

Efficient Synthesis, Characterization and Biological Activity of Tetrazole- Derivatives

Veeranna Guguloth¹, M. Subramanyam², Ravinder Vadde³

¹Department of Chemistry, JVR Government College Sathupally, Khammam Telangana-507303

²Department of Chemistry, SR &BGNR Government Arts and Science College (A) Khammam Telangana-507002

³Department of Chemistry, Kakatiya University Warangal Telangana -506009

Abstract:

A few derivatives of Tetrazole were synthesised by copper mediated N-arylation. The compounds synthesised were characterized by spectral analysis. These compounds were screened for biological activity and found to inhibit L1210 leukemia cell Proliferation and SK-BR-3 breast cancer cell growth.

Keywords: Tetrazole, Boronic acids, characterization, culture, Cu-Catalyzed, Heterogeneous catalytic transfer reductions, Suzuki coupling.

INTRODUCTION

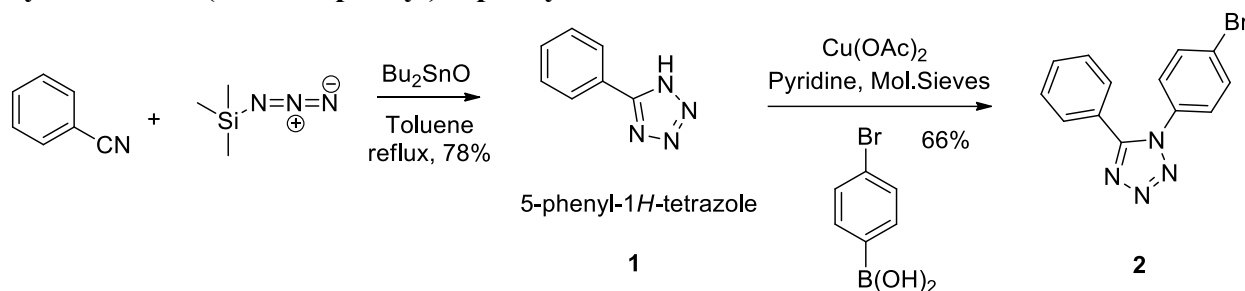
A class of organic heterocyclic compounds rich in nitrogen, tetrazoles have the typical chemical formula $R-CHN_4$. Tetrazoles and their derivatives have garnered a lot of interest because of their usefulness in industry, medicine, and agriculture, among other fields^[1-9]. Drugs patented for the treatment of illnesses of the central nervous system, HIV^[10], sexual dysfunction^[11], asthma^[2], obesity^[3], diabetes^[5], antihypertensive^[12], antiallergic^[13], antibiotics^[14], and anticonvulsants all contain tetrazole ring systems.^[15] Tetrazole derivatives are also used as fungicides, herbicides, and plant growth regulators in agriculture^[8]. They also serve as stabilizers in photoimaging and photography^[9]. These scaffolds have been investigated as explosives, missile propellants, and gas generators in air bags because tetrazole molecules release a lot of energy and lose dinitrogen during decomposition due to their high enthalpy of synthesis^[16].

Herein, we report the facile synthesis of a series of 1,5-diarylated derivatives of these ring systems **4a-d** (Scheme 2) and antimicrobial evaluation in rapidly-growing suspension cultures of L1210 leukaemia cells and slow-growing monolayer cultures of SK-BR-3 mammary tumour cells, using tests of metabolic activity and DNA synthesis in vitro.^[17,18] Since the fraction of Ki-67-positive tumour cells is often correlated with the clinical course of cancer, it was also of interest to determine whether treatments with synthesized tetrazole derivatives would inhibit the expression of human Ki-67 nuclear protein, which is an excellent marker of tumour cell proliferation.^[19]

Although, there are several reports in the literature that discuss the preparation of 1,5-phenyl alkyl substituted tetrazole, the synthesis of the corresponding 1,5-diaryl derivatives has not been directly carried out. We successfully synthesized 1,5-diaryl tetrazole derivatives **4a-d** by the Suzuki coupling of a series of ortho substituted boronic acids **3a-d** ($R = OMe, Cl, I$) with 1-(4-bromophenyl)-5-phenyl-1H-tetrazole **2**²⁰ in the presence of pyridine and molecular sieves (Scheme 2).

RESULTS AND DISCUSSION

Synthesis of 1-(4-Bromophenyl)-5-phenyl-1H-tetrazole 2

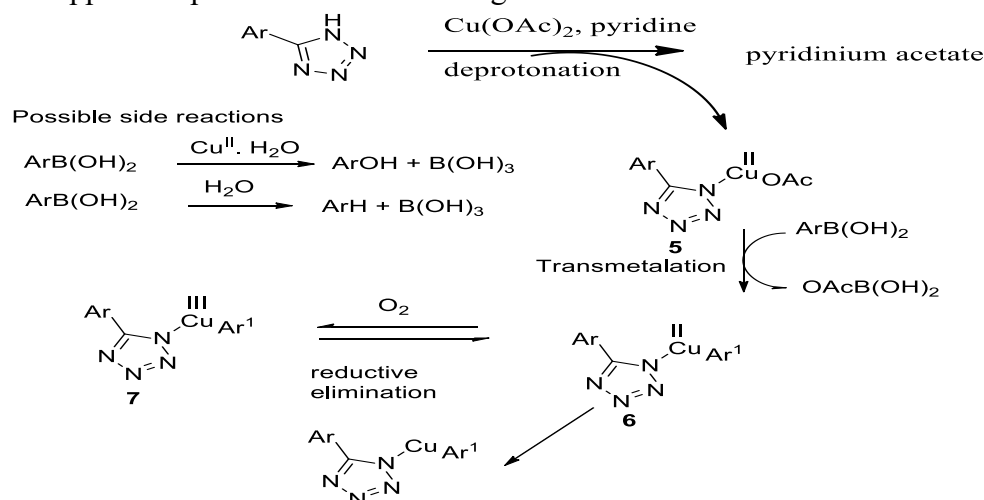


Scheme1. Synthesis of 1-(4-bromophenyl)-5-phenyl-1H-tetrazole

The synthesis of 1-(4-bromophenyl)-5-phenyl-1H-tetrazole **2** was started from Cyano benzene and trimethylsilylazide in the presence of catalytic amount of *t*-butyl tin oxide in toluene to obtain 5-Phenyl-1H-Tetrazole **1** in 78% yield. The N-arylation of the tetrazole **1** was treated with 4-bromophenyl boronic acid and Copper (II) acetate in the presence of molecular sieves to furnish 1-(4-bromophenyl)-5-phenyl-1H-tetrazole **2** in 66% yield. Pyridine was used as a base in the reaction and 3 Å molecular sieves were added to absorb the excess water generated during the course of the reaction (Scheme 1).²¹

Mechanism of Copper catalyzed N-arylation with aryl boronic acid

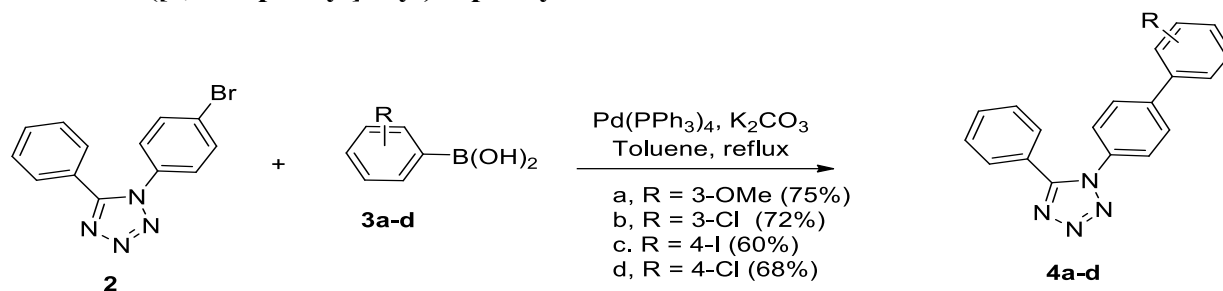
The plausible mechanism for the N-arylation reaction is shown²¹ in Scheme 2. Pyridine assists in the formation of the copper complex **5**. The latter undergoes transmetalation with



Scheme2. Mechanism of N-arylation with phenyl boronic acid

Arylboronic acid to yield Cu (II) complex **6**. The reaction is carried out in the presence of air that oxidizes copper (II) complex **6** to Cu (III) complex **7** which undergoes reductive elimination faster. The excess water formed during the course of the reaction may result in possible side reactions with the arylboronic acids. For this reason, the arylboronic acid is added in excess to the reaction mixture. Molecular sieves are also added to the reaction mixture to absorb the water (Scheme 2).²¹

Synthesis of 1-([1,1'-Biphenyl]-4-yl)-5-phenyl-1*H*-tetrazole derivatives 4a-d



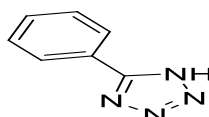
Scheme3. Synthesis of 1-([1,1'-biphenyl]-4-yl)-5-phenyl-1*H*-tetrazole

Additionally, we propose a facile synthesis of novel 1-([1,1'-biphenyl]-4-yl)-5-phenyl-1*H*-tetrazole **4a-d** derivatives in one step. Suzuki coupling of bromo substituted tetrazole **2** with boronic acid (**3a-d**) to afford the desired product (**4a-d**) in moderate to good yields (Scheme 3).

Experimental Section

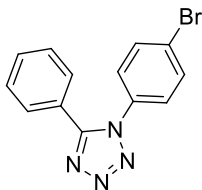
Thin layer chromatography was carried out on 250 μm silica gel plates and UV-light was used as a visualizing agent. Standard column chromatography was performed using 63–200 μm silica gel. Methylene chloride was dried by distillation over calcium hydride. Dry THF was obtained by distillation over sodium and benzophenone. ^1H and ^{13}C NMR spectra were recorded on 400 MHz and 200 MHz spectrometers. The infrared frequencies are reported in cm^{-1} . High resolution mass spectra were acquired on an MDS SCIEX/Applied Bio systems QStar Elite hybrid quadrupole/time-of-flight mass spectrometer (Applied Biosystems, Foster City, CA). In most cases, both $[\text{M} + \text{H}]^+$ and $[\text{M} + \text{Na}]^+$ ions were detectable for each species.

Synthesis of 5-Phenyl-1*H*-tetrazole (1)



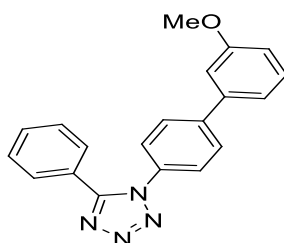
To a stirred solution of Cyano benzene (0.8 g, 7.76 mmol) in toluene (50 mL) under a nitrogen atmosphere, were added azide (TMSA) (1.1 g, 9.32 mmol) and catalytic amount of *t*-butyl tin oxide (30 mg) and the resulting mixture was heated under reflux at 100 $^{\circ}\text{C}$ for 12 h. After completion of the reaction (monitored by TLC), excess TMSA was removed under reduced pressure to afford the crude product which on purification by silica gel column chromatography (EtOAc /hexane = 2:3) furnished the desired product **1** (0.9 g, 79%) as white solid. $[\alpha]_{\text{D}}^{20} = -14.9$ ($c = 3.6$, CHCl_3); IR (neat): $\nu = 3248, 2854, 2743, 2725, 1742, 1539, 1035 \text{ cm}^{-1}$. ^1H NMR (500 MHz, CDCl_3): $\delta = 9.83$ (s, 1H), 8.12 (d, $J = 8.4$ Hz, 1H), 7.78 (d, $J = 8.6$ Hz, 1H), 7.56 (dd, $J = 12.6, 5.3$ Hz, 2H), 7.24 (s, 1H) ppm. ^{13}C NMR (125 MHz, CDCl_3): $\delta = 156.6, 135.6, 132.1, 132.0, 129.7, 128.9, 128.4, 127.9, 120.2, 119.0, 118.9$ ppm. MS (ESI): m/z 147 $[\text{M} + \text{H}]^+$.

Synthesis of 1-(4-Bromophenyl)-5-phenyl-1H-tetrazole (2)



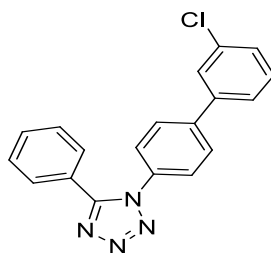
To a stirred suspension of dry copper acetate (0.43 g, 2.4 mmol), 5-Phenyl-1H-tetrazole **1** (0.6 g, 1.2 mmol), phenyl boronic acid (0.52 g, 2.6 mmol) and activated molecular sieves 3A° (0.2 g) in dry methylene chloride (30 mL), was added pyridine (0.23 mL, 2.6 mmol). The reaction mixture was stirred over night at room temperature in air. Upon completion of the reaction, the mixture was diluted with methylene chloride, filtered through celite and washed with aqueous EDTA solution (1.7 mmol). The colorless organic phase was dried over anhydrous Na₂SO₄ and concentrated under reduced pressure. The residue was purified by silica gel column chromatography (EtOAc/hexanes = 1:1) to give 1-(4-Bromophenyl)-5-phenyl-1H-tetrazole **2** (0.8 g, 65%) as a colorless liquid oil. IR (neat): ν = 3439, 2985, 2930, 2854, 1724, 1646, 1027 cm⁻¹. ¹H NMR (500 MHz, CDCl₃): δ = 8.29 (d, *J* = 7.3 Hz, 2H), 8.12 (d, *J* = 8.3 Hz, 2H), 7.78 (d, *J* = 8.5 Hz, 2H), 7.42-7.56 (m, 2H), 7.25-7.35 (m, 2H) ppm. ¹³C NMR (125 MHz, CDCl₃): δ = 165.2, 132.1, 132.0, 130.6, 130.2, 128.9, 128.5, 128.4, 128.2, 127.2, 127.0, 120.2, 119.2 ppm. HRMS (ESI): *m/z* calcd. for C₁₃H₁₀BrN₄ [M + H]⁺: 302.3116, found: 302.3146.

1-(3'-Methoxy-[1,1'-biphenyl]-4-yl)-5-phenyl-1H-tetrazole (4a)



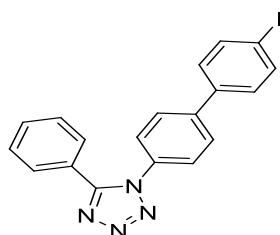
To a stirred solution of **2** (100 mg, 0.0003 mmol), Pd(PPh₃)₄ (0.006 g, 0.009 mmol), K₂CO₃ (0.003 g, 0.01 mmol) in distilled toluene (5 mL) under argon at 40 °C, was added (3-methoxyphenyl)boronic acid (0.05 g, 0.06 mmol) dropwise over a period of 10 min. The reaction mixture was heated at 85 °C until the starting material was completely consumed. The solution was concentrated under reduced pressure. The residue was purified by silica gel column chromatography, yellow solid (75 mg, 75%), mp 119-120 °C. R_f (hexane:ethyl acetate 3:1) 0.3; FTIR (neat): 3860, 3733, 3619, 3002, 2360, 2252, 1440, 1038; ¹H NMR (500 MHz, CDCl₃): δ = 8.31 (d, *J* = 8.3 Hz, 2H), 7.92 (d, *J* = 8.7 Hz, 2H), 7.80 (d, *J* = 8.1 Hz, 2H), 7.42-7.56 (m, 4H), 7.22-7.31 (m, 3H), 3.64 (s, 3H) ppm. ¹³C NMR (125 MHz, CDCl₃): δ = 191.6, 164.2, 137.6, 132.9, 132.8, 132.2, 130.3, 127.6, 121.3, 120.5, 56.9 ppm. HRMS (ESI): *m/z* calcd. for C₂₀H₁₇N₄O [M + H]⁺: 329.3116, found: 329.3146.

1-(3'-Chloro-[1,1'-biphenyl]-4-yl)-5-phenyl-1H-tetrazole (4b)



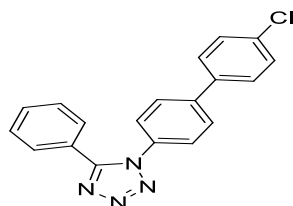
To a stirred solution of **2** (100 mg, 0.0003 mmol), Pd(PPh₃)₄ (0.006 g, 0.009 mmol), K₂CO₃ (0.003 g, 0.01 mmol) in distilled toluene (5 mL) under argon at 40 °C, was added (3-chlorophenyl)boronic acid (0.05 g, 0.06 mmol) dropwise over a period of 10 min. The reaction mixture was heated at 85 °C until the starting material was completely consumed. The solution was concentrated under reduced pressure. The residue was purified by silica gel column chromatography, solid (80 mg, 72%), FTIR (neat): 3860, 3740, 3304, 2906, 2850, 2366, 1043, 984, 753; ¹H NMR (500 MHz, CDCl₃): δ = ¹H NMR (500 MHz, CDCl₃): δ = 8.41 (d, *J* = 7.5 Hz, 2H), 8.16 (d, *J* = 8.6 Hz, 2H), 7.71 (d, *J* = 8.6 Hz, 2H), 7.41-7.56 (m, 4H), 7.15-7.36 (m, 3H) ppm. ¹³C NMR (125 MHz, CDCl₃): δ = 165.2, 132.1, 132.0, 130.6, 130.2, 128.9, 128.5, 128.4, 128.2, 127.2, 127.0, 125.2, 120.2 ppm; HRMS (ESI): *m/z* calcd. for C₁₉H₁₄ClN₄ [M + H]⁺: 333.3016, found: 333.3036.

1-(4'-Iodo-[1,1'-biphenyl]-4-yl)-5-phenyl-1H-tetrazole (4c)



To a stirred solution of **2** (100 mg, 0.0003 mmol), Pd(PPh₃)₄ (0.006 g, 0.009 mmol), K₂CO₃ (0.003 g, 0.01 mmol) in distilled toluene (5 mL) under argon at 40 °C, was added (4-iodophenyl)boronic acid (0.05 g, 0.06 mmol) dropwise over a period of 10 min. The reaction mixture was heated at 85 °C until the starting material was completely consumed. The solution was concentrated in vacuo and reaction mixture was purified by silica gel column chromatography. Pale yellow solid (85 mg, 60%), mp 119-120 °C. R_f (hexane:ethyl acetate 4:1) 0.3; FTIR (neat): 3164, 3003, 2944, 2292, 2252, 1439, 1375, 1303, 984, 753; ¹H NMR (500 MHz, CDCl₃): δ = 8.29 (d, *J* = 7.3 Hz, 2H), 8.14 (d, *J* = 8.5 Hz, 2H), 7.68 (d, *J* = 8.6 Hz, 2H), 7.41-7.56 (m, 4H), 7.15-7.35 (m, 3H) ppm. ¹³C NMR (125 MHz, CDCl₃): δ = 191.6, 132.7, 132.1, 131.9, 131.1, 130.2, 129.3, 129.1, 128.6, 128.4, 128.1, 126.4, 117.4, 77.2 ppm; HRMS (ESI): *m/z* calcd. for C₁₉H₁₄IN₄ [M + H]⁺: 425.3216, found: 425.3126.

1-(4'-Chloro-[1, 1'-biphenyl]-4-yl)-5-phenyl-1H-tetrazole (4d)



To a stirred solution of **2** (100 mg, 0.3 mmol), Pd(PPh₃)₄ (0.006 g, 0.009 mmol), K₂CO₃ (0.003 g, 0.01 mmol) in distilled toluene (5 mL) under argon at 40 °C, was added (4-chlorophenyl)boronic acid (0.05 g, 0.6 mmol) dropwise over a period of 10 min. The reaction mixture was heated at 85 °C until the starting material was completely consumed. The solution was concentrated in vacuo and reaction mixture was purified by silica gel column chromatography, yellow solid (75 mg, 68%), mp 119-120 °C. R_f (hexane:ethyl acetate 2:1) 0.3; FTIR (neat): 3741, 2360, 1439, 1366, 1320, 1039, 1071, 1043, 984, 753; ¹H NMR (500 MHz, CDCl₃): δ = 8.29 (d, *J* = 7.3 Hz, 2H), 8.14 (d, *J* = 8.5 Hz, 2H), 7.68 (d, *J* = 8.6 Hz, 2H), 7.41-7.56 (m, 4H), 7.15-7.35 (m, 3H) ppm. ¹³C NMR (125 MHz, CDCl₃): δ = 191.6, 164.2, 137.6, 132.9, 132.8, 132.2, 130.3, 127.6, 121.3, 120.5 ppm: HRMS (ESI): *m/z* calcd. for C₁₉H₁₄ClN₄ [M + H]⁺: 333.2016, found: 333.2146.

CONCLUSION

In conclusion, we synthesised of the title compounds 1-([1,1'-biphenyl]-4-yl)-5-phenyl-1H-tetrazole **4a-d** with good yield. The compound 1-([1,1'-biphenyl]-4-yl)-5-phenyl-1H-tetrazole **4a** has obtained is an excellent precursor to introduce other type of substituents on tetrazole through metalation followed by electrophilic quenching, or *via* palladium catalyzed Suzuki coupling reactions. The above reactions also demonstrate the stability of the tetrazole ring to diverse reaction conditions.

REFERENCES:

- Andres-Gil, J. I., Alcazar-Vaca, M. J., Pastor-Fernandez, J., Drinkenburg, W. H. I. M., Langlois, X. J. M., Oyarzabal-Santamarina, J. and Vega-Ramiro, J. A. Preparation of triazolone, tetrazolone and imidazolone derivatives for use as alpha 2C-adrenoreceptor antagonists, EP569512006067139, 2006.
- Goulet, M., Ashton, W. T., Chu, L., Fisher, M. H., Lin, P., Ponpipom, M. M., Wyvratt, M. J., Girotra, N. N. and Young, J. Preparation of indoles as antagonists of gonadotropin releasing hormone, 1154976200957, 2001.
- Goulet, M., Walsh, T. F., Ujjainwalla, F. and Wyvratt, M. J., Jr. Preparation of indole-5-alkanamides and analogs as gonadotropin releasing hormone antagonists, 3121926156772, 2000.
- Janssens, F., Torremans, J., Janssen, M., Stokbroekx, R. A., Luyckx, M. and Janssen, P. A. New antihistaminic N-heterocyclic 4-piperidinamines. 3. Synthesis and antihistaminic activity of N-(4-piperidinyl)-3H-imidazo[4,5-b]pyridin-2-amines, J. Med. Chem., 1985, 28, 1943–1947.
- Chartrain, M. M., Chung, J. Y. L. and Roberge, C. Antidiabetic antiobesity agent intermediate manuf. with Candida, US126159803672, 1998.
- Foldenyi, R. New 1H-tetrazol-5-thiol derivatives as pesticides. 2. Cyanodithiomidocarbonic acid-esters (Short commun), Monatshefte Für Chemie, 1995, 126, 1035–1043.

7. Nishi, Y. and Miyauchi, H. New formulation and application method with fentrazamide in transplanted rice, *Pflanzenschutz-Nachr. Bayer (Engl. Ed.)*, 2001, 54, 43–50.
8. Theodoridis, G. Herbicidal 1-aryl-4-substituted-1,4-dihydro-5H-tetrazol-5-ones and sulfuranalogs thereof, US17998501939, 1985.
9. Ishikawa, W., Fujiwhara, M., Kojima, T., Endo, T. and Kato, K. Color image production, 29020742902074, 1979.
10. Abell, A. D. and Foulds, G. J. Synthesis of a cis-conformationally restricted peptide bond isostere and its application to the inhibition of the HIV-1 protease, *J. Chem. Soc., Perkin Trans. 1*, 1997, 17, 2475–2482.
11. Shaw-Reid, C. A., Miller, M. D., Hazuda, D. J., Ferrer, M., Sur, S. M., Summa, V., Lyle, T. A., Kinzel, O., Pescatore, G., Muraglia, E., Orvieto, F. and Williams, P. D. Preparation of tetrazolyl derivatives as HIV reverse transcriptase inhibitors, US166712005115147, 2005.
12. Mavromoustakos, T., Kolocouris, A., Zervou, M., Roumelioti, P., Matsoukas, J. and Weisemann, R. An effort to understand the molecular basis of hypertension through the study of conformational analysis of losartan and sarmesin using a combination of nuclear magnetic resonance spectroscopy and theoretical calculations, *J. Med. Chem.*, 1999, 42, 1714–1722.
13. Toney, J. H., Fitzgerald, P. M., Grover-Sharma, N., Olson, S. H., May, W. J., Sundelof, J. G., Vanderwall, D. E., Cleary, K. A., Grant, S. K., Wu, J. K., Kozarich, J. W., Pompliano, D. L. and Hammond, G. G. Antibiotic sensitization using biphenyl tetrazoles as potent inhibitors of *Bacteroides fragilis* metallo-beta-lactamase, *Chem. Biol.*, 1998, 5, 185–196.
14. Hashimoto, Y., Ohashi, R., Kurosawa, Y., Minami, K., Kaji, H., Hayashida, K., Narita, H. and Murata, S. Pharmacologic profile of TA-606, a novel angiotensin II-receptor antagonist in the rat, *J. Cardiovasc. Pharmacol.*, 1998, 31, 568–575.
15. DeSarro, G. B., Zappala, M., Grasso, S., Chimirri, A. and DeSarro, A. Molecular requirement for anticonvulsant activity in a series of thiazolo-1,4-benzodiazepine derivatives and comparison with classical benzodiazepines, *Gen. Pharmacol.*, 1993, 24, 877–884.
16. Burns, S. P., Moquin, L. A. and Khandhadia, P. S. Selective non-catalytic reduction of toxic gaseous effluents, 6386067575648, 2009.
17. Araujo, N. C. P., Barroca, P. M. M., Bickley, J. F., Brigas, A. F., Cristiano, M. L. S., Johnstone, R. A. W. and Loureiro, R. M. S. Structural effects on sigmatropic shifts in heteroaromatic allyl ethers, *J. Chem. Soc., Perkin Trans. 1*, 2002, 9, 1213–1219.
18. Cristiano, M. L. S., Johnstone, R. A. W. and Price, P. J. Metal-assisted reactions. Part 25. Heterogeneous and homogeneous catalytic transfer hydrogenolysis of allyloxytetrazoles to yield alkenes or alkanes, *J. Chem. Soc., Perkin Trans. 1*, 1996, 12, 1453–1459.
19. Araujo, N. C. P., Brigas, A. F., Cristiano, M. L. S., Frija, L. M. T. and Guimaraes, E. M. O. Heteroaromatic benzyl ethers as intermediates for palladium-catalysed transfer hydrogenolysis of benzyl alcohols, *J. Mol. Catal. A: Chem.*, 2004, 215, 113–120.
20. Rayat, S., Chhabra, R., Alawode, O. and Gundugola, A. S. *J. Mol. Struct.*, 2009, 38, 933.
21. Ley, S. V. and Thomas, A. W. Modern synthetic methods for copper-mediated C(aryl)-O, C(aryl)-N, and C(aryl)-S bond formation, *Angew. Chem. Int. Ed.*, 2003, 42, 5400–5449.