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Research Article

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Biological activity and mass spectra investigation of synthesized 1, 2, 4-triazine derivatives

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ABSTRACT

1, 2, 4-Triazine derivatives (3, b) were prepared via condensation of oxazolinones (2a, b) with thiosemicarbazide in acetic acid. Reaction of compound 3b with carbon disulphide, ethyl chloro acetate and p-methylphenacylbromid gives triazolo- [2,1,a]-1,2,4-triazine (4) and triazino- [2,1-a]-1,2,4-triazine derivatives (6 and 8). Acylation of compounds 4, 6 and 8 with acetic acid anhydride yielded the corresponding N-acetyl derivatives (5, 7 and 9). The mass spectral fragmentation patterns of some prepared compounds have been investigated in order to elucidate the structure of the synthesized compounds. Antimicrobial activities and antitumar activity were assayed against test bacteria, fungi and Breast carcinoma cells (MCF-7-cell line).

Key words: Oxazolinone,1, 2, 4-Triazine derivatives, Spectral studies, Synthesis, Biological activity**.**

INTRODUCTION

1, 2, 4-Triazine derivatives have been reported to posses a broad spectrum of biological activities, including antifungal[1, 2], Anti-HIV[3], anticancer[4], antiflammatory[5], analgesic[6], and antihypertensive activities[7]. Besides this triazines were used as herbicides, pesticides and dyes[8, 9]. This prompted us to synthesize five and six membered ring heterocycles fused with1,2,4-triazines and evaluate them for antimicrobial and anticancer activities. The electron impact (EI) ionization mass spectral fragmentation of some prepared compounds is also described.

EXPERIMENTAL SECTION

The melting points were determined in capillaries with a MEL-TEMP II laboratory Devices, USA, and are uncorrected. Infrared spectra were recorded on perkin-Elmer 337 spectrophotometer. Using KBr wafers. Proton NMR spectra were obtained on a varian EM 360 spectrometer using solution in hexadeuteriodimethyl sulfoxide with tetra- methyl silane as the internal standard. Mass spectra were recorded on a VG Autspec GEIF AB$^+$ and a Hewlett Packard MS-Engine thermospray and ionization by electron impact at 70 eV. The accelerating voltage was 6 KV, and the emission current 100 m *A*. Microanalysis was conducted using a perkin-Elmer 2408 CHN analyzer.

5-(Substituted) benzylidene-2-(p-chlorophenyl)-4-oxo-3, 1-oxazolinones (2a, b)
A mixture of **1** (0.01 mole), aromatic aldehydes (such as 4-methoxybenzaldehyde and 3-bromo -4-methoxybenzaldehyde) (0.01mole) and fused sodium acetate (0.01 mole) in acetic anhydride (0.01 mole) was fused on a hot-plat for 10-15 min. The reaction mixture was heated on a water-bath for 2 hr, then cooled and poured into water. The solid formed was filtered off, washed with hot water, dried, and recrystallization from ethanol to give **2**.

5-(4-Methoxy)benzylidene-2-(p-chlorophenyl)-4-oxo-3, 1-oxazolinone ***(2a)*** as yellow crystals(benzene), yield 76%, m.p. 152 ºC ; IR(KBr): 1782 (C=O), 1644 (C=N), 1596, 1501 (C=C), 1258, 1089, 1024 (C−O)cm^{-1}. ^{1}H-NMR(DMSO-d_6) δ: 3.91 (s, 3H, OCH_3), 6.89-7.73(m, 9H, Ar-H and H-olefinic) ppm .MS: m/z (%)= 315(M^++2,

5.20),314(M^++1, 2.90), 313(M^+, 11.90(8.90), 147(0.70), 146(2.9), 141(27.80), 140(11.70), 139(100), 138(12.60), 113(7.00), 111(23.30), 110(3.60), 105(0.40), 104(2.30), 103(4.00), 102(1.30), 92(0.50), 91(2.50), 90(0.8), 89(2.40), 88(1.40), 77(2.30), 76(7.60), 75(15.70), 65(1.30), 64(0.90), 63(2.90), 62(1.80), 51(4.30), 50(6.40). Anal. Found: C, 65.03; H, 3.72; N, 4.23. $C_{17}H_{12}N\ ClO_3$ requires: C, 65.17; H, 3.38; N, 4.47.

5-(3-Bromo-4-methoxy) benzylidene-2-(P-chlorophenyl) - 4-oxo-3, 1-oxazolinone ***(2b)*** as yellow crystals ethanol, yield 73%, m.p. 235 ºC; IR (KBr): 1780(C=O), 1641(C=N), 1602, 1583(C=C), 1221, 1078, 1032(C−O) cm^{-1}. 1H-NMR (DMSO-d_6): δ 3.92(s, 3H, OCH_3), 6.91-7.78(n, 8H, Ar-H and H-olefinic) ppm. MS: m/z(%)= 393(M^++2, 12.30), 392(M^++1, 6.50), 391(M^+, 12.030), 390(6.70), 373(3.30), 228(4.90),227 (21.30), 226(16.30), 225(21.60), 212(11.50), 211(15.60), 210(13.50), 197(2.30), 196(9.80), 173(6.80), 155(1.50), 154(11.10), 153(19.20), 141(30.20), 140(40.60), 139(100), 138(16.20), 137(14.20), 113(20.80), 111(45.90), 102(33.20), 101(14.10), 06(6.81), 103(13.60), 102(33.20), 101(14.10), 00(11.20), 89(20.80), 88(19.20), 87(13.60), 80(19.60), 79(2.30), 76(13.10), 75(38.10), 74(16.80), 73(20.80), 6(19.20), 64(2.60), 63(13.60), 62(19.20), 57(20.90), 51(21.90), 50(25.60). Anal. Found: C, 52.01; H, 2.69; N, 3.47. $C_{17}H_{11}NClBrO_3$ requires: C, 52.17; H, 2.81; N, 3.58.

5-(Substituted) benzylidene-3-(p-chlorophenyl)-2-aminothiocarbonyl-1, 2, 4-triazin-6-one (3a, b)

A mixture of **2a, b** (0.01 mole), and thiosemicarbazid (0.01 mole) in acetic acid (30 ml) was heated under reflux for 2-3 hr, then cooled and poured into water. The resulting solid was filtered off, washed with water, dried, and purified by recrystallization from suitable solvent to give **3**.

5-(4-methoxy)benzylidene-3-(P-chlorophenyl)-2-aminothiocarbonyl-1,2,4-triazin-6-one ***(3a)*** as yellow crystals(acetic acid), yield 79%, m.p. 201 ºC; IR(KBr): 3412, 3159(NH_2), 3225(NH), 1698(C=O), 1641(C=N), 1603, 1581(C=S), 1358(C=S), 1210, 1071(C−O) cm-1. 1H-NMR (DMSO-d_6) δ: .88(s, 3H, OCH_3), 6.63(s, 2H, NH_2), 6.98-7.78(m, 9H, Ar-H and H-olefinic), 10.30(S, 1H, NH)ppm . MS: m/z (%)= 388(M^++2, 6.30), 387(M^++1, 3.20), 386(M^+, 18.20), 385(7.20), 314(3.20), 313(13.90), 312(10.50), 148(0.10), 147(0.60), 146(3.60), 145(0.70), 142(2.00), 141(31.40), 140(11.20), 139(100), 113(9.60), 112(2.60), 111(27.10), 110(2.30), 105(0.60), 104(2.80), 103(5.70), 102(1.60), 91(2.60), 90(0.90), 89(2.70), 88(1.20), 87(1.10), 77(2.60), 76(9.90), 75(15.90), 74(4.00), 65(1.90), 64(0.90), 63(3.40), 62(3.50), 51(5.90), 50(10.00). Anal. Found: C, 55.81; H, 3.69; N, 14.39; S, 8.09. $C_{18}H_{15}N_4ClO_2S$ requires: C, 5.96; H, 3.89; N, 14.51; S, 8.29.

5-(3-Bromo-4-methoxy)benzylidene-3-(p-chlorophenyl)-2-aminothiocarbonyl-1,2,4-triazin-6-one ***(3b)*** as yellow crystals acetic acid, yield 77%, m.p. 192ºc; IR(KBr): 3421, 3187(NH_2), 3229(NH), 1693(CO),1635(C=N), 1603, 1589(C=C), 1339(C=S), 1225, 1073, 1035(C−O) cm^{-1}.^{1}H-NMR(DMSO-d_6).δ: 3.89(S, 3H, OCH_3), 6.61(s, 2H, NH_2), 6.91-7.89(m, 8H, Ar-H and H-olefinic), 10.51 (s, 1H, NH)ppm. MS: m/z (%)= 466(M^++2, 21.20), 465(M^++1, 10.20), 464(22.30, 425(9.40), 4424(7.80), 393(10.90), 392(35.90), 391(23.40), 390(21.90), 389(14.10), 377(9.40), 376(6.30), 228(9.40), 227(23.40), 226(18.60), 225(23.40), 224(17.20), 212(12.50), 211(15.60), 210(15.60), 197(4.70), 196(12.50), 173(7.80), 155(18.80), 154(14.10), 153(20.30), 142(7.80), 141(32.80), 140(43.60), 139(100), 138(18.80), 137(28.10), 131(9.40), 130(15.60), 119(9.40), 117(6.30), 113(21.90), 112(10.90), 111(46.90), 110(32.80), 106(7.80), 103(15.60), 102(39.10), 101(25.00), 100(12.50), 99(9.40), 89(21.90), 88(20.30), 87(15.60), 79(4.70), 76(14.10), 75(39.10), 74(18.80), 73(21.90), 66(30.30), 63(15.60), 62(20.30), 61(9.40), 59(20.30), 57(21.90), 55(12.50), 51(21.90), 50(26.60).Anal. Found:C, 46.33; H, 2.98; N, 11.89; S, 6.66. $C_{18}H_{14}N_4BrClO_2S$ requires: C, 46.55; H, 3.02; N, 12.07; S, 6.89.

6-(3-Bromo-4-methoxy)benzylidene-4-(P-chlorophenyl)-1,3-dithioxo-1,2,4-triazolo-[2, 1-a]-1, 2, 4-triazine-7-one (4)

A mixture of **3b** (0.01 mole), carbon disulphide (0.03 mole) and potassium hydroxide (0.03 mole) in ethanol (50 ml) was heated under reflux for 6 hr, and then cooled. The reaction mixture was acidified with dilute hydrochloric acid (1N). The solid formed was filtered, washed with water, dried and purified by recrystallization from benene to give 4 as pale yellow crystals, yield 56%, m.p. 134 ºC; IR(KBr): 3285(NH), 1689(C=O), 1635(C=N), 1381(C=S), 1605, 1581(C=C), 1071, 1034(C−O)cm^{-1}. ^{1}H-NMR (DMSO-d_6): δ 3.90(S, 3H, OCH_3), 6.98-7.89(m, 8H, Ar-H and olefinic-H), 10.71(S, 1H, NH) ppm. MS: m/z (%)= 508(M^++2, 10.20), 507(M^++1,3.20), 506(M^+, 4.20), 463(6.20), 462(8.60), 437(6.50), 436(5.10), 394(8.60), 393(11.80), 392(21.50), 391(8.60), 390(12.90), 389(12.90), 285(5.40), 254(6.50), 227(11.80), 226(8.80), 225(10.80), 214(5.40), 213(12.90), 212(12.90), 211(12.90), 210(11.80), 209(7.50), 195(5.40), 194(10.80), 168(5.40), 155(11.80), 154(8.60), 153(14.00), 152(8.60), 141(32.30), 140(33.30), 139(93.50), 138(100), 137(19.40), 133(5.40), 132(7.50), 118(4.30), 117(9.70), 116(7.50), 113(16.10), 111(30.10), 110(16.10), 105(4.20), 104(16.10), 103(11.80), 102(21.50), 101(11.80), 91(8.60), 90(6.50), 89(14.00), 87(12.90), 77(12.40), 76(15.10), 75(40.90), 74(21.50), 67(12.90), 65(11.60), 64(15.10), 63(20.40), 62(11.80), 60(34.40), 55(21.50), 53(11.80), 51(20.40), 50(25.80). Anal. Found: C, 44.89; H, 2.22; N, 10.97; S, 12.47. $C_{19}H_{12}N_4BrClO_2S_2$ requires: C, 45.06; H, 2.37; N, 11.07; S, 12.65.

7-(Substituted)benzylidene-5-(p-chlorophenyl)-4-thioxo-1-substituted-1,2,4-triazino-[2, 1-a]-1, 2, 4-triazine (6 and 8)

A mixture of **3b** (0.01 mole), ethyl chloro acetate and 4-methyl phenacyl bromide (0.01 mole) in acetic acid (30 ml) in presence of fused sodium acetate (0.03 mole) was heated under reflux for 4 hr, then cooled and poured into water. The resulting solid was filtered off, washed with water, dried and purified by recrystallization from a suitable solvent to give **6** and **8**.

7-(3-Bromo-4-methoxy) benzylidene-5-(P-chlorophenyl)-4-thioxo-2, 3-dihydro-1,2,4-triazino [2,1-a]-1,2,4-triazin-1,8-dione ***(6)*** as yellow crystals (ethanol), yield 63%, m.p. 233 ºc; IR(KBr): 3227(NH), 1705-1689(C=O), 1633(C=N), 1606, 1583(C=C), (C=S), 1125, 1071(C−O), cm^{-1}.^{1}H-NMR(DMSO-d_6). δ: 3.32(s, 2H,N− CH_2 CO), 3.89(S, 3H, OCH_3), 6.91-7.81(m, 8H, Ar-H and H-olefinic), 10.35 (S, 1H, NH)ppm. MS: m/z (%) = 506(M^++2, 48.30), 505(M^++1, 34.50), 504(M^+, 41.40), 503(13.80), 393(34.50), 392(13.80), 391(31.00), 390(11.20), 254(31.0), 252(58.60), 251(31.00), 227(31.00), 226(13.80), 216(13.80), 215(24.10), 214(31.00), 213(27.60), 210(24.10), 183(10.30), 182(24.10), 142(24.10), 141(3.40), 139(62.10), 138(41.40), 117(20.70), 116(41.40), 114(31.00), 113(34.50), 111(6.90), 110(27.60), 103(55.30), 102(37.90), 101(41.40), 91(37.90), 89(24.10), 88(27.60), 87(100), 89(34.50), 81(41.40), 80(37.90), 77(20.70), 76(20.70), 75(31.00), 74(24.10), 66(13.80), 63(24.10), 62(4.80), 51(20.70), 50(6.90). Anal. Found: C, 47.47; H, 2.63; N, 11.02; S, 6.22. $C_{20}H_{14}N_4BrClO_3S$ requires: C, 47.62; H, 2.78; N, 11.11; S, 6.35.

7-(3-bromo-4-methoxy) benzylidene-5-(p-chlorophenyl)-4-thioxo-1-(p-tolyl)-1, 2,4-triazino-[2,1-a]-1,2,4-triazin-8-one ***(8)*** as yellow crystal benzene, yield 61%; m.p. 110 ºC; IR(KBr):3230(NH), 1698(C=O), 1631(C=N), 1607, 1589(C=C), 1338(C=S), 1210, 1071(C−O) cm^{-1}. ^{1}H-NMR (DMSO-d_6): δ 2.35(S, 3H, CH_3), 3.89(S, 3H, OCH_3), 6.89-7.89(m, 13H, Ar-H and H- olefiic), 10.56(S, 1H, NH) ppm. Anal. Found: C, 55.89; H, 3.28; N, 9.51; S, 5.33. $C_{27}H_{20}N_4BrClO_2S$ requires; C, 6.06; H, 3.46; N, 9.69; S, 5.5.

Acetylation of compounds 4, 6 and 8. Formation of N-acetyl derivatives (5, 7 and 9)

A solution of **4**, **6** and **8** (0.01 mole) in acetic anhydride (20 ml) was heated under reflux for 2 hr, then cooled and poured into ice-water. The solid obtained was filtered off, washed with water, dried, and purified by recrystallization from suitable solvent to give **5**, **7** and **9**.

6-(3-bromo-4-methoxy)benzylidene-4-(P-chlorophenyl)-1,3-dithioxo-2-2-acetyl-1,2,4-triazolo-[2,1-a]-1,2,4-triazine-7-one ***(5)*** as yellow crystals (benzene), yield 57%, m.p. 90 ºC; IR(KBr): 1705-1889(C=O), 1629(C=N), 1607, 1589(C=C), 138(CS), 1221, 1073(C−O) cm^{-1}. ^{1}H-NMR (DMSO-d_6): δ 2.41(s, 3H, $COCH_3$), 3.88(S, 3H, OCH3), 6.99-7.91(m, 8H, Ar-H and H-olefinic) ppm. MS: m/z(5) = 550(M^++2, 10.20), 549(6.20), 548(M^+, 11.30), 493(1.5), 492(1.90), 491(.10), 490(3.90), 489(2.20), 488(4.30), 452(1.20), 451(5.70), 450(4.0), 449(12.70), 448(13.60), 447(9.60), 446(10.50), 439(2.70), 438(2.80), 437(2.10), 436(2.80), 409(3.00), 407(9.00), 406(9.10), 405(6.70), 404(5.80), 393(4.90), 392(9.60), 391(10.00), 390(6.70), 389(7.60), 364(1.30), 361(1.60), 327(1.80), 325(2.50), 324(1.90), 312(2.10), 311(3.40), 310(2.10), 296(1.20), 287(2.10), 284(1.60), 282(1.20), 25(1.80), 254(2.10), 253(2.70), 252(1.60), 239(1.80), 238(1.60), 227(7.20), 226(6.60), 225(10.30), 213(6.00), 212(10.80), 211(10.20), 210(7.60), 201(3.10), 200(2.40), 197(8.50), 196(7.90), 195(20.90), 194(13.80), 188(1.60), 187(2.10), 186(1.20), 185(2.10), 173(2.50), 172(2.20), 171(1.60), 170(2.70), 169(3.40), 167(3.90), 157(2.50), 155(14.10), 153(3.80), 152(32.10), 141(35.00, 140(29.30), 139(100), 138(34.10), 132(1.80), 131(2.80), 130(4.80), 125(3.90), 119(5.10), 118(6.60), 177(3.90), 116(5.80), 113(12.40), 111(41.30), 110(6.30), 105(3.90), 104(4.60), 103(8.50), 102(22.40), 101(6.10), 91(3.10), 90(7.80), 89(8.20), 88(9.00), 87(5.80), 76(16.90), 75(26.00), 74(9.90), 65(2.10), 63(10.80), 62(8.10), 51(14.20), 50(14.10). Anal. Found: C, 45.72; H, 2.35; N, 10.09; S, 11.51. $C_{21}H_{14}N4BrClO_3S$ requires: C, 45.98; H, 2.55; N, 10.22; S, 11.68.

7-(3-bromo-4-methoxy) benzylidene-5-(p-chlorophenyl)-4-thioxo-3-acetyl-1, 2, 4-triazino-[2,1-a]-trizine-1,8-dione ***(7)*** as yellow crystals (ethanol), yield 71%, m.p. 215 ºC; IR(KBr): 1705-1693(C=O), 1632(C=H), 1603, 1583(C=C), 1338(C=S), 1215, 1071(C−O)c,-1. H-NMR(DMSO-d_6): δ 2.35(s, 3H, $COCH_3$), 3.35(S, 2H,CH_2CO), 3.91(S,3H, OCH_3), 6.91-7.79(m, 8H, Ar-H, H-olefinic) pm. MS: m/z (%) = 548(m^++2, 8.70), 547(m^++1, 15.20), 546(15.20), 507(15.20), 506(1.00), 504(13.00), 503(10.90), 450(13.00), 449(13.00), 447(15.20), 433(13.00), 432(6.50), 423(13.00), 422(10.90), 394(23.90), 392(41.30), 391(32.60), 390(45.70), 389(32.60), 388(10.90), 377(1.00), 360(15.20), 58(15.20), 357(15.20), 356(10.90), 339(10.90), 338(10.90), 310(13.00), 276(13.00), 275(6.50), 252(8.70), 251(10.90), 227(39.10), 226(32.60), 225(47.80), 224(26.10), 215(10.90), 214(13.00), 213(17.40), 212(37.00), 211(32.60), 210(26.10), 201(15.20), 198(15.20), 197(21.70), 196(17.40), 195(17.40), 194(15.20), 180(10.90), 178(15.20), 168(15.20), 167(13.00), 158(13.00), 156(21.70), 155(21.70), 152(15.20), 147(13.00), 144(13.00), 142(15.70), 141(41.30), 140(50.00), 139(100), 138(80.40), 137(78.30), 130(21.70), 129(21.70), 119(13.00),118(13.00), 117(23.90), 116(34.80), 115(30.40), 113(39.10), 111(54.30), 110(34.80), 105(15.20), 104(14.50), 103(28.30), 102(50.00), 101(19.80), 90(6.50), 89(34.80), 88(32.60), 87(21.70), 82(39.10), 80(43.50),

79(30.10), 77(30.40), 76(45.70), 75(58.70), 74(3.60), 73(21.70) 68(19.60), 64(34.80), 63(28.30), 62(10.90), 60(43.50), 51(50.00), 50(34.80) Anal. Found: C, 48.19; H, 2.79; N, 10.07; S, 5.66. $C_{22}H_{16}N_4BrClO_4S$ requires: C, 48.35; H, 2.93; N, 10.26; S, 5.86.

7-(3-bromo-4-methoxy) benzylidene-5-(P-chlorophenyl)-4-thioxo-3-acetyl-1-(P-tolyl)-1,2,4-triazino-[2,1-a]-1,2,4-triazin-8-one **(9)** as yellow crystals (benzene), yield 58%, m.p. 160 ºC; IR(KBr): 1703-1695(C=O), 1632(C=N), 1606, 1593(C=C),1338(C=S), 1215, 1069(C−O)cm^{-1}. ^{1}H-NMR (DMSO-d_6): δ 2.21(S, 3H, CH_3), 2.35(S, 3H, $COCH_3$), 3.88(S, 3H, OCH_3), 6.89-7.91(m, 13H, Ar-H and H-olefinic) ppm. Anal. Found: C, 56.02; H, 3.33; 8.99; S; 5.01. $C_{29}H_{22}N_4BrClO_3S$ requires: C, 56.13; H.3.55; N, 9.03; S, 5.16

RESULTS AND DISCUSSION

1- Chemistry

5-(Substituted) benzylidene-2-(p-chlorophenyl)-3, 1-oxazol-4-ones (**2a**, **b**) were prepared by the reaction of 4-methoxy benzaldehyde and 3-bromo-4-methoxy benzaldehyde with N-(p-chlorobenzoyl)-glycine in presence of fused sodium acetate and acetic anhydride under fusion. Treatment of 3,1-oxaszolinone (**2**a, b) with thiosmicarbazide in acetic acid under reflux, yielded the corresponding 5-(substituted)benzylidene-3-(p-holorophenyl) 2-aminothiocarbonyl-1,2,4-triazin-6-one (**3a**, **b**). The reaction of 1, 2, 4-triazine derivative (**3a)** with carbon disulophide in alcoholic potassium hydroxide under reflux give the corresponding 6-(3-bromo-4-methoxy) benzylidene-4-(p-chlorophenyl)-1, 3-dithioxo-1, 2, 4-triazolo-[2, 1-a]-1, 2, 4-triazine-7-one (**4**). Treatment[10] of substituted 1, 2, 4-triazine (**3b**) with ethyl chloro acetate and 4-methyl penacyl bromide in presence of fused sodium acetate under reflux, afforded the corresponding 7-(3-bromo-4-methoxy)benzylidene-(p-hlorophenyl)-4-thioxo-2,3-dihydro-1,2,4-triazino-[2,1-a]-1,2,4-triazin-1,8- diones (**6**) and7-(3-bromo-4-methoxy)benzylidene-5-(p-chlorophenyl)-4-thioxo-1-(p-tolyl)-1, 2, 4-triazino-[2,1-a]-1,2,4-triazen-8-one (**8**, scheme1). Acetylation[11] of compounds 3, 6 and 8 with acetic anhydride under reflux led to the formation of 6-(3-bromo-4-methoxy)benzylidene-4-(p-chlorophenyl)-1,3-dithioxo-2-acetyl-1,2,4-triazino-[2,1-a]-1,2,4-triazine-7-one(5),7-(3-bromo-4-methoxy)benzylidene-5-(p-chlorophenyl)-4- thioxo-3-acetyl-1,2,4-triazino-[2,1-a]-1,2,4-triazine-7-one(**5**),7-(3-bromo-4- methoxy)benzylidene-5-(p-chlorophenyl)-4-thioxo-3-acetyl-1,2,4-riazino[2,1-a]-1,2,4-triazin-1,8-dienes(**7**)and7-(3-bromo-4-methoxy)benzylidene-5-(p-chlorophenyl)-4-thioxo-3-acetyl-1-(p-tolyl)-1,2,4-triazino-[2,1-a]-1,2,4-triazine-8-one (**9**, scheme1), respectively.

2- Mass spectroscopy

The mass spectral decomposition modes of [12, 13] of the prepared 1, 2, 4-triazine derivatives have been investigated. The mass spectra of compounds **3a** (Figure **1**) and **3b** (Figure **2**) showed an intense molecular ion peak at m/z 386 and m/z 464, corresponding to the molecular formula $C_{18}H_{15}N_4ClO_2S$ and $C_{18}H_{14}N_4BrClO_2S$, respectively. The molecular ion of compounds **3a** and **3b** (Scheme 2) underwent fragmentations to produce peaks of m/z 312 and m/z 390, corresponding to the molecular ion of 4-(3-substituted-4-methoxybenzylidene)-2-p-chlorophenyl-imidazolidin-5-ones. The ion of m/z 312 and m/z 390 fragmented with rearrangement processes to give the stable ion of m/z 139. It further underwent loss of CH_2=NH and hydrochloric acid (HCl) to give peaks at m/z 111 and m/z 75, respectively. Also, the ion of m/z 312 and 390 underwent fragmentation with rearrangement to give the ion of m/z 147 and m/z 225. The loss of NH group from the ions of m/z 147 and m/z 225 gave a peak at m/z 132 and m/z 210, respectively

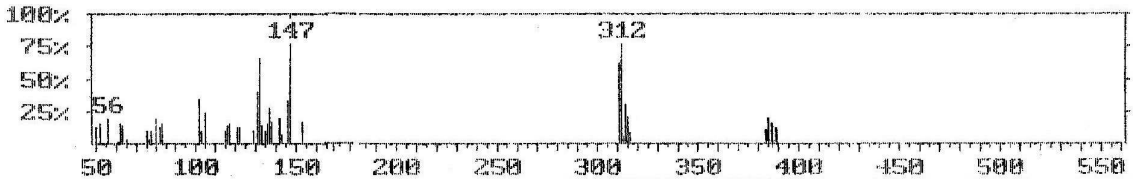


Figure 1: 70 eV mass spectrum of 3a

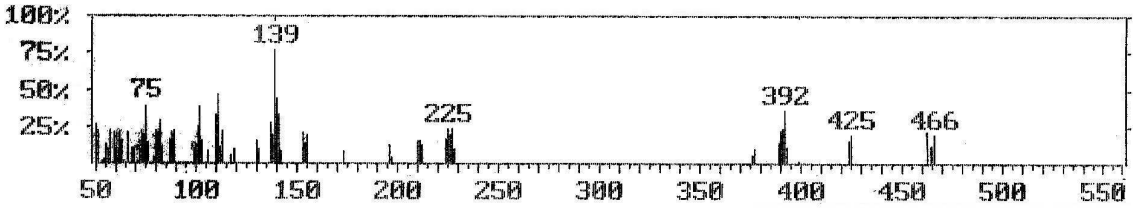
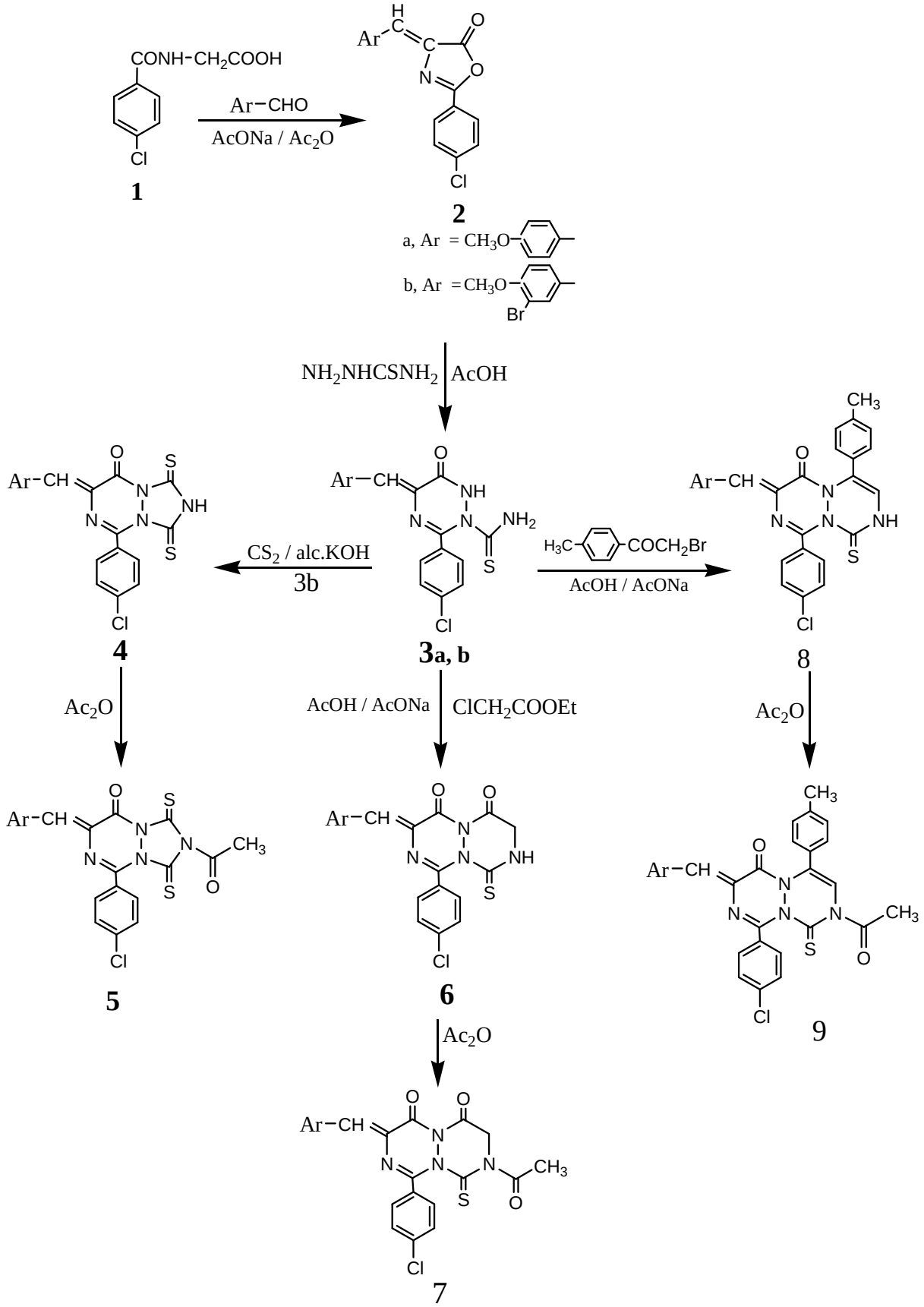


Figure 2: 70 eV mass spectrum of 3b


CONH-CH2COOH
Ar−CHO
AcONa / Ac2O
1
2
a, Ar = CH3O
b, Ar = CH3O
Br
NH2NHCSNH2 | AcOH
3a, b
CS2 / alc.KOH
3b
4
H3C-COCH2Br
AcOH / AcONa
8
Ac2O
AcOH / AcONa | ClCH2COOEt
Ac2O
5
6
9
Ac2O
7

Scheme 1

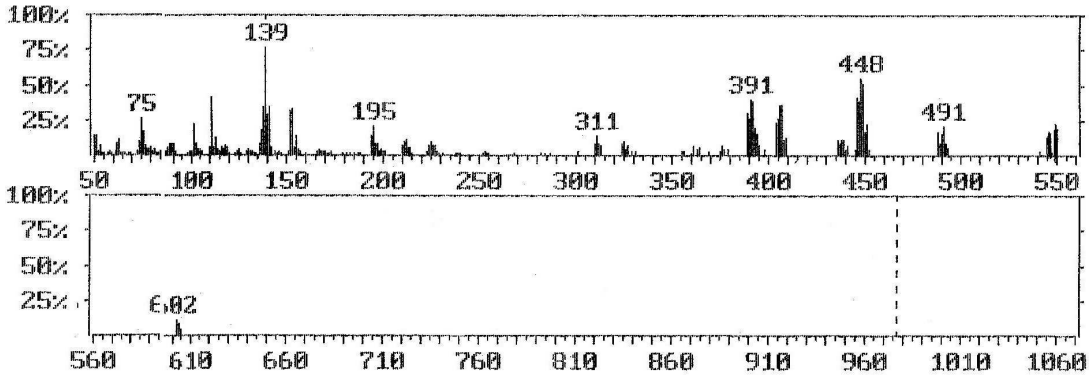


Figure 3: 70 eV mass spectrum of 5

The mass spectrum of compound **5** (Figure 3) showed an intense molecular ion peak at m/z 548 corresponding to the molecular formula $C_{21}H_{14}N_4BrClO_3S$. The molecular ion of compound **5** underwent fragmentation (Scheme 3) to produce the peak at m/z 448 by losing ketene (CH_2CO) molecule and isothiocyanate group (NCS).The loss of isothiocyanate group (NCS) from the ion with m/z 448 gave ion at m/z 390.The fragmentation ion of m/z 390 which has further broken via pathway similar to compound 3b to give the stable ion at m/z 139

The mass spectrum of compound **6** and **7** (Figure 4 and 5) are fully consistent with the assigned structures. In most cases intense molecular ion peaks were observed. Thus, compounds **6 a**nd **7** showed intense molecular ion peaks at m/z 504 and m/z 546, consistent with the molecular formula $C_{20}H_{14}N_4BrClO_3S$ and $C_{22}H_{16}N_4BrClO_4S$, respectively

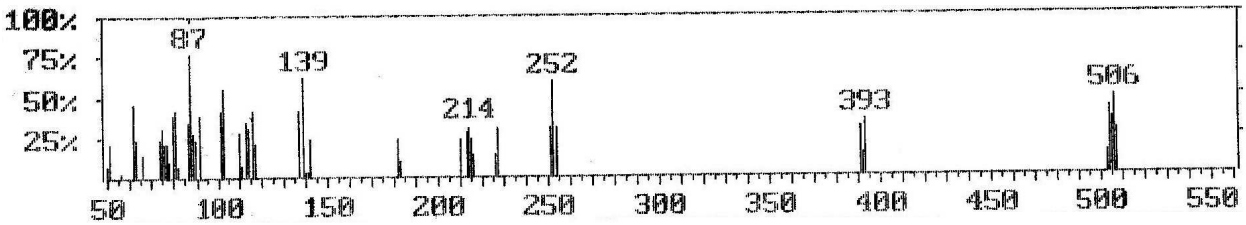


Figure 4: 70 eV mass spectrum of 6

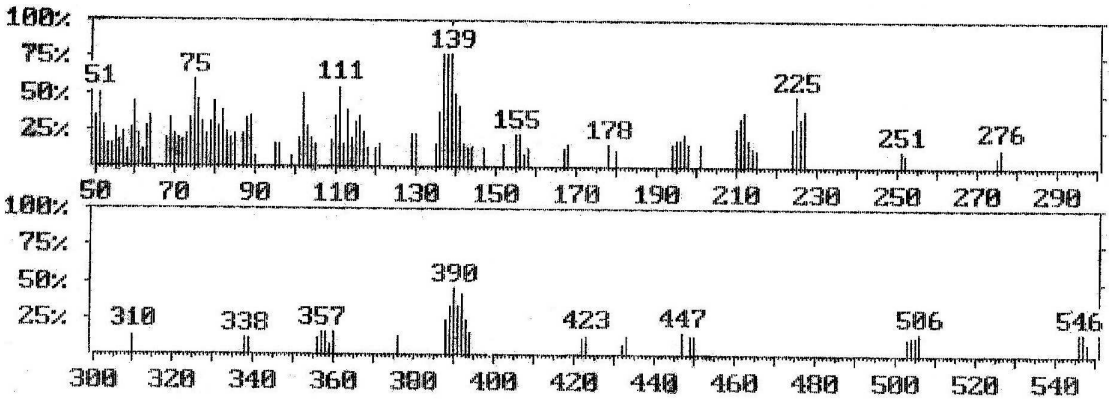
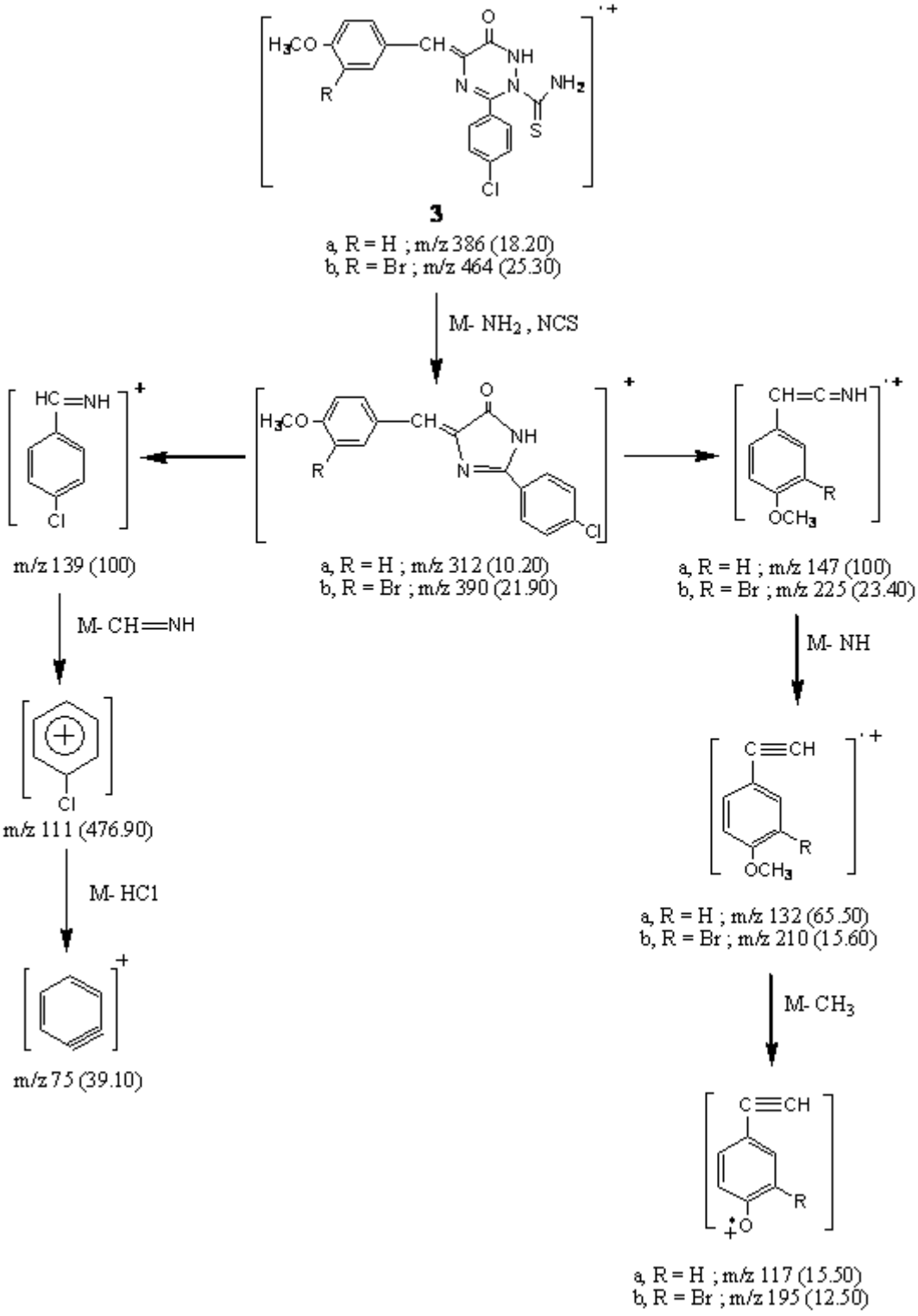


Figure 5: 70 eV mass spectrum of 7

The M+2 peak was also observed at m/z 506 and m/z 548 along with the molecule ion peak due to the presence of isotopes of bromine and chlorine atoms present in the compound. From the mass spectrum of compound 7, it was concluded that the molecular ion was at m/z 546. The ion of m/z 546 underwent fragmentation to produce a peak at m/z 504 by losing ketene molecule CH_2CO), corresponding to the molecule ion of compound **6**. The loss of formyl group (CHO) and hydrogen cyanide from the molecular ion of compound 6 at m/z 504 gave a peak at m/z 448 which we obtain from the compound **5** by losing ketene molecule and isothiocyanate group (NCS).The fragment ion of m/z 448 further broke via pathway similar to compound **5** (scheme 4).

Scheme 2: Main fragmentation pathway of compounds 3a and 3b



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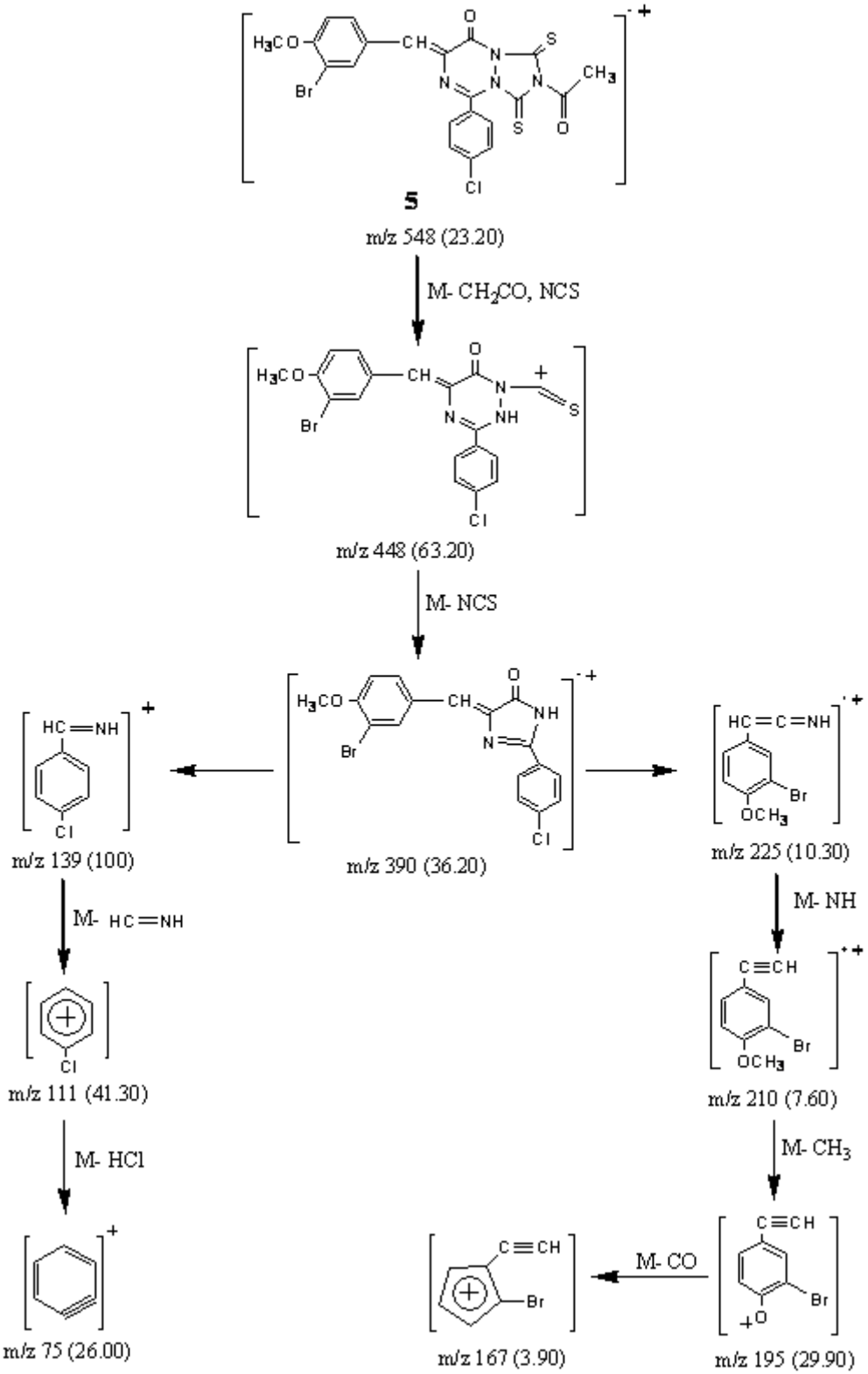
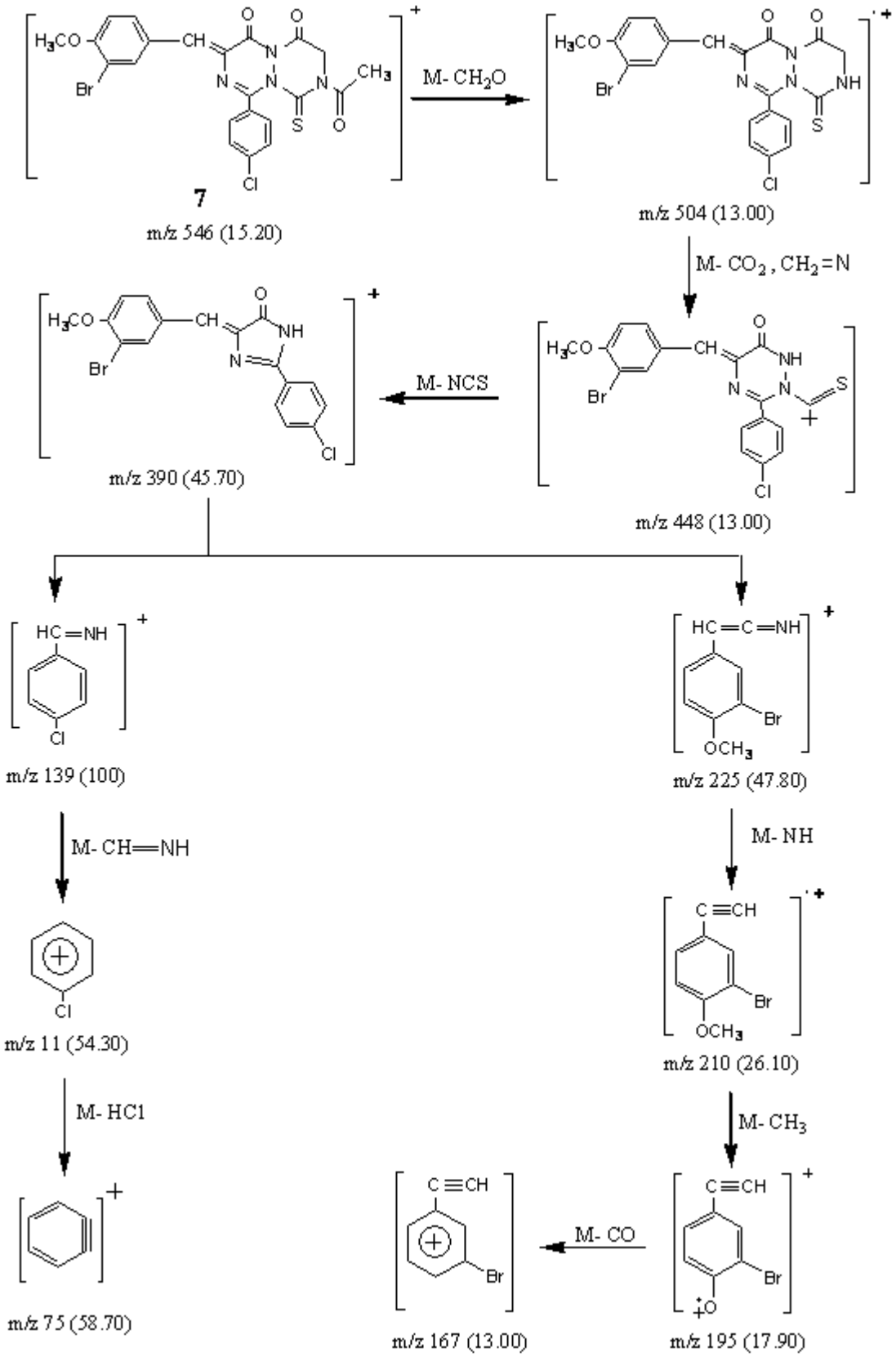
Scheme 3: Main fragmentation pathway of compound 5



Scheme 4: Main fragmentation pathway of compound 7



Biological Assay

1- Antimicrobial activity

Antimicrobial activity was measured by the following agar-diffusion technique [14-16]. All the newly synthesized compounds were tested in vitro for several at strains of bacteria such as Bacillus subtilis, Streptocccus pneumonia, Staphylococcus aureus, Escherichia coli and Pseudomonas. Also, antifungal activity against Aspergillus niger and Penicillium sp. Using agar-plate diffusion technique. The compounds were tested at 100 µg/mL concentration and the activity was determined by measuring the zone of inhibition. The screening results are listed in table 1. All compounds showed activity against bacteria, while compounds 4 and 5 did not exhibit any activity against streptococcus pneumonia. Also, all compounds were active against fungi, except compound 4 was non active against penicillium sp.

2- Anticancer evaluation

Cytotoxic and antitumor activity of some synthesized compounds **3b**, 4, **5,** 6, and **7** which were tested against MCF-7 cell line according to the method of Masmann (1983) and Vijayen et al.(2004). Inhibitory activity against Breast carcinoma cells (MCF-7 cell line) was detected by using different concentration of the tested compounds (50, 25, 12.5, 6.25, 3.125 and 1.56 µg) and viability cells (%) were determined by colorimetric method. Also, inhibitory concentration fifty (IC_{50}) was calculated from tables 2-6 and figures 6-10. Inhibtory concentration fifty (IC_{50}) was found to be 3.1 µg for compound 3b, 4.8 µg for compound 4, 3.80 µg for compound 5, 6.20 µg for compound 6 and 5.60 µg for compound 7. Results revealed that, all tested compounds have cytotoxic and antitumor activity against Breast carcinoma cell line with superiorty of compound 3b with 3.1 µg and compound 5 with 3.8 µg. Compound 3b contained triazine skeleton and aminothiocarbonyl group with attached nitrogen atom number two in triazine ring. Compounds **4** and **5** contained fused triazine skeleton with five memberd ring, while the compounds **6** and **7** contained fused triazine skeleton with six memberd ring. Acetyl derivatives **5** and **7** were found to exhibit potent anticancer activity which were better than compounds **4** and **6**.From the inhibitory concentration fifty (IC_{50}) we can apply the compounds **3**b and **5** as drug against tumor.

Table1: Antimicrobial activity of some prepared compounds 3-9

Co*mpd* No	Antibacterial Activity					Antifungal Activity	
	Gram Positive Bacteria			Gram Negative Bacteria			
	Bacillus Subtilis	*Staphylococcus Aureas*	*Streptococcus Penumonia*	*Escherichia Coli*	*Pesudomonas Solanarium*	*Aspergillus Niger*	*Penicillium*
3b	+	+	-	+	+	+	+
4	+	+	-	+++	+++	++	-
5	+	+	-	+++	+++	+++	+ +
6	++	++	+++	+	+	+ +	+++
7	+ +	+	++	+ +	++	+ + +	+
8	+++	++	+ + +	+++	+	+	+ + +
9	++	+++	+++	+ + +	+ + +	+++	+++

No antimicrobial activity, (++) Moderate activity , (+) Mild activity, (+++) Marked activity

Table 2: For compound 3b

Sample conc. (µg)	Viability %
50	9.16
25	20.64
12.5	32.41
6.25	39.18
3.125	48.30
1.56	70.82
0	100.00

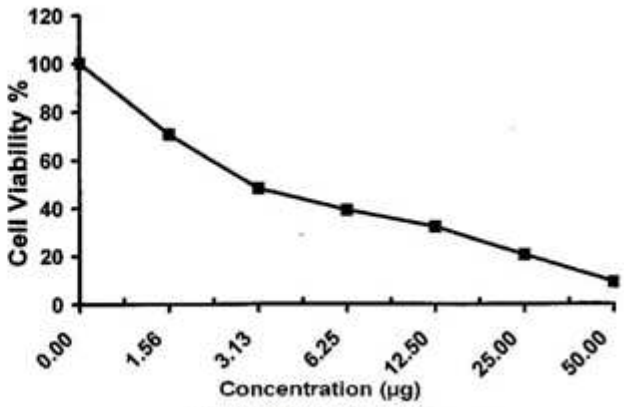


Figure 6: For compd 3b

Table 3: For compound 4

Sample conc. (µg)	Viability %
50	21.72
25	28.44
12.5	36.35
6.25	47.46
3.125	56.12
1.56	70.88
0	100.00

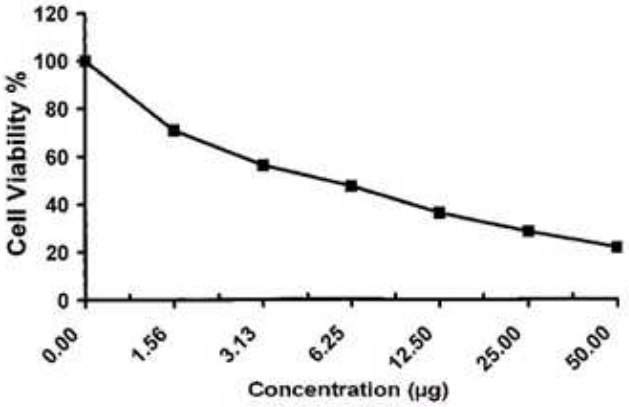


Figure 7: For compd 4

Table 4: For compound 5

Sample conc. (µg)	Viability %
50	21.72
25	28.44
12.5	36.35
6.25	47.46
3.125	56.12
1.56	70.88
0	100.00

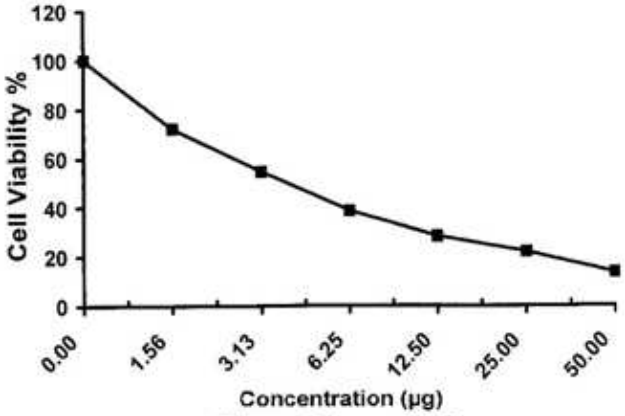


Figure 8: For compd 5

Table 5: For compound 6

Table 5: For compound 6

Sample conc. (µg)	Viability %
50	21.72
25	**28.44**
12.5	36.35
6.25	47.46
3.125	56.12
1.56	70.88
0	100.00

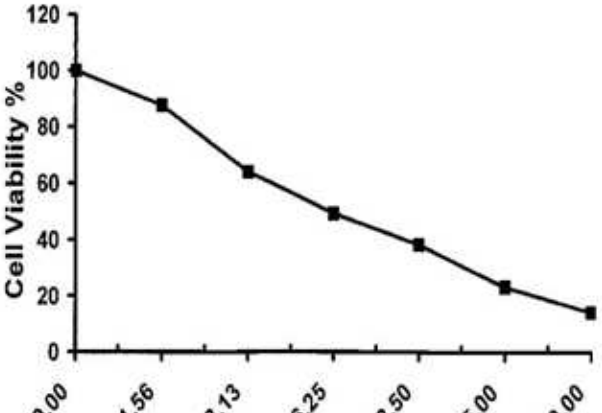


Figure 9: For compd 6

Table 6: For compound 7

Sample conc. (µg)	Viability %
50	21.72
25	28.44
12.5	36.35
6.25	47.46
3.125	56.12
1.56	70.88
0	100.00

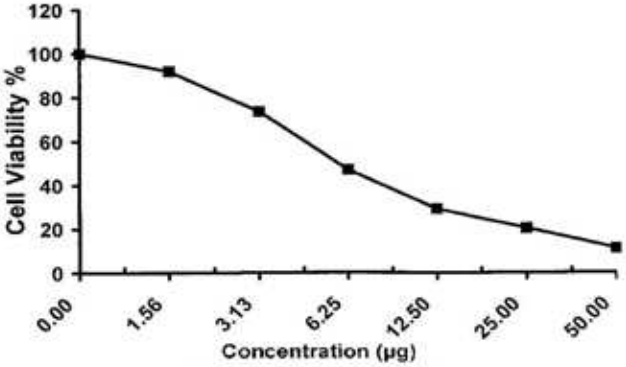


Figure 10: For compd 7

CONCLUSION

In conclusion, we have described the synthesis and biological activities of a new 1, 2, 4-triazine derivatives 3-9. The compounds showed in vitro growth inhibitory activity against the tested organisms or higher than streptomycin. The biological data revealed that with slight modifications in the structure one can plan for the drug design**.**

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