Rajaram Arulmozhi et al /J. Pharm. Sci. & Res. Vol. 11(5), 2019, 1974-1978

ISSN:0975-1459
Journal of Pharmaceutical Sciences and Research
www.jpsr.pharmainfo.in

# Synthesis, characterization and anticancer activity of some new Tetrazoles derived from Quinazolin-4-one

**Rajaram Arulmozhi[1], Natarajan Abirami[1,*], Helen P. Kavitha[2], Subramanian Arulmurugan[2]**

*[1]Department of Chemistry, Faculty of Engineering and Technology, SRMIST, Kattankulathur, Chennai, Tamil Nadu, India*

*[2]Department of Chemistry, Faculty of Engineering and Technology, SRMIST, Ramapuram, Chennai, Tamil Nadu, India*

**Abstract**

The present study was aimed in the synthesis of novel Tetrazoles from 3-[(4-(4-Amino-benzenesulfonyl)-phenyl]-2-phenyl-quinazolin-4(3H)-one.N-{4-[4-(4-Oxo-2-Phenyl-4H-Quinazolin-3-yl)-Benzenesulfonyl]-Phenyl}-Benzamide (2a-i) and 2-Phenyl-3-{4-[4-(5-Phenyl-Tetrazol-2-yl)-Benzenesulfonyl]-Phenyl}-Quinazolin-4(3H)-one (3a-i) were prepared in accordance with the proposed scheme**.** The compounds were characterized by IR, 1H NMR, $^{13}$C NMR, Mass and elemental analysis. Some of the Tetrazole derivatives 3a, 3b and 3g were tested for anticancer activity against MTT assay method using 5-Flurourocil as the standard drug. The data reveals that all the three compounds 3a, 3b and 3g taken for the study show anti-proliferative activity against the two tested cancer cell lines (MCF-7 and HT-29). Among them, the compound **3g** shows better anti-proliferative activity than the rest of the compounds with the $IC_{50}$ value of 62µg/ml for HT-29. However, the activity of the test compounds is less than the standard drug 5-Flurourocil. Cytotoxic effects of compound **3g** to MCF-7 reveal that morphological changes includes decrease of cell density, cell rounding and floating cells under inverted phase contrast microscope. A new class of tetrazolo quinazoline heterocycles were synthesized and characterized. The representative compounds were screened for anticancer by MTT assay method using 5-Flurourocil as the standard drug. The results of anticancer activity revealed, the compound **3g** is found to have potent anti-cancer activity.

**Keywords**: Anticancer activity, MTT assay, Quinazolinone, Tetrazoles

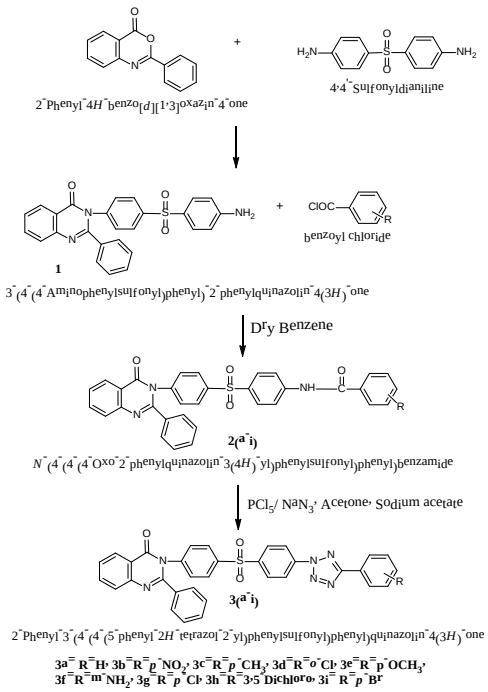
## INTRODUCTION

Tetrazole cycle is a promising pharmacophore fragment frequently used in the development of novel drug [1] development of the Tetrazole chemistry has been largely associated with ring flexibility, stability which provides easily to different binding modes and toxicity decreasing properties [2]. This Tetrazole is used in MTT assay to quantify the respiratory activity of live cells in cell culture, although it kills cells in the process [3]. Interest in Tetrazole chemistry over the past few years has been increasing rapidly because of its wide range of applications, mainly as a result of the role played by this heterocyclic functionality in medicinal chemistry as these offer a more favorable pharmacokinetic profile and a metabolically stable surrogate for carboxylic acid functionalities [4]. The ability of Tetrazole compounds to mimic the carboxylic functionality has motivated the incorporation of Tetrazole derivatives into biologically active molecules. This has led to applications in therapy resulting in compounds with antibacterial [5] antifungal [6], antiviral [7], analgesic [8,9], anti-inflammatory [10-12] antiulcer [13] and anti-hypertension activities [14].

Quinazolines are nitrogen containing compounds having broad spectrum of medicinal values, such as antifungal [15], anticancer [16], anti-HIV [17], anti-inflammatory [18], analgesic [19], antiviral [20], antimicrobial [21], anticonvulsant [22], anticoagulant [23] , anti fibrillatory [24], cardiac stimulant [25],and diureric [26] etc. In view of the literature evidence, a scheme was proposed combining the moieties of Tetrazole and Quinazolone, with a view to synthesize new compounds with enhanced activities.

## MATERIALS AND METHODS

The proposed scheme for the preparation of Quinazoline and its derivatives is given in the scheme.




**Scheme:** Preparation of Quinazoline and its derivatives

### Cyto-toxic studies

### MTT Assay

The anticancer activity of the compounds, such as 2-Phenyl-3-(4-(4-(5-phenyl-2H-tetrazol-2-yl)phenylsulfonyl)phenyl) quinazoline-4(3H)-one **(3a)**, 2-Phenyl- 3-(4-(4-(5-(4-Nitrophenyl)-2H-tetrazol-2-yl)phenylsulfonyl)phenyl)-2-phenylquinazolin-4(3H)-one **(3b)**, 2-Phenyl-3-{4-[4-(5-(4-chloro)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one **(3g)** was

carried out by MTT Assay in two different human cell lines such as, HT-29(Colon cancer) and MCF-7(Breast cancer).
Human colon cancer HT-29 and human breast cancer MCF-7 cell lines were obtained from National Centre for Cell Sciences (NCCS), Pune. The cells were maintained in RPMI-1640 supplemented with 10% FBS, penicillin (100 U/ml) and Streptomycin (100 µg/ ml) in a humidified atmosphere of 50 µg/ml $CO_2$ at 37 °C.
Cells (1 × 105/well) were plated in 100 µl of medium/well in 96-well plates. After 48 hours of incubation, the cell reaches the confluence. The cells were then incubated in the presence of various concentrations of the samples in 0.1% DMSO for 48 h at 37°C and 5% $CO_2$. After the removal of the sample solution and washing with phosphate-buffered saline (pH 7.4), 20µl/well (5mg/ml) of 0.5% 3-(4, 5-dimethyl-2-thiazolyl)-2,5-diphenyl-tetrazolium bromide cells (MTT) phosphate-buffered saline solution was added. After 4h incubation, 0.04M HCl/ isopropanol were added. Viable cells were determined by the absorbance at 570nm with reference at 655nm. Measurements were performed three times and the concentration required for 50% inhibition of viability ($IC_{50}$) was determined graphically. The absorbance at 570 nm was measured with a microplate reader using wells without sample containing cells as blanks. All experiments were performed in triplicate. The effect of the samples on the proliferation of cancer cells was expressed as the % cell viability using the following formula:
% Cell Viability = A570 of Treated Cells / A570 of Control Cells × 100% (1)

## RESULTS AND DISCUSSION

### Characterization studies for synthesized compounds

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3a)**

Pale green powder, yield 62 %; mp 291 – 294 $^oC$; IR(KBr): 1403.63 (N = N), 1643.86( C = N), 1255.50( N – N =N), 1073.94 &1020.42 ( Tetrazole ring),; $^1H$ NMR (DMSO D6) δ in ppm 7.55-8.13(m, 22H, Ar-H); $^{13}C$ NMR: 165.42, 164.31, 152.52, 143.21, 138.42, 133.44, 132.9, 134.1, 130.02, 129.0, 128.6, 127.4, 127.0, 126.7,126.0, 122.15, 121.6; EI-MS, m/z(%): 582(100); Analytical calculation for $C_{33}$ $H_{22}N_6O_3S$ (582) C-68.03; H- 3.81; N-14.42; Found: C-68.12; H-3.84; N-14.76

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-(4-nitro)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3b)**.

Pale yellow powder, yield 69 %; mp 281-283 $^oC$; IR(KBr): 1407.72(N = N ), 1590.87 (C =N), 1183.26(N – N = N), 1105.26 & 1070.71 (Tetrazole ring); $^1H$ NMR (DMSO D6) δ ppm 8.36 (d, 2H, J= 2.5, H19, 21), 7.75-8.25 (m, 19H, Ar-H) ; $^{13}C$ NMR: 175.04, 165.19, 161.67, 154.82, 153.20, 149.76, 147.62, 140.61, 135.50, 131.59, 131.50, 131.17, 129.99, 128.98, 128.77, 128.55, 127.96, 127.86, 127.32, 126.90, 123.98, 121.08, 121.01; EI-MS, m/z(%): 628(15); Analytical calculation for $C_{33}$ $H_{21}N_7O_5S$ (628) C-63.15; H-3.37;N- 15.62; Found:C-63.12; H- 3.32; N- 15.64

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-(4-methyl)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3c)**

Pale brown powder, yield 71 %; mp182-184 $^oC$; IR (KBr): 1666.59 (C = N), 1399.32 (N = N), 1259.32 (N – N = N), 1108.96 & 1018.98 (Tetrazole ring),; $^1H$ NMR (DMSO D6) 7.49-7.80(m, 13H, Ar-H), 2.3 (s, 1H, CH3), 7.88 (d, J= 7, 2H, H18,22 Phenyl), 7.36 (d, J= 5.5, 2H, H19, 20, phenyl), 8.06 (d, 1H, J= 7, H1), 7.34(t, 1H, J= 5.5, H2 Q) 7.21(t, 1H, J=8, H3), 7.84 (d, 1H, J= 7, H4); $^{13}C$ NMR : 166.37, 161.61, 155.00, 147.48, 134.82, 141.71, 142.63, 135.55, 133.20, 129.43, 131.86, 128.98, 128.70, 128.43, 126.96, 121.14, 20.9; EI-MS, m/z(%): 596 (6); Analytical calculation for $C_{34}H_{24}N_6O_3S$ (597) C-68.44; H- 4.05;N-14.09; Found:C-68.47; H- 4.01; N- 14.11

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-(2-chloro)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3d)**

Pale brown powder, yield 68 %; mp198-200 °C; IR (KBr): 1678.26 (C = N), 1469.68 (N = N), 1274.48 (N – N = N), 1073.06 & 1020.06 (Tetrazole ring),; $^1H$ NMR (DMSO D6) 7.45-7.67(m, 4H, Quinazoline), 7.86-8.0(m, 13H, Ar-H), 7.34(t, 1H, J= 8, H19 ), 7.81(t, 1H, J=8, H20), 8.19(d, 1H, J= 8, H21), 8.22(d, 1H, J=8, H18); $^{13}C$ NMR : 166.02, 165.97, 154.92, 147.46, 140.53, 136.74, 136.35, 135.53, 135.39, 135.32, 131.97, 131.58, 131.50, 130.38, 130.19, 129.80, 129.69, 129.46, 129.41; EI-MS, m/z(%): 618(5); Analytical calculation for $C_{33}$ $H_{21}ClN_6O_3S$ (618) C-64.23; H- 3.43;N- 13.62; Found:C-64.20; H- 3.45; N- 13.64

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-(4-methoxy)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3e)**

Pale yellow powder, yield 69 %; mp149 – 150 °C; IR (KBr): 1662.81 (C = N), 1466.74 (N = N), 1251.86 (N – N = N) 1106.48 & 1022.02 (Tetrazole ring),; $^1H$ NMR (DMSO D6): 3.8 (s, 3H, -OCH3), 7.62 (d, 2H, J= 8.5, H19, 20), 8.19 (d, 2H, J=8, H18, 21) 7.76-8.07 (m, 17H, Ar-H); $^{13}C$ NMR: 165.95, 165.88, 162.77, 162.74, 147.36, 144.63, 141.70, 135.55, 135.29, 134.67, 130.35, 129.65, 129.42, 129.27, 128.97, 128.131, 128.043, 127.91, 127.76, 126.951, 121.010, 120.592, 120.526, 114.15, 113.93, 55.95; EI-MS, m/z(%): 613 (5); Analytical calculation for $C_{34}$ $H_{24}N_6O_4S$ (613) C- 66.65; H- 3.95;N- 13.72; Found: C-66.62; H-3.91; N- 13.69

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-(3-amino)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3f)**

Pale green powder, yield 65 %; mp 159 – 161 °C; IR (KBr): 1627.81 (C = N), 1403.31 (N = N), 1281.41 (N – N = N), 1104.46 & 1072.02 ( Tetrazole ring), ; $^1H$ NMR (DMSO D6): 3.44 (s, 2H, NH2) 6.23 (s, 1H, H18) 7.40-7.60 (m, 20H, Ar-H); $^{13}C$ NMR: 165.16, 153.84, 153.19, 131.20, 130.12, 130.05, 129.97, 129.68, 129.62, 129.40, 129.26, 128.89, 128.62, 128.11, 128.01, 127.93, 127.38, 120.82, 120.21, 119.89, 119.57, 113.46, 113.32, 113.04; EI-MS, m/z(%): 598 (13 Analytical calculation for $C_{33}$ $H_{23}N_7O_3S$ (598) C- 66.32; H- 3.88;N- 16.41; Found:C-66.30; H- 3.85; N- 16.43

**Spectral characterization of 2-Phenyl-3-{4-[4-(5-(4-chloro)phenyltetrazol-2-yl)-benzenesulfonyl]-phenyl}-quinazolin-4(3H)-one (3g)**.

Pale brown powder, yield 68 %; mp 103 – 105 °C; IR (KBr): 1642.10 (C = N ), 1414.26 (N = N), 1156.16 (N – N = N), 1018.42 & 1098.14 (Tetrazole ring ),781.30(C-Cl), ; $^{1}$H NMR (DMSO D6): 7.44 (d, 2H, J=10, H19, 20), 7.54 (d. 2H, J=8.5, H18, 21), 7.9 (d, 1H, J= 12, H1), 7.20 (t, 1H, J=8, H2), 7.55 (t, 1H, J=8.5, H3), 7.75 (d, 1H, J=8, H4), 7.22-7.36 (m, 13H, Ar); $^{13}$C NMR: 165.53,164.20, 153.62, 152.34, 143.46, 139.62, 138.42, 134.12, 133.70, 130.26, 129.9, 128.6, 127.23, 126.72, 126.05, 122.27, 121.42; EI-MS, m/z(%): 617 (5); Analytical calculation for $C_{33}H_{21}ClN_6O_3S$ (618) C-64.23; H-3.43;N-13.62; Found: C-64.20; H- 3.45; N-13.68.

In the IR spectra of the compounds analyzed, it is revealed that the absorption bands in the region 1590-1678 $cm^{-1}$ (C=N), 1399-1466 $cm^{-1}$ (N=N), 1156-1281 $cm^{-1}$ (N-N=N) and 1110-1020 $cm^{-1}$ (tetrazole ring) revealing the presence of tetrazole ring in the synthesized compounds. The $SO_2$ (symmetric) and $SO_2$ (asymmetric) stretching vibrations are observed at 1353-1315 $cm^{-1}$ and 1143-1156 $cm^{-1}$ respectively. The $^{1}$H NMR and $^{13}$C NMR spectra also show convincing evidence regarding the structure of the synthesized compounds. In the $^{1}$H NMR spectra, the aromatic protons appear as a multiplet in the regions δ 7.15 to 8.04.The appearance of singlet peaks at δ 2.3, δ 3.8 and δ 3.44 confirms the presence of $CH_3$, $OCH_3$ and $NH_2$ groups in the Tetrazole derivatives 3c, 3e & 3f respectively. In the $^{13}$C NMR spectra, the appearance of signals in the region 121.0-153.7 may be attributed to the quinazolinone carbons. Signals at 121.5-135.6 may be assigned to the phenyl carbons of the compounds. The mass spectrum of all the nine tetrazole derivatives shows molecular ion peaks at their respective molecular weights which further lend support to the assigned structures.

**MTT assay**

The results of the cytotoxic activity of the representative compounds **3a, 3b**, and **3g** against human cancer cell lines such as, HT-29 (Colon Cancer) and MCF-7(Breast Cancer) are presented(Table 1 and 2) along with their graphs (Figure 1 and 2). The $IC_{50}$ values are also given (Table 3). The data from the tables reveals that all the three compounds taken for the study show anti proliferative activity against the two tested cancer cell lines. Among the three tested compounds, the compound **3g** shows better anti proliferative activity than the rest of the compounds with the $IC_{50}$ value of 62µg/ml for HT-29. However, the activity of the test compounds is less than the standard drug 5-Flurourocil. Cytotoxic effects of compound **3g** to MCF-7 (Figure 3) reveal that morphological changes includes decrease of cell density, cell rounding and floating cells under inverted phase contrast microscope.

**Table 1:** Cell viability of compounds 3a, 3b and 3g against HT-29 colon cancer cell line

| S.No | C (µg/ml) | Dilutions | 3a | | 3b | | 3g | | 5-Flurourocil [5Fu] | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | Abs (O.D) | Cell viability (%) | Abs (O.D) | Cell viability (%) | Abs (O.D) | Cell viability (%) | Abs (O.D) | Cell viability (%) |
| 1 | 1000 | Neat | 0.07 | 14.58 | 0.04 | 8.33 | 0.05 | 10.41 | 0.04 | 12.35 |
| 2 | 500 | 1:1 | 0.13 | 27.08 | 0.10 | 20.83 | 0.14 | 29.16 | 0.11 | 17.30 |
| 3 | 250 | 1:2 | 0.18 | 37.50 | 0.17 | 35.41 | 0.19 | 39.58 | 0.15 | 21.05 |
| 4 | 125 | 1:4 | 0.23 | 47.91 | 0.21 | 43.75 | 0.25 | 52.08 | 0.24 | 24.17 |
| 5 | 62.5 | 1:8 | 0.31 | 64.58 | 0.27 | 56.25 | 0.31 | 64.58 | 0.28 | 29.00 |
| 6 | 31.2 | 1:16 | 0.38 | 79.16 | 0.33 | 62.50 | 0.36 | 75.00 | 0.34 | 37.25 |
| 7 | 15.6 | 1:32 | 0.43 | 89.58 | 0.38 | 79.16 | 0.40 | 83.33 | 0.37 | 43.46 |
| 8 | 7.8 | 1:64 | 0.46 | 95.83 | 0.43 | 89.58 | 0.45 | 93.75 | 0.44 | 48.36 |
| 9 | Cell control | - | 0.48 | 100 | 0.48 | 100 | 0.48 | 100 | 0.48 | 100 |

C- Concentration; Abs-Absorbance

**Table 2**: Cell viability of compounds 3a, 3b and 3g against MCF-7 breast cancer cell line

| S.No | C (µg/ml) | Dilutions | 3a | | 3b | | 3g | | 5-Flurourocil | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | Abs (O.D) | Cell viability (%) | Abs (O.D) | Cell viability (%) | Abs (O.D) | Cell viability (%) | Abs (O.D) | Cell viability (%) |
| 1 | 1000 | Neat | 0.08 | 14.81 | 0.03 | 5.55 | 0.04 | 7.04 | 0.04 | 12.35 |
| 2 | 500 | 1:1 | 0.15 | 27.77 | 0.09 | 16.66 | 0.11 | 20.37 | 0.11 | 17.30 |
| 3 | 250 | 1:2 | 0.20 | 37.03 | 0.13 | 24.07 | 0.17 | 31.48 | 0.15 | 21.05 |
| 4 | 125 | 1:4 | 0.26 | **48.14** | 0.19 | 35.18 | 0.24 | **50.00** | 0.24 | 24.17 |
| 5 | 62.5 | 1:8 | 0.30 | 55.55 | 0.24 | 44.44 | 0.34 | 62.96 | 0.28 | 29.00 |
| 6 | 31.2 | 1:16 | 0.37 | 68.51 | 0.28 | 51.85 | 0.42 | 77.77 | 0.34 | 37.25 |
| 7 | 15.6 | 1:32 | 0.42 | 77.77 | 0.34 | 62.96 | 0.47 | 87.03 | 0.37 | 43.46 |
| 8 | 7.8 | 1:64 | 0.46 | 85.18 | 0.39 | 72.22 | 0.51 | 94.44 | 0.44 | 48.36 |
| 9 | Cell control | - | 0.48 | 100 | 0.48 | 100 | 0.48 | 100 | 0.48 | 100 |

C- Concentration; Abs-Absorbance

**Table 3**: $IC_{50}$ Values of the test compounds

| S.No | Samples | IC $_{50}$ Concentration (µg/mL) | |
|---|---|---|---|
| | | HT-29 | MCF-7 |
| 1. | 3a | 116 | 110 |
| 2. | 3b | 92 | 39 |
| 3. | 3g | 146 | 126 |
| 4. | 5Fu | 5.75 | 5.5 |

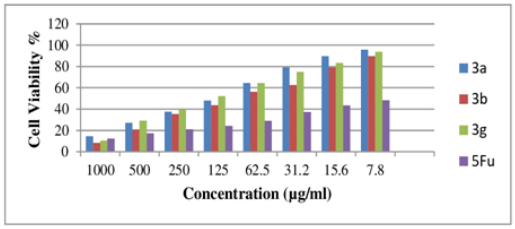



**Figure 1**. Cell viability of compounds 3a, 3b and 3g against HT-29 colon cell lines

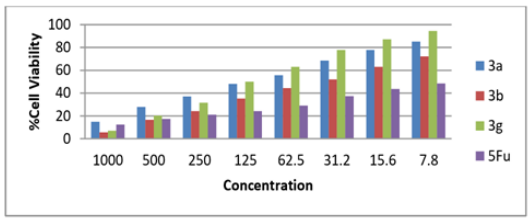



**Figure 2**. Cell viability of compounds 3a, 3b and 3g against MCF-7 breast cell lines

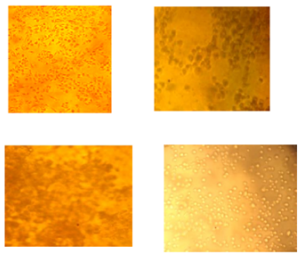


**Figure 3**. Cytotoxic effects of 3g to MCF-7 (breast cancer cell line) (morphological changes including decrease of cell density, cell rounding and floating cells were observed with an inverted phase contrast microscope and photographed).

## CONCLUSIONS

A new class of tetrazolo quinazoline heterocycles were synthesized and characterized. The representative compounds were screened for anticancer by MTT assay method using 5-Flurourocil as the standard drug. The results of anticancer activity revealed that the compounds taken for the study possess significant activity. Among the tested compounds, the compound **3g** is found to have potent activity.

It is convincing that this class of compounds certainly holds great promise towards the pursuit to discover novel classes of anticancer agents. Further biological studies are in progress to know the potency of the compounds.

## ACKNOWLEDGEMENTS

We acknowledge DST-FIST (fund for improvement of S&T infrastructure) for financial assistance for Department of Chemistry, SRM University of Science & Technology, No.SR/FST/CST-266/2015(c).

## CONFLICT OF INTERESTS

The authors declare that they have no conflicts of interest.

## REFERENCES

1. Popova Elena EA, Protas V, TrifonovRostislav R E. Tetrazole Derivatives as Promising-Anticancer Agent, Anti-cancer agents in medicinal chemistry. 2017; 17: 999.
2. Kaplancikli ZA, Yurttaş L, Özdemir A, Zitouni GT, İşcan G, Akalın G. Journal of enzyme inhibition and medicinal chemistry. 2014; 29.
3. Mohite PB, Bhaskar VH. Potential Pharmacological Activities of Tetrazoles in The New Millennium International Journal of PharmTech Research. 2011; 3: 1557-1566.
4. Myznikov LV, Hrabalek A, Koldobskii G. Drugs in the tetrazole series,Chem.Heterocycl. Compound. 2007; 43: 1.
5. Genin MJ, Alwine DA, Anderson DJ, Barbachyn MR, Emmert DE, Graber SA, Grega KC, Hester JB, Hutchinson DK, Morris J, Reischer RJ, Ford CW, Zurenko GE, Hemel JG, Schchaatt RD, Stapert D, Yagi BH. Substituent effects on the antibacterial activity of nitrogen-carbon-linked (azolylphenyl)oxazolidinones with expanded activity against the fastidious gram-negative organisms Haemophilus influenzae and Moraxella catarrhalis. J. Med. Chem. 2000;43: 953-970.
6. Rex JH, Rianaldi MG, Pfallar MA. Antimicrob.Agents. Chemother. 1995; 39: 1.
7. Evstopov AN, Yavorovskaya VE, Vorobev ES, Kudonogova ZP, Gritsenko LN, Schmidt EN, Medevedeva SG, Filimonov VD, Prishchep TP, Saratikov AS. Synthesis and antiviral activity of antipyrine derivatives. Pharm. Chem. J; 1992:26, 426-429.
8. Bachar SC, Lahiri SC. Synthesis of chloro and bromo substituted 5-(indan-1′-yl)tetrazoles and 5-(indan-1′-yl)methyltetrazoles as possible analgesic agents. Pharmazie.2004; 59: 435.
9. Stewart KD. Discovery of a new cyclooxygenase-2 lead compound through 3-D database searching and combinatorial chemistry Bioorg. Med. Chem. Lett. 1998; 529.
10. Aliasaghar H, Khalili D, Clercq ED, Salmi C, Brunel JM. Synthesis, Antibacterial, Antifungal and Antiviral Activity Evaluation of Some New bis-Schiff Bases of Isatin and their Derivatives. Molecules. 2007; 12: 1720.
11. Mohite PB, Pandhare RB, Khanage SG, Bhaskar VH. Synthesis and Anti-Inflammatory Activity of Some 5-Phenyl-1-(Acyl)-1, 2, 3, 4-Tetrazole. Journal of Pharmacy Research. 2010;3: 1.
12. Natrajan U, Kaliappan I, Singh NK. A facile design and efficient synthesis of schiff's bases of tetrazolo [1,5-a] quinoxalines as potential anti-inflammatory and anti-microbial agents Der PharmaChemica. 2010; 21: 159-167.
13. Hallinan EA, Tsymbalov S, Dorn CR, Pitzele BS, Jr. Hansen DW Synthesis and Biological Characterization of L-$N^6$-(1-Iminoethyl)lysine 5-Tetrazole-amide, a Prodrug of a Selective iNOS Inhibitor.. J. Med. Chem. 2002; 45: 1686-1689.
14. Upadhayaya RS, Sinha N, Jain S, Kishore N, Chandra R, Arora SK. Synthesis and Antifungal Activity of (2R,3S)-2-(2,4-Difluorophenyl)-3-(5-{2-[4-aryl-piperazin-1-yl]-ethyl}-tetrazol-2-yl/1-yl)-[1,2,4]-triazol-1-yl-butan-2-ol Bioorg. Med. Chem. 2004; 12: 2225-2238.
15. Ouyang P, Zhang G, Xu P, Song S, Yang L, Jin W, Xue D, Hu P, Lu Chen Z. Synthesis and Antifungal Bioactivities of 3-Alkylquinazolin- 4-one Derivatives,Molecules. 2006; 11: 383-392.
16. Desai N C, Shah BR, Bhatt JJ, Patel H, Undavia NK, Trived BB. Synthesis of 2, 3- disubstituted-3, 1- quinazolin-4(4H)ones as potential anticancer and anti-HIV agents Indian J. Chem. 1995; 34: 201.
17. Desai NC, Undavia LK, Trivedi PB, Dave D, Vyas GD. Synthesized and screened anti-HIV activity of some non-nucleoside 2,3-disubstituted quinazoline derivatives Indian J.Exp.Biol. 1998;, 36:1280.

18. Alagarsamy V, Solomon VR, Murugesan S. Synthesis and Pharmacological Evaluation of Some 3-(2-Methylphenyl)-2-substituted amino-quinazolin-4(3*H*)-ones as Analgesic and Antiinflammatory Agents. Arzneimittelforsch. 2008;58: 174-181.
19. Rohit D Patel, Manish P Patel and Ranjan G Patel, Synthesis, characterization and anti-bacterial activity of some new 2, 3, 6-trisubstituted quinazolin-4(3*H*)-ones, Indian Journal of Chemistry-B, 44B, 2005, 1944-1946.
20. Pandey VK, Tusi S, Tusi Z, Raghubir R, Dixit M, Joshi MN. Heterocyclic Compounds Thiazolyl quinazolones as Potential Antiviral and Antihypertensive Agents,Indian J. Chem. 2004; 43: 180-184.
21. Wasfy AAF. Synthesis and antimicrobial evaluation of some 2,2-disubstituted-3,3-biquinazolin-4(3H)-ones Indian J. Chem. 2003; 42B: 3102.
22. Georgey H, Abdel-Gawad N, Abbas S. Synthesis and Anticonvulsant Activity of Some Quinazolin-4-(3*H*)-one Derivatives. Molecules. 2008; 13: 2557-2569.
23. Seth PK, Parmar SS. In Vitro Inhibition Of Rat Brain Respiration By An Anticonvulsant (2-Methyl-3-Ortho-Tolyl-4-Quinazolone), Can. J. Pharmacol. 1965; 43:1019.
24. Bekhit AA, Habbib NS, EI. Bekhit A. Boll Synthesis and antimicrobial evaluation of chalcone and syndrome derivatives of 4(3H)-quinazolinone.. Chim.Farm. 2001; 140 : 297.
25. Chohan ZH, Spuran CT, Scozzafava A, Farroq MA. Antibacterial Schiff bases of oxalyl-hydrazine/diamide incorporating pyrrolyl and salicylyl moieties and of their zinc(II) complexes. J. Enzym.Inhib.Med. Chem. 2002; 17: 1-7.
26. K. Siddappa, Tukaram Reddy, M .Mallikarjun And C V. Reddy, Synthesis, Characterization and Antimicrobial Studies of 3-[(2-Hydroxy-quinolin-3-ylmethylene)-amino]-2-phenyl3H-quinazolin-4-one and its Metal(II) Complexes, E-Journal of Chemistry. 2008; 5(1), 155-162,.
27. ThilagavathiR, Kavitha HP, Arulmozhi R, Babu SM. Synthesis of 3-{4-[4-(Benzylideneamino)benzenesulfonyl]-phenyl}-2-phenylquinazolin-4(3*H*)-one Molbank, 2009;M589.