Journal of Pharmaceutical and Health Sciences 2016;4(2), 101-108.

Original Article

JPHS

Synthesis and docking study on thiadiazolo[3,2-a][1,3]diazepin-8(5H)-one derivatives as selective GABA(A) agonists

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Abstract

HIE-124 is a new member of ultra-short acting hypnotics' drug family. In this research, the synthesis of analogues of HIE-124 drug in the heterocyclic thiazole ring replaced to thiadiazole, will be presented. Thiadiazolodiazepines during a two-step reaction starting from the amino thiadiazole resulted from-various derivatives of benzoic acid and thiosemicarbazide were synthesized. In the first step, the reaction of synthetic raw material 2-amino thiadiazole and 4-chlorobutyrilchloride in toluene solvent give the 4-chloro-N-(5-(methyl/aryl)-1,3,4-thiadiazol-2-yl) butanamide intermediate. In the next step, from the cyclization reaction of this intermediate ring in the presence of base under reflux, the target products are synthesized. Structure of products was identified based on IR, HNMR and CNMR spectroscopy analysis. Then, the procedure of docking of ligands were performed on the active site of GABAA that the common residues involved in allosteric modulators such as benzodiazepines and HIE-124 include ASN82, ASN81, PHE79, MET1, TYR106, ALA38 and AlA168. Consequently, These Docking calculations suggest that these new compounds might be having better interaction results between receptor (GABAA) than HIE-124.

Keywords: HIE-124, Thiopental Sodium, 4-Chlorobutyryl chloride, Thiadiazolodiazepines, Piperidine, Thiosemicarbazide, Docking, GABAA

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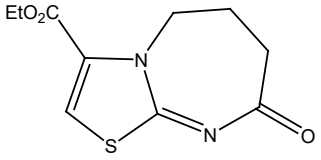
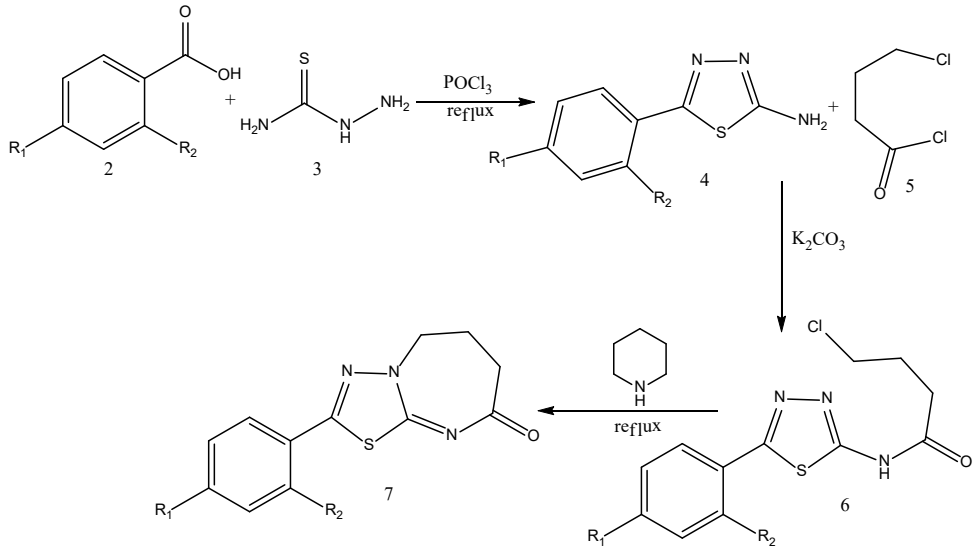
INTRODUCTION

General anesthesia most often is initiated by an injection of thiopental, an ultra-short acting barbiturate, to induce sleep prior to administration of the agents that are necessary for maintaining anesthesia during the surgical procedure. Ultra-short acting barbiturates and benzodiazepines have an important place in the practice of anesthesiology. Thiopental sodium remains the standard for comparison with new agents (Hughes et al. 1992). Thiopental is metabolized slowly in the liver, which together with other factors, may influence the depth of anesthesia, time of recovery, and duration of action (Breimer, 1997). The concentration employed of thiopental should not exceed 2.5% in aqueous solution. When greater concentration is injected extra vascular, severe pain and tissue necrosis may occur. Intra-arterial injection of a concentrated solution of thiopental may result in damage to the arterial endothelium, followed by endarteritis, often with thrombosis exacerbated by subsequent arteriolar spasm. Vascular ischemia and even gangrene may result (Marshall & Longnecker, 1996). Thiopental produces a long lasting hangover (Rang et al. 2003). Patients using a large initial dose of thiopental will awake despite plasma concentrations that normally would cause sleep. The nature of this acute tolerance is not known. For this reason, thiopental cannot be used to maintain surgical anesthesia, but only as an induction agent. Thiopental produces a dose-related depression of respiration that can be profound (Rang et al. 2003; Marshall et al. 1995; Hapiro, 1975). Larger doses of thiopental sodium cause more profound changes and respiration is maintained only by movements of the diaphragm. Diazepines have showed a large spectrum of biological activities, for example, Thiazolo-[3, 2-a][1,3] Diazepine and their derivatives show a wide range of ultra-short acting hypnotics (Lehmann et al. 2004; El-Subbagh et al. 2011; Cheeseman & Eccleshall, 1986). Several range of chemical compounds such as benzodiazepines and some new thiazolodiazepines analogs as CNS active agents. Consequently they bind to the GABAA chloride ion channel complex (Sieghart, 1995). GABAA, 4-aminobutyric acid or 1, 4-aminobutanoic was discovered in 1950, is the main inhibitory transmitter in the brain and inhibition of neurons (Smith & Olsen, 1995; McKernan et al. 2000). HIE-124 1 has been shown to not only induce anesthesia but also maintain the anesthetic state during the surgical procedure. HIE-124 exhibited a very rapid onset of action and a shorter duration of action with no acute tolerance or noticeable side effects when compared with thiopental sodium. This new finding granted the issuance of a patent (Lehmann et al. 2004). In the present investigation, we would like to report the synthesis of 6, 7-dihydro-[1,3,4] thiadiazolo-[3,2-A][1,3] diazepin derivatives (HIE-124 analogous), a member of a novel class which might overcomes many of the disadvantages and problems that are usually associated with the use of thiopental or benzodiazepines as intravenous anesthetic agents. The thiadiazolo [3, 2-a][1,3] diazepine nucleus, can be obtained by the use of published methods (Imming, 1995). 6,7-dihydro-[1,3,4] thiadiazolo-[3,2-A] [1,3] diazepin derivatives 7, was synthesized according to an inventive method (Scheme 2). For the synthesis of HIE-124 analogues 7 in the first step, from reaction of benzoic acid derivatives 2 with thiosemicarbazide in the solvent of phosphoryl chloride, 5-aryle-1,3,4-thiadiazol-2-amine 4 were prepared (Salih et al. 2011; Daoud & Eisa, 2005). Then, 5-aryle-1, 3, 4-thiadiazol-2-amine with 4-chlorobutyryl chloride 5 and potassium carbonate in toluene was heated under reflux for 4 hr. The solid obtained 6 was washed and dried. In the next step, Compound 6 was cyclized using piperidine as a base to produce HIE-124 analogues 7. The reaction mixture was cooled, poured into water and stirred. The toluene was then evaporated under reduced pressure to give a crude product which was purified by column chromatography. Structure elucidation of

compounds 6 and 7 was obtained based on analysis of the IR, 1H- and 13C-NMR spectra spectrometry for each compound.



Scheme 1: HIE-124



Scheme 2: Synthesis of 6, 7-dihydro-[1, 3, 4] thiadiazolo-[3,2-A][1,3] diazepin derivatives

Table 1: Synthesis of 6,7-dihydro-[1, 3, 4] thiadiazolo-[3,2-A][1,3]diazepin derivatives

Entry	R1	R2	Products		
			4	6	7
a	NO2	67	93	93	H
b	Cl	64	85	91	H
c	H	61	89	88	H
d	OMe	55	61	85	H
e	H	56	84	76	F

RESULT AND DISCUSSION

The IR spectrum of 7b showed single absorption at 1711 cm-1 indicating the presence of carbonyl group. The 1HNMR spectrum of 6b exhibits two triplet signal at δ = 2.69 ppm, δ = 3.71 ppm and a multiplet at δ = 2.09 ppm with 3JHH of about 7.5 Hz which are related tree methylene group which confirmed unambiguously the formation of a amide group. The off resonance decoupled 13C-NMR spectra of 6b exhibited characteristic peaks for the carbonyl group and methylene group attached to the carbonyl at 171.3 and 32.6 ppm respectively.

The 1H-NMR spectrum of 7b exhibits two triplet signal at δ = 2.68 ppm, δ =4.13 ppm and a multiplet at δ = 2.22 ppm with 3JHH of about 7.5 Hz which are related tree methylene group which confirmed the formation of a cyclic. The 13C-NMR spectra of 7b exhibited characteristic peaks for the carbonyl group and methylene group attached to the carbonyl at 174.8 and 31.2 ppm respectively.

In present docking study, the procedure of docking of ligands were performed on the active site of GABAA that the common residues involved in allosteric modulators such as benzodiazepines and HIE-124 include ASN82, ASN81, PHE79, MET1, TYR106, ALA38 and AlA168 (figure 1). The analysis of the docking parameter file in this software AutoDock have showed that every docking contains some useful knowledge which includes binding energy (Eb) which is the sum of the intermolecular energy, the torsional energy and the internal energy which are reported in table 2. The study and compare of all dockings indicates that HIE-124 and thiadiazolo[3,2-a][1,3]diazepin-8(5H)-one derivatives with the receptor protein, GABAA, according to table 2, the complex of HIE-124 has the lowest amount of binding energy (-4.35) with other compounds binding energy (Eb).

Table 2: Docking results Thiadiazolo[3,2-a][1,3]diazepin-8(5H)-one Derivatives by Autodock 4 software

Compound	Intermolecular energy	Vdwhbdesolve energy	Inhibconstant	Electrostatc energy	Total internal energy	Torsional energy	Unbound energy	Lg and efficiency	Ref RMS	Building energy
HIE-124	-5.2	693.3	-5.13	0.89	-0.4	-4.31	8.99	-0.27	-0.4	0.89
7a	-5.99	67.59	-5.96	0.3	-0.25	-5.69	12.33	0.32	-0.25	0.3
7b	-6.97	21.45	-5.92	0.6	-0.32	-6.37	8.8	-0,32	-0.32	0.6
7c	-6.54	43.7	-6.47	0.6	-0.29	-5.95	9.29	-0.31	-0.29	0.6
7d	-5.86	83.83	-5.94	0.3	-0.24	-5.56	12.81	-0.33	-0.24	0.3
7e	-5.87	82.85	-5.8	0.3	-0.22	-5.57	13.2	-0.31	-0.22	0.3

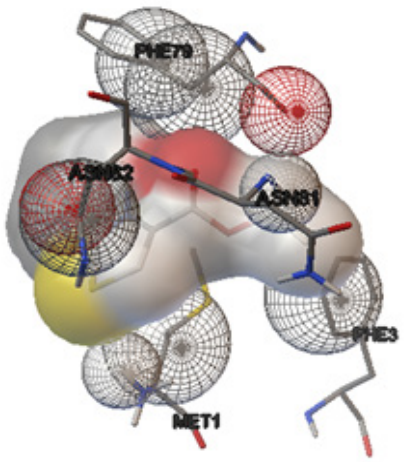


Figure 1: Docked structures of HIE-124 in GABA with pdb code 1KJT are represented.

Our docking results show that HIE-124 forms one hydrogen bonding interactions with ASN82 (distance = 2.033) (energy = -4.021) while don't form any π-cation or π-π interactions (Figure

2). The Compound 7b has a hydrogen bonding interaction with MET1 (distance = 2.156) (energy = -1.16) while don't form any π-cation or π-π interactions (Figure 3).

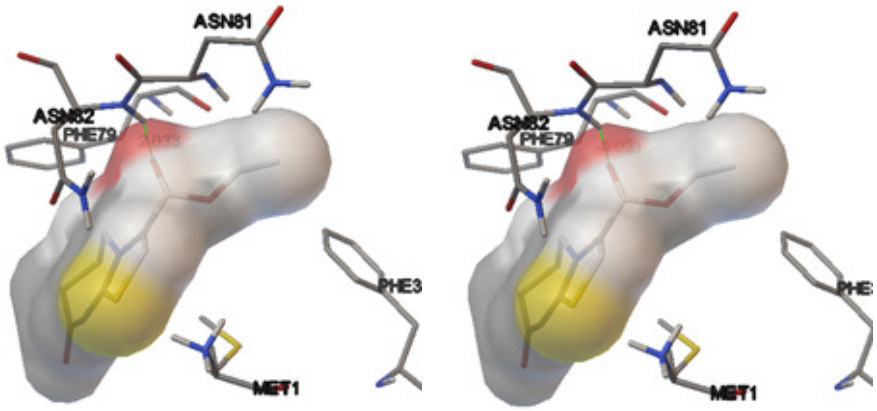


Figure 2: Docked structures of HIE-124 in GABAA with pdb code 1KJT as hydrogen bond distance and hydrogen bond energy are represented.

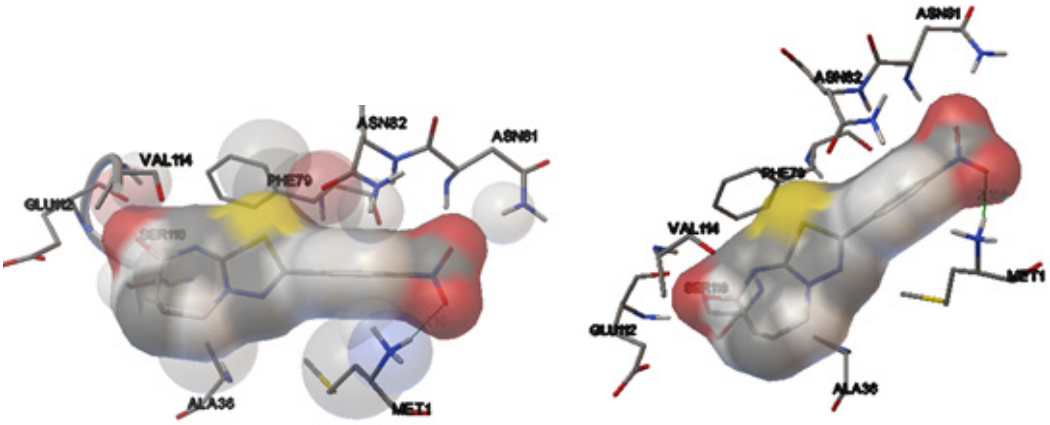


Figure 3: Docked structures of 7b in GABAA with pdb code 1KJT as hydrogen bond distance and hydrogen bond energy are represented.

In the compounds 7a, 7c, 7d and 7e don't have any hydrogen bonding interaction, any π-π interactions and any π-cation interactions. According to the Ki, all of compounds 7a-7e with 67.59, 21.45, 43.7, 83.83, and 82.85 inhibition-constant can inhibit the enzyme more efficiently when compared to HIE-124 with 693.3 inhibition-constant. Docking studies have suggested that Thiadiazolo [3, 2-a][1,3] diazepin-8(5H)-one Derivatives have important role in their high binding energies than HEI-124. Furthermore, our findings suggest that interaction results between receptor (GABAA) than HIE-124.

MATERIALS AND METHODS

In the present work, all the ligands used were made using GaussSum program. Before the docking calculation of the ligands, the structures were fully optimized. Docking calculations were calculated by using Autodock 4.2.6. Crystal structure of the GABA (A) receptor as-

sociated were retrieved from Protein Databank (PDB) (http://www.pdb.org/), PDB id 1KJT, having resolution of 2.0 Å. All heteroatoms were removed from the PDB files and water molecules removed in the Notepad++ software. Kollman partial charges were assigned to all protein atoms. Autogrid was carried out for the preparation of the grid map using grid boxes of 28-28-28 A°points of 1.00A° spacing. We studied docking calculations in some grid boxes, finally we had found the best grid box for calculations it was grid boxes of 28-28-28 A°points of 1.00A°. A Lamarckian genetic algorithm (Amber force field) was used. A population of 150 individuals and 2,500,000 function evaluations were applied. Numbers of GA Runs 100 were applied. At the end of calculations, the best superimposing poses were chosen for the analysis.

Experimental

General melting point was recorded on an electrothermal digital melting point apparatus. Infrared spectra were recorded on a mattson 1000 FTIR. 1H, 13CNMR spectra were measured with a Bruker DRX-300 AVANCE instrument with CDCl3 as solvent at 300.1 MHz. Mass spectra were recorded on a Finnigan MAT 8430 mass spectrometer operating at an electron energy of 70 eV. thiosemicarbazide and benzoic acid derivatives, were obtained from Fluka (Buchs, Switzerland) and were used without further purification, and amino thiadiazole derivatives were obtained via synthesized.

General procedure for synthesis of 5-aryle-1, 3, 4-thiadiazol-2-amine (4)

A mixture of benzoic acid derivatives (2, 0.02 mol) and thiosemicarbazide (3, 0.02 mol) in phosphoryl chloride (150 mL) was heated under reflux for 3 h. The phosphoryl chloride was then evaporated under reduced pressure. Then, the residue was quenched with water, stirred, and filtered. The solid obtained was washed, dried, and recrystallized from water to give the required product 4.

General procedure for synthesis of 4-chloro-N-(5- aryle -1,3,4-thiadiazol-2-yl)butanamide (6)

A mixture of 5-aryle-1,3,4-thiadiazol-2-amine (4, 0.02 mol) and 4-chloro-butyryl chloride (5, 0.03 mol) in toluene (100 mL) was heated under reflux for 4 h. The toluene was then evaporated under reduced pressure. Then, the residue was mixed with water, stirred, and filtered. The solid obtained was washed, dried, and recrystallized from water to give the required product 6.

4-chloro-N-(5-(nitrophenyl)-1,3,4-thiadiazol-2-yl)butanamide(6a): M.p. 218–220 C; 1HNMR (CDCl3) 2.09 (m, 2H, CH2), 2.69 (t, 2H, J=7.5, CH2), 3.71 (t, 2H, J=7.5 Hz, CH2), 7.66 (d, 2H, J=7.5 Hz, Ar-H), 7.98 (d, 2H, J=7.5 Hz, Ar-H), 12.20 (brs, 1H, NH). 13CNMR 27.8, 32.6, 45.3, 129.2, 129.8, 130.9, 135.9, 159.8, 161.2, 171.3.

4-chloro-N-(5-(4-chlorophenyl)-1,3,4-thiadiazol-2-yl)butanamide(6b): M.p. 224–226 C; 1HNMR (CDCl3) 2.09 (m, 2H, CH2), 2.68 (t, 2H, J=7.5, CH2), 3.71 (t, 2H, J=7.5 Hz, CH2), 7.59 (d, 2H, J=7.5 Hz, Ar-H), 7.96 (d, 2H, J=7.5 Hz, Ar-H), 12.4 (brs, 1H, NH). 13CNMR 27.8, 32.6, 45.2, 129.0, 129.5, 129.9, 135.6, 159.0, 161.2, 171.2.

4-chloro-N-(5-(phenyl)-1,3,4-thiadiazol-2-yl) butanamide (6c): M.p. 198–200 C; 1HNMR (CDCl3) 2.07 (m, 2H, CH2), 2.68 (t, 2H, J=7.5, CH2), 3.70 (t, 2H, J=7.5 Hz, CH2), 7.53, 7.93 (m, 5H, Ar-H), 12.4 (brs, 1H, NH). 13CNMR 27.8, 32.6, 45.2, 127.4, 129.9, 130.6, 131.1, 159.1, 161.2, 171.2.

4-chloro-N-(5-(4-methoxyphenyl)-1,3,4-thiadiazol-2-yl)butanamide(6d): M.p. 171–172 C; 1HNMR (CDCl3) 2.06 (m, 2H, CH2), 2.67

(t, 2H, J=7.5, CH2), 3.71 (t, 2H, J=7.5 Hz, CH2), 7.57 (d, 2H, J=7.5 Hz, Ar-H), 7.91 (d, 2H, J=7.5 Hz, Ar-H), 12.4 (brs, 1H, NH). 13CNMR 27.6, 32.6, 45.2, 127.4, 129.9, 130.6, 131.2, 159.1, 161.2, 171.2.

4-chloro-N-(5-(2-fluorophenyl)-1,3,4-thiadiazol-2-yl)butanamide(6e): M.p. 188–190 C; 1HNMR (CDCl3) 2.06 (m, 2H, CH2), 2.67 (t, 2H, J=7.5, CH2), 3.71 (t, 2H, J=7.5 Hz, CH2), 7.31 (m, 2H, Ar-H), 7.45 (d, 1H, Ar-H), 8.35 (t, 1H, Ar-H), 12.4 (brs, 1H, NH). 13CNMR 27.6, 32.6, 45.2, 116.0, 116.4, 124.7, 124.8, 128.7, 128.8, 132.0, 132.1, 159.1, 161.2, 171.2.

General procedure for synthesis of 4-chloro-N-(5- aryle -1,3,4-thiadiazol-2-yl)butanamide (7)
A mixture of 4-chloro-N-(5- aryle -1,3,4-thiadiazol-2-yl)butanamide (6, 0.004 mol) and piperidine (0.8 mL, 0.008 mol) in toluene (50 mL) was heated under reflux for 3 h. The reaction mixture was cooled, poured into water, and stirred. Toluene was separated, dried, and evaporated to give a crude product, which was purified by repeated silica gel column chromatography eluting with CH2Cl2/hexane (80 : 20 v/v) to give 7.

2-(4-nitrophenyl)-6,7-dihydro-[1,3,4]thiadiazolo [3,2-a] [1,3]diazepin-8(5H)-one (7a): M.p. 188–190 C; 1HNMR (CDCl3) 2.25 (m, 2H, CH2), 2.69 (t, 2H, J=7.5, CH2), 4.14 (t, 2H, J=7.5 Hz, CH2), 8.24 (d, 2H, J=8.3 Hz, Ar-H), 8.35 (d, 2H, J=8.3 Hz, Ar-H). 13CNMR 27.8, 32.6, 45.3, 129.2, 129.8, 130.9, 135.9, 159.8, 161.2, 173.3.

2-(4-chlorophenyl)-6,7-dihydro-[1,3,4] thiadiazolo[3,2-a][1,3]diazepin-8(5H)-one (7b): M.p. 215–217 C; 1HNMR (CDCl3) 2.22 (m, 2H, CH2), 2.68 (t, 2H, J=7.5, CH2), 4.13 (t, 2H, J=7.5 Hz, CH2), 7.60 (d, 2H, J=7.5 Hz, Ar-H), 7.95 (d, 2H, J=7.5 Hz, Ar-H). 13CNMR 18.2, 31.2, 45.2, 129.0, 129.4, 129.9, 135.8, 157.4, 163.9, 174.8.

2-phenyl-6,7-dihydro-[1,3,4]thiadiazolo[3,2-a] [1,3]diazepin-8(5H)-one(7c): M.p. 224–227 C; 1HNMR (CDCl3) 2.31 (m, 2H, CH2), 2.71 (t, 2H, J=7.5, CH2), 4.25 (t, 2H, J=7.5 Hz, CH2), 7.46, 7.95 (m, 5H, Ar-H). 13CNMR 18.2, 31.3, 47.9, 127.4, 128.8, 129.1, 130.6, 157.4, 163.9, 173.7.

2-(4-methoxyphenyl)-6,7-dihydro-[1,3,4] thiadiazolo[3,2-a][1,3]diazepin-8(5H)-one(7d): M.p. 210–213 C; 1HNMR (CDCl3) 2.32 (m, 2H, CH2), 2.72 (t, 2H, J=7.5, CH2), 4.21 (t, 2H, J=7.5 Hz, CH2), 7.44 (d, 2H, J=7.5 Hz, Ar-H), 7.95 (d, 2H, J=7.5 Hz, Ar-H). 13CNMR 18.6, 32.2, 47.2, 129.4, 129.9, 130.6, 131.2, 159.1, 161.4, 172.2.

2-(2-fluorophenyl)-6,7-dihydro-[1,3,4]thiadiazolo [3,2-a][1,3]diazepin-8(5H)-one(7e): M.p. 202–204 C; 1HNMR (CDCl3) 2.35 (m, 2H, CH2), 2.75 (t, 2H, J=7.5, CH2), 4.29 (t, 2H, J=7.5 Hz, CH2), 7.31 (m, 2H, Ar-H), 7.45 (d, 1H, Ar-H), 8.35 (t, 1H, Ar-H). 13CNMR 18.4, 31.3, 47.9, 116.0, 116.4, 124.7, 124.8, 128.7, 128.8, 132.0, 132.1, 159.1, 161.2, 173.7.

CONCLUSION

In conclusion, We synthesized of a new series of 1,3-benzodiazepine derivatives 7 during two-step reaction starting from the amino thiadiazole resulted from-various derivatives of benzoic acid and thiosemi-carbazide were synthesized. In the first stage, the reaction of synthetic raw material 2-amino thiadiazole and 4-chlorobutyrilchloride in toluene solvent give the 4-chloro-N-(5-(methyl/aryl)-1,3,4-thiadiazol-2-yl) butanamide intermediate. In the next step, from the cyclization reaction of this intermediate ring in the presence of base under reflux, the target products are synthesized. The structures of the products were elucidated us-

ing IR, 1H NMR and 13CNMR spectral data. The procedure of docking of ligands were performed on the active site of GABAA that the common residues involved in allosteric modulators such as Benzodiazepines and HIE-124 include ASN82, ASN81, PHE79, MET1, TYR106, ALA38 and AlA168. Consequently, These Docking calculations suggest that these new compounds might be having better interaction results between receptor (GABAA) than HIE-124.

Acknowledgments

We gratefully acknowledge financial support from the Research Council of Imam Hossein University.

Supplementary Material

Supplementary data (included are general procedures and IR, 1H and 13C NMR data of all compounds) associated with this article can be found in the online version.

References

Breimer DD. Clinical pharmacokinetics of hypnotics. Clin. Pharmacokinet 1977; 2(2): 93–109.

Cheeseman GWH, Eccleshall, SA. Synthesis of pyrrolo[1,2-a]naphtho[1,8-ef][1,4] diazepines. J. Heterocycl. Chem, 1986; 23(1), 65–67.

Daoud KM, Eisa MAA. Synthesis of some Substituted Multinuclear 1,3,4-Oxadiazoles and 1,3,4-Thiadiazoles. National J of Chem, 2005; 19: 405-411.

El-Subbagh HI, Hassan GS, El-Azab AS, Alaa AM, Kadi AA, Al-Obaid AM, Al-Shabanah OA, Sayed-Ahmed MM. Synthesis and anticonvulsant activity of some new thiazolo[3,2-a][1,3] diazepine, benzo[d] thiazolo[5,2-a][12,6] diazepine and benzo[d]oxazolo [5,2-a][12,6]diazepine analogues. Euro J of Medi Chem, 2011; 46(11): 5567-5572.

Hapiro HM. Intracranial hypertension: therapeutic and anesthetic considerations. Anesthesiology 1975; 43(4): 445–471.

Hughes MA, Glass PS, Jacobs JR. Context-sensitive half-time in multicompartment pharmacokinetic models for intravenous anesthetic drugs. Anesthesiology 1992; 76: 334–341.

Imming P. Synthesis of the First Penicillin Derivatives With Medium-Sized Lactam Ring and of Related Thiazolidines. Arch. Pharm 1995; 328: 207-215.

Lehmann J, El-Subbagh HI, El-Kashef HA. Thiazolodiazepnes: New ultra-short Acting Hypnotics, German Patent, 2004; DE 103 2004 732 A1.

Marshall BE, Hanson CW, Marshall C. In A Practice of Anesthesia, (Eds.: D. Healy, P. J. Cohen), Edward Arnold, London, 1995; 119–145.

Marshall BE, Longnecker DE. In The Pharmacological Basis of Therapeutic, (Eds.: J. G. Hardman, L. E. Limbird), 9th Ed, McGraw-Hill, New York, 1996; 321–323.

McKernan RM, Rosahl TW, Reynolds DS, Sur C, Wafford KA, Atack JR, Farrar S, Myers J, Cook G, Ferris P, Garrett L, Bristow L, Marshall G, Macaulay A, Brown N, Howell O, Moore KW, Carling RW, Street LJ, Castro JL, Ragan CI, Dawson GR, Whiting PJ. Sedative but not anxiolytic properties of benzodiazepines are mediated by the GABA(A) receptor alpha1 subtype. Nat. Neurosci, 2000; 3(6): 587-592.

Rang HP, Dale, MM, Ritter JM, Moore PK. Pharmacology, 5th Ed, Churchill Livingstone, New York, 2003; 503–515.

Salih N, Salimon J, Yousif E. Synthesis and antimicrobial evaluation of 5-(p-substituted phenyl)-N-(3-(5-nitrofur-2-yl) allylidene)-1, 3, 4-thiadiazo l-2-amine derivatives. Orient. J. Chem, 2011; 27(2): 373-383.

Sieghart W. Structure and pharmacology of gamma-aminobutyric acidA receptor subtypes. Pharmacol Rev, 1995; 47(2): 181–234.

Smith GB, Olsen RW. Functional domains of GABAA receptors. Trends Pharmacol Sci, 1995; 16(5): 162-168.