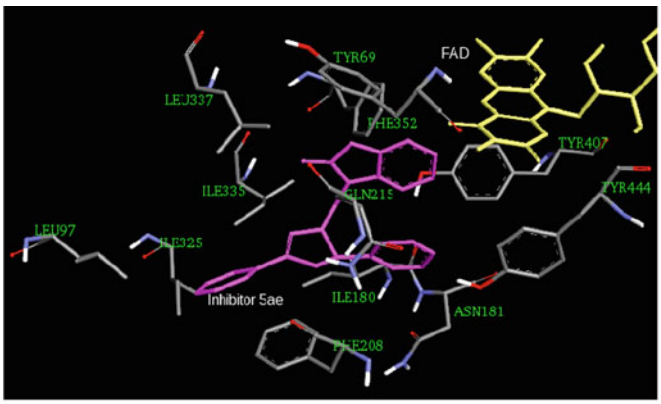
Table 2 Calculated and experimental K_i values corresponding to the inhibition of MAO isoforms by the newly synthesized *2-pyrazoline* derivatives

Compounds	Calculated K_i value for MAO-A (μM)	Calculated K_i value for MAO-B (μM)	Calculated SI*	Experimental K_i value for MAO-A (μM)**	Experimental K_i value for MAO-B (μM)**	Experimental SI*	Inhibition type, selectivity, reversibility
5ae (R)	0.001	1.20	0.000833	0.003 ± 0.00001	1.80 ± 0.13	0.002	MAO-A, competitive, reversible
5ae (S)	0.001	1.97	0.000508				
5af (R)	0.009	3.69	0.002	0.010 ± 0.001	3.80 ± 0.17	0.003	MAO-A, competitive, reversible
5af (S)	0.007	5.82	0.001				
5ag (R)	0.00093	11.29	0.0000823	0.050 ± 0.002	32.00 ± 1.60	0.002	MAO-A, competitive, reversible
5ag (S)	0.031	55.15	0.000562				
5ah (R)	24.32	63.85	0.381	45.260 ± 1.250	590.00 ± 15.21	0.076	MAO-A, competitive, reversible
5ah (S)	38.81	566.75	0.068				
5be (R)	0.003	3.74	0.000802	0.100 ± 0.009	3.00 ± 0.01	0.03	MAO-A, competitive, reversible
5be (S)	0.133	3.29	0.040				
5bf (R)	0.070	9.74	0.007	0.080 ± 0.002	1.00 ± 0.09	0.080	MAO-A, competitive, reversible
5bf (S)	0.070	0.70	0.1				
5bg (R)	0.002	12.71	0.000157	0.090 ± 0.001	23.10 ± 1.60	0.004	MAO-A, competitive, reversible
5bg (S)	0.044	19.94	0.002				
5bh (R)	31.61	254.54	0.124	15.20 ± 1.05	153.00 ± 8.06	0.099	MAO-A, competitive, reversible
5bh (S)	461.06	58.55	7.875				
5ce (R)	0.014	2.37	0.006	0.050 ± 0.002	1.90 ± 0.009	0.027	MAO-A, competitive, reversible
5ce (S)	0.063	1.15	0.055				
5cf (R)	0.054	29.39	0.002	0.095 ± 0.002	7.00 ± 0.23	0.014	MAO-A, competitive, reversible
5cf (S)	0.034	0.298	0.114				
5cg (R)	0.009	18.39	0.000489	0.120 ± 0.015	7.00 ± 2.11	0.017	MAO-A, competitive, reversible
5cg (S)	0.548	10.53	0.052				
5ch (R)	8.980	755.08	0.012	9.26 ± 0.35	805.20 ± 36.45	0.011	MAO-A, competitive, reversible
5ch (S)	62.189	292.00	0.213				
Selegiline (MAO-B inhibitor)	22.02	34.07	0.646	9.06 ± 0.44	0.09 ± 0.004	100.67	MAO-B, competitive irreversible
Moclobemide (MAO-A inhibitor)	5.71	250.74	0.023	0.005 ± 0.001	1.22 ± 0.08	0.004	MAO-A, competitive, reversible

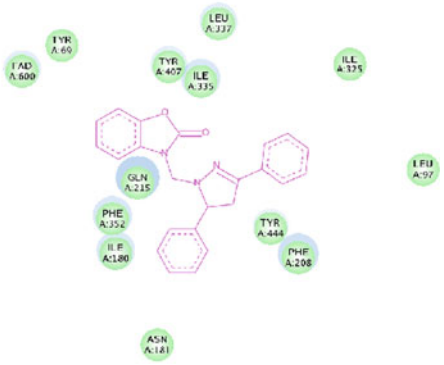
* Selectivity index. It was calculated as K_i(MAO-A)/K_i(MAO-B)

** Each value represents the mean ± SEM of three independent experiments

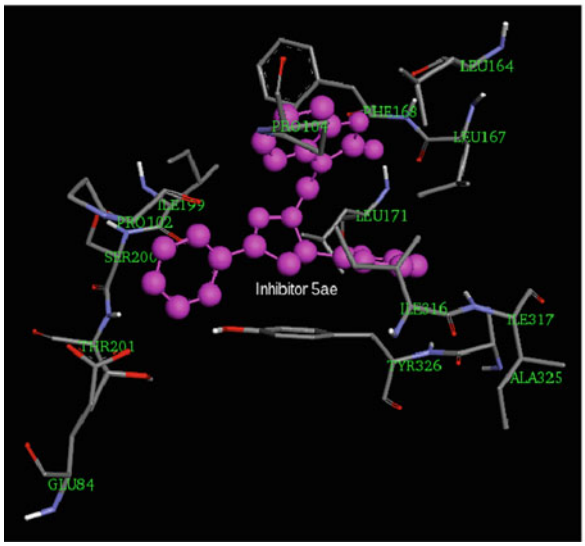
significant σ–π interactions with the side chains of PHE352 and PHE208. ASN181, ILE325, LEU97, GLN215, ILE335, LEU337, and TYR69 contribute to the other attractions. The last two pictures of Fig. 1 show the poses of **5ae** in the active side of MAO-B in 3-D and 2-D depictions, respectively. On the contrary, MAO-A compound **5ae** occupies a space in the entrance cavity of MAO-B very far from the main cavity and hydrophobic packet. The phenyl rings of **5ae** make two σ–π interactions with SER200 and TYR326. The selectivity and potency of compound **5ae** on MAO-A compared to MAO-B can be noted in the above poses in MAO-A and MAO-B. The experimental data given in Table 2 are in agreement with these observations. All the computational results may suggest why the MAO-A



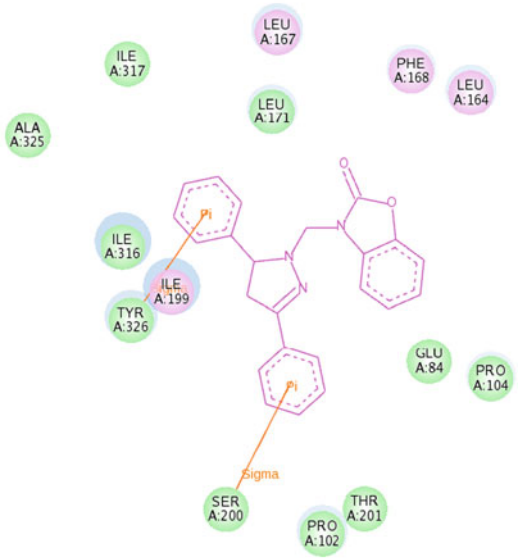
MAO-A 3-D



MAO-A 2-D



MAO-B 3-D



MAO-B 2-D

Fig. 1 Docked pose of compound **5ae (R)** in MAO-A and MAO-B active site in 3-D and 2-D, respectively. Amino acid side chains are shown as *sticks*, the inhibitor is shown as a *ball* and *stick* (*magenta*), and the cofactor FAD is depicted as a *yellow stick*. Residues involved in hydrogen bonding or polar interactions are represented by *magenta-colored circles*, and residues involved in vdW and hydrophobic interactions are shown by *green circles* in all 2-D figures

inhibitory potency of inhibitor **5ae** ($K_i = 0.001$ μM) is much better and more selective in comparison to MAO-A ($K_i = 1.20$ μM).

References

Anderson MC, Hasan F, McCrodden JM, Tipton KF (1993) Monoamine oxidase inhibitors and the cheese effect. Neurochem Res 18:1145–1149

Bilgin AA, Palaska E, Sunal R (1993) Studies on the synthesis and antidepressant activity of Some 1-thiocarbamoyl-3,5-diphenyl-2-pyrazolines. Arzneimittel-Forsch 43:1041–1044

Binda C, Wang J, Pisani L, Caccia C, Carotti A, Salvati P, Edmondson DE, Mattevi A (2007) Structures of human monoamine oxidase B complexes with selective noncovalent inhibitors: safinamide and coumarin analogs. J Med Chem 50:5848–5852

Bradford MM (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal Biochem 72:248–254

Çakır B, Dağ Ö, Yıldırım E, Erol K, Şahin MF (2001) Synthesis and anticonvulsant activity of some hydrazones of 2[(3H)-oxobenzoxazolin-3-yl-aceto]hydrazide. J Fac Pharm Gazi 18:99–106

Chen SQ, Zhang YC, Liu FM (2011) Synthesis and spectral characterization of some new thiazolyl-pyrazolines bearing 1,2,4-triazole moiety. Phosphorus Sulfur 186:319–325

Chimenti F, Bolasco A, Manna F, Secci D, Chimenti P et al (2004) Synthesis and selective inhibitory activity of 1-acetyl-3,5-diphenyl-4,5-dihydro-(1*H*)-pyrazole derivatives against monoamine oxidase. J Med Chem 47:2071–2074

Chimenti F, Maccioni E, Secci D, Bolasco A, Chimenti P, Granese A et al (2005) Synthesis, molecular modeling studies, and selective

inhibitory activity against monoamine oxidase of 1-thiocarbamoyl-3,5-diaryl-4,5-dihydro-(1*H*)-pyrazole derivatives. J Med Chem 48:7113–7122

Chimenti F, Bolasco A, Manna F, Secci D, Chimenti P, Granese A, Befani O, Turini P, Alcaro S, Ortuso F (2006a) Synthesis and molecular modelling of novel substituted-4,5-dihydro-(1H)-pyrazole derivatives as potent and highly selective monoamine oxidase-A inhibitors. Chem Biol Drug Des 67:206–214

Chimenti F, Bolasco A, Manna F, Secci D, Chimenti P, Granesea A, Befani O, Turini P, Cirilli R et al (2006b) Synthesis, biological evaluation and 3D-QSAR of 1,3,5-trisubstituted-4,5-dihydro-(1H)-pyrazole derivatives as potent and highly selective monoamine oxidase A inhibitors. Curr Med Chem 13:1411–1428

Chimenti F, Fioravanti R, Bolasco A, Manna F, Chimenti P et al (2007) Monoamine oxidase isoform-dependent tautomeric influence in the recognition of 3,5-diaryl pyrazole ınhibitors. J Med Chem 50:425–428

Chimenti F, Fioravanti R, Bolasco A, Manna F, Chimenti P et al (2008a) Synthesis, molecular modeling studies and selective inhibitory activity against MAO of N1-propanoyl-3,5-diphenyl-4,5-dihydro-(1H)-pyrazole derivatives. Eur J Med Chem 43:2262–2267

Chimenti F, Maccioni E, Secci D, Bolasco A, Chimenti P et al (2008b) Synthesis, stereochemical identification, and selective inhibitory activity against human monoamine oxidase-B of 2-methylcyclohexylidene-(4-arylthiazol-2-yl)hydrazones. J Med Chem 51:4874–4880

Chimenti F, Carradori S, Secci D, Bolasco A, Bizzarri B et al (2010) Synthesis and inhibitory activity against human monoamine oxidase of N1-thiocarbamoyl-3,5-di(hetero)aryl-4,5-dihydro-(1*H*)-pyrazole derivatives. Eur J Med Chem 45:800–804

Close WJ, Tiffany BD, Spielman MA (1949) The analgesic activity of some benzoxazolone derivatives. J Am Chem Soc 71:1265–1268

Dawey W, Tivey DJ (1958) Chalcones and related compounds. Part IV. Addition of hydrogen cyanide to chalcones 242:1230–1236

Dong F, Jian C, Zhenghao F, Kai G, Zuliang L (2008) Synthesis of chalcones via Claisen–Schmidt condensation reaction catalyzed by acyclic acidic ionic liquids. Catal Commun 9:1924–1927

Drukarch B, van Muiswinkel FL (2000) Drug treatment of Parkinson's disease. Time for phase II. Biochem Pharmacol 59:1023–1031

Gökçe M, Geciken AE, Yıldırım E, Tosun AU (2001) Synthesis and anticonvulsant activity of 5-chloro-2(3H)-benzoxazolinone-3-acetyl-2-(o/p-substituted benzal)hydrazone derivatives. Arzneimittelforschung 58:537–542

Gökhan N, Yeşilada A, Uçar G, Erol K, Bilgin AA (2003) 1-N-Substituted thiocarbamoyl-3-phenyl-5-thienyl-2-pyrazolines: synthesis and evaluation as MAO inhibitors. Arch Pharm Pharm Med Chem 336:362–371

Gökhan-Kelekçi N, Yabanoğlu S, Küpeli E, Salgın U, Özgen Ö et al (2007) A new therapeutic approach in alzheimer disease: some novel pyrazole derivatives as dual MAO-B inhibitors and antiinflammatory analgesics. Bioorg Med Chem 15:5775–5786

Gökhan-Kelekçi N, Koyunoğlu S, Yabanoğlu S, Yelekçi K, Özgen Ö et al (2009) New pyrazoline bearing 4(3H)-quinazolinone inhibitors of monoamine oxidase: synthesis, biological evaluation, and structural determinants of MAO-A and MAO-B selectivity. Bioorg Med Chem 17:675–689

Holla BS, Akberali PM, Shivananda MK (2000) Studies on arylfuran derivatives-Part X. Synthesis and antibacterial properties of arylfuryl-Δ^2-pyrazolines. Il Farmaco 55:256–263

Irie K, Watanabe K (1980) Aldol condensations with metal(II) complex catalysts. Bull Chem Soc Jpn 53:1366–1371

Kubota Y, Ikeya H, Sugi Y, Yamada T, Tatsumi T (2006) Organic-inorganic hybrid catalysts based on ordered porous structures for Michael reaction. J Mol Catal A-Chem 249:181–190

Lipson VV, Desenko SM, Shirobokova MG, Borodina VV, Musatov VI (2005) Chemical reactions of 2-methyl-5,7-diphenyl-6,7-dihydropyrazolo[1,5-*a*]pyrimidine (New York, NY, United States). Chem Heterocycl Compd 41:492–495

Manna F, Chimenti F, Bolasco A, Secci D, Bizzarri B et al (2002) Inhibition of amine oxidases activity by 1-acetyl-3,5-diphenyl-4,5-dihydro-(1H)-pyrazole derivatives. Bioorg Med Chem Lett 12:3629–3633

Milcent R, Akhnazarian A, Lensen N (1996) Synthesis of 1-(2-hydroxyphenyl)-2,4-imidazolidinedione derivatives through cyclic transformations of ethyl 2-oxo-3(2H)-benzoxazoleacetate derivatives. J Heterocycl Chem 33:1829–1833

Morris GM, Huey R, Lindstrom W, Sanner MF, Belew RK, Goodsell DS, Olson AJ (2009) Autodock4 and AutoDockTools4: automated docking with selective receptor flexibility. J Comp Chem 16:2785–2791

Önkol T, Gökçe M, Tosun AU, Polat S, Serin MS, Tezcan S (2008) Microwave synthesis and antimicrobial evaluation of 5-chloro-2(3H)-benzoxazolinone-3-acetyl-2-(p-substituted benzal)hydrazone and 5-chloro-2(3H)-benzoxazolinone-3-acetyl-2-(p-substituted acetophenone)hydrazone derivatives. Turk J Pharm Sci 5:155–166

Özdemir Z, Kandilci HB, Gümüşel B, Çalış Ü, Bilgin AA (2007) Synthesis and studies on antidepressant and anticonvulsant activities of some 3-(2-furyl)-pyrazoline derivatives. Eur J Med Chem 42:373–379

Özdemir Z, Kandilci HB, Gümüşel B, Çalış Ü, Bilgin AA (2008) Synthesis and studies on antidepressant and anticonvulsant activities of some 3-(2-thienyl)-pyrazoline derivatives. Arch Pharm Chem Life Sci 341:701–707

Palaska E, Aytemir M, Uzbay İT, Erol D (2001) Synthesis and antidepressant activities of some 3,5-diphenyl-2-pyrazolines. Eur J Med Chem 36:539–543

Palaska E, Aydın F, Uçar G, Erol D (2008) Synthesis and monoamine oxidase inhibitory activities of 1-thiocarbamoyl-3,5-diphenyl-4,5-dihydro-1H-pyrazole derivatives. Arch Pharm Chem Life Sci 341:209–215

Potts KT, Bhattacharjee D, Kanemasa S (1980) Mesoionic compounds. 52. Attempted synthesis of the anhydro-2-hydroxyoxazolo[2,3*b*-]oxazolium hydroxide system. J Org Chem 45:4985–4988

Şahin ZS, Salgın-Gökşen U, Gökhan-Kelekçi N, Işık Ş (2011) Synthesis, crystal structures and DFT studies of 1-[2-(5-methyl-2-benzoxazolinone-3-yl)acetyl]-3-phenyl-5-(3,4-dimethoxyphenyl)-4,5-dihydro-1H-pyrazole and 1-[2-(5-chloro-2-benzoxazolinone-3-yl)acetyl]-3-phenyl-5-(4-methoxyphenyl)-4,5-dihydro-1H-pyrazole. J Mol Struct 1006:147–158

Salgın-Gökşen U, Gökhan-Kelekçi N, Göktaş Ö, Köysal Y, Kılıç E et al (2007) 1-Acylthiosemicarbazides, 1,2,4-triazole-5(4H)-thiones, 1,3,4-thiadiazoles and hydrazones containing 5-methyl-2-benzoxazolinones: synthesis, analgesic-anti-inflammatory and antimicrobial activities. Bioorg Med Chem 15:5738–5751

Sarabhai KP, Mathur KBL (1963) Some arylation reactions with diazotized 3,4,5-trimethoxyaniline. Preparation of 3,4,5-trimethoxychalcone and 3,4,5-trimethoxybenzaldehyde. Indian J Chem 1:482–483

Schapira AH (2007) Treatment options in the modern management of Parkinson disease. Arch Neurol 64:1083–1088

Secci D, Bolasco A, Chimenti P, Carradori S (2011) The state of the art of pyrazole derivatives as monoamine oxidase inhibitors and antidepressant/anticonvulsant agents. Curr Med Chem 18:5114–5144

Shekarchi M, Pirali-Hamedani M, Navidpour L, Adib N, Shafiee A (2008) Synthesis, antibacterial and antifungal activities of 3-aryl-5-(pyridin-3-yl)-4,5-dihydropyrazole-1-carbothioamide derivatives. J Iran Chem Soc 5:150–158

Son S-Y, Ma J, Kondou Y, Yoshimura M, Yamashita E, Tsukihara T (2008) Structure of human monoamine oxidase A at 2.2-Å resolution: the control of opening the entry for substrates/inhibitors. P Natl Acad Sci USA 105:5739–5744

Ueno Y, Yadav LDS, Okawara M (1983) Carbon-carbon bond formation via phosphine-initiated cleavage of oxosulphides. Chem Lett 6:831–834

Ünlü S, Erdoğan H, Sunal R (1992) Synthesis of some (2-benzoxazolinones-3-yl)alkonoic acid derivatives and their analgesic properties. Hacettepe University J Faculty of Pharm 12:23–31

Yamada M, Yasuhara H (2004) Clinical pharmacology of MAO inhibitors: safety and future. Neurotoxicology 25:215–221

Yáñez M, Fraiz N, Cano E, Orallo F (2006) Inhibitory effects of cis- and trans-resveratrol on noradrenaline and 5-hydroxytryptamine uptake and on monoamine oxidase activity. Biochem Biophys Res Commun 344:688–695

Yelekçi K, Karahan Ö, Toprakci M (2007) Docking of novel reversible monoamine oxidase-B inhibitors: efficient prediction of ligand binding sites and estimation of inhibitors thermodynamic properties. J Neural Transm 114:725–732