
MW assisted synthesis of some heterocyclic compounds and their Mannich bases under solvent free conditions and their biological assay

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Síntesis asistida por microondas de algunos compuestos heterocíclicos y sus bases de Mannich en condiciones de ausencia de disolvente y ensayos biológicos

Síntesi assistida per microones d'alguns compostos heterocíclics y les seves bases de Mannich en condicions d'absència de dissolvent i assaigs biològics

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RESUMEN

Se presenta una síntesis muy eficiente de distintos compuestos heterocíclicos nitrogenados, en condiciones de ausencia de disolvente asistida por microondas. Su aminometilación se llevó a cabo sobre alúmina ácida con radiación de microondas para obtener las correspondientes bases de Mannich. El método es limpio, rápido, fácil, eficiente, económico y medioambientalmente benigno. La estructura de los compuestos sintetizados se caracterizó en base a su análisis elemental y datos espectroscópicos. La actividad antimicrobiana de estos compuestos se evaluó frente a bacterias y hongos.

Palabras clave: Aminometilación, Alúmina ácida, irradiación con microondas.

SUMMARY

A highly efficient MW assisted synthesis of different nitrogen containing heterocyclic compounds under solvent free conditions was reported. Their aminomethylation was carried out on acidic alumina under microwave irradiation to give the corresponding Mannich bases. The method is clean, fast, facile, efficient, low cost and environmentally benign. The structure of synthesized compounds has been characterized on the basis of their elemental analysis and

spectral data. These compounds were evaluated for their antimicrobial activity against bacteria and fungi.

Keywords: Aminomethylation, Acidic alumina, Microwave irradiation

RESUM

Es presenta una síntesi molt eficient de diferents compostos nitrogenats heterocíclics, en condicions d'absència de dissolvent, i assistida per microones. La seva aminometilació es va realitzar sobre alúmina àcida, amb radiació de microones per obtenir les corresponents bases de Mannich. El mètode es net, ràpid, eficient, econòmic, i mediambientalment benigne. La estructura dels compostos sintetitzats se va caracteritzar en base al seu anàlisi elemental i les dades espectroscòpiques. La activitat antimicrobiana d'aquests compostos es va avaluar amb bacteris i fongs.

Paraules clau: Aminometilació, Alúmina àcida, irradiació amb microones.

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INTRODUCTION

The growth of chemistry has been closely associated with the development of new bioactive molecules, which have been synthesized using newer and greener routes. Conventional methods of synthesis of chemical compounds with desired biological properties are time consuming and expensive, too. Thus, traditional medicinal chemistry remains the bottlenecks in the drug discovery processes. Consequently, there is an increased interest in technologies and concepts that facilitates more rapid synthesis and screening of chemical substances to identify compounds with appropriate qualities. One such high-speed technology is Microwave Assisted Organic Synthesis (MAOS). In recent years, the acceleration of a wide range of chemical reactions using microwave dielectric heating has been reported¹. The solvent free conditions under microwave irradiation offer several advantages; solvents are often expensive, toxic and difficult to remove and are polluting agents. A considerable amount of work has been reported on the synthesis and pharmacological activity of various Mannich bases for analgesic, antispasmodic, anesthetic and antimicrobial activity as well as intermediate in drug synthesis²⁻¹⁴. In this context, literature survey has revealed a number of reports on antimicrobial activity of N-Mannich bases derived from different heterocycles such as pyrrole, pyrazole, benzimidazole, benzotriazole etc¹⁴⁻²⁰. Microwave induced aminomethylation using solid support fulfils the need of 'Green chemistry' to a greater extent because it minimizes the use of solvents. Moreover, these supports are reusable. In the present paper, a simple, highly efficient and eco-friendly process for synthesis of some heterocyclic compounds and corresponding Mannich bases has been reported.

RESULTS AND DISCUSSION

All heterocyclic compounds have been synthesized under microwave exposure. The structures of these compounds have been confirmed by their physical, analytical and spectral data. The synthesized compounds on reaction with formaldehyde and secondary amines (morpholine / N-methyl piperazine) yielded the corresponding Mannich bases on alumina under MW irradiation. When compound (1) was treated with formaldehyde and secondary amines, 1-[(morpholin-4-yl/4-methyl piperazin-1-yl)methyl]-1H-benzotriazole were obtained (**2a** & **2b**). Disappearance of N-H stretching band at 3500-3400 cm⁻¹ and appearance of IR bands at 1226/1215, 2832/2850 & 1097 cm⁻¹ due to C-N, CH₂ and C-O groups indicate presence of morpholine / N-methyl piperazine moiety in the desired compound. ¹H NMR and mass spectrum were also confirmed the structure of product.

Compound (3) on reaction with formaldehyde and secondary amines afforded (**4a** & **4b**). The structure of compounds (**4a** & **4b**) has been confirmed by disappearance of N-H stretching at 3480 cm⁻¹ and bands appearing at 1203 and 1072 cm⁻¹, respectively due to C-N and C-O stretching, which shows that morpholine / N-methyl piperazine moiety has been entered. ¹H NMR and molecular ion peaks appearing in mass spectra further supported the structure of Mannich bases.

Compound (5) on aminomethylation yielded (**6a** & **6b**). In the IR spectrum, disappearance of N-H stretching at

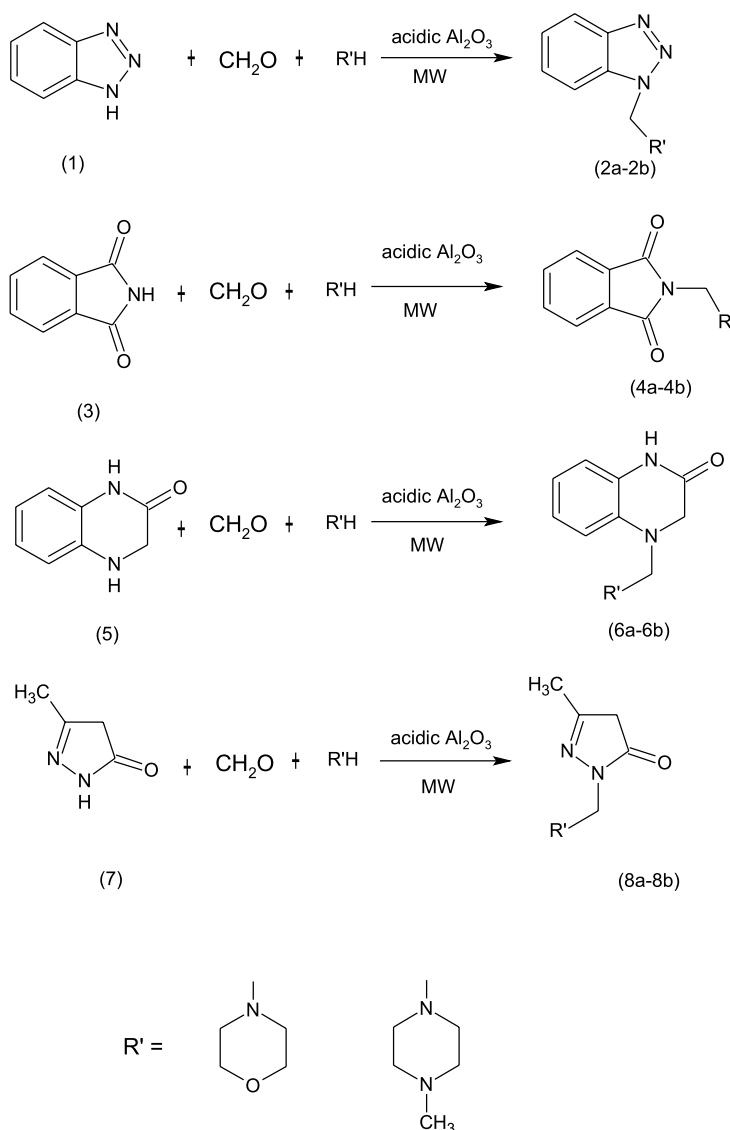
3420 cm⁻¹ and appearance of bands at 1213 and 1081 cm⁻¹ due CN & C-O groups indicate the formation of Mannich bases. In ¹H NMR spectrum, single singlet appeared at δ 9.12 due to the presence of one N-H group indicating that one N-H group underwent aminomethylation reaction. Similarly, reaction of compound (7) afforded (**8a** & **8b**). Disappearance of broad band due to N-H group between 3300-3100 and appearance of peaks at 1225 and 1122 cm⁻¹ due to C-N and C-O groups favour the structure of compounds (**8a** & **8b**). Further, ¹H NMR and mass spectra also supported the structure of aminomethylated compounds. The physical and analytical data are presented in **Table (1)** and spectral data are given in **Table (2)**.

Table 1 : Characterization data of synthesized compounds

| Compound | R' | Molecular formula | Molecular weight | Melting point | Yield (%) | Analysis of N (%) Calcd. (Found) |
|-----------|----|---|------------------|---------------|-----------|----------------------------------|
| 2a | | C ₁₁ H ₁₄ N ₄ O | 218 | 135 | 78 | 25.67 (25.64) |
| 2b | | C ₁₂ H ₁₇ N ₅ | 231 | 163 | 75 | 30.28 (30.25) |
| 4a | | C ₁₃ H ₁₄ N ₂ O ₃ | 246 | 230 | 80 | 11.38 (11.35) |
| 4b | | C ₁₄ H ₁₇ N ₃ O ₂ | 259 | 200 | 75 | 16.20 (16.18) |
| 6a | | C ₁₃ H ₁₇ N ₃ O ₂ | 247 | 187 | 82 | 16.99 (16.95) |
| 6b | | C ₁₄ H ₂₀ N ₄ O | 260 | 233 | 70 | 21.52 (21.50) |
| 8a | | C ₉ H ₁₅ N ₃ O ₂ | 197 | 287 | 80 | 21.30 (21.28) |
| 8b | | C ₁₀ H ₁₈ N ₄ O | 210 | 215 | 75 | 26.64 (26.62) |

Table 2 : Spectral data of synthesized compounds

| Compd. | | Spectral Data |
|-----------|----------------------------------|--|
| 2a | IR (cm ⁻¹): | 3064 (C-H str., Ar-H), 2832 (C-H str., CH ₂), 1600 (N=N str.), 1411 (CH ₂ bend.), 1226 (C-N str.), 1097 (C-O str.), 795 (Ar-H bend.). |
| | ¹ H NMR (δ): | 7.50-7.32 (m, 4H, ArH), 4.32 (s, 2H, N-CH ₂ -N), 3.86 (t, 4H, CH ₂ -O-CH ₂), 2.50 (t, 4H, CH ₂ -N-CH ₂) |
| | MS (m/z) | 218 [M] ⁺ , 190 [M-N ₂] ⁺ , 146 [M-C ₄ H ₈ O] ⁺ , 132 [M - C ₄ H ₈ NO] ⁺ , 119 [C ₂ H ₅ N] ⁺ , 118 [M - C ₂ H ₁₀ NO] ⁺ , 104 [C ₆ H ₄ N ₂] ⁺ , 100 [C ₅ H ₁₀ NO] ⁺ , 86 [C ₄ H ₈ NO] ⁺ . |
| 2b | IR (cm ⁻¹): | 3075 (C-H str., Ar-H), 2850 (C-H str., CH ₂), 1580 (N=N str.), 1251 (C-N str.), 790 (Ar-H bend.). |
| | ¹ H NMR (δ): | 8.0- 7.52 (m, 4H, ArH), 4.23 (s, 2H, N-CH ₂ -N), 3.55 (t, 4H, CH ₂ -N-CH ₂), 2.85 (s, 3H, N-CH ₃), 2.58 (t, 4H, CH ₂ -N(Me)CH ₂). |
| | MS (m/z) | 231 [M] ⁺ , 203 [M-N ₂] ⁺ , 202 [M-NCH ₃] ⁺ , 174 [M-C ₃ H ₇ N] ⁺ , 146 [M-C ₅ H ₁₁ N] ⁺ , 132 [M-C ₅ H ₁₁ N] ⁺ , 119 [C ₆ H ₅ N ₃] ⁺ , 113 [C ₆ H ₁₃ N ₂] ⁺ , 104 [C ₆ H ₄ N ₂] ⁺ . |
| 4a | IR (cm ⁻¹): | 3150 (C-H str., Ar-H), 2855 (C-H str., CH ₂), 1750, 1705 (C=O str.), 1072 (C-O str.), 790 (Ar-H bend.). |
| | ¹ H NMR (δ): | 7.89-7.54 (m, 4H, ArH), 4.46 (s, 2H, N-CH ₂ -N), 3.90 (t, 4H, CH ₂ -O-CH ₂), 2.65 (t, 4H, CH ₂ -N-CH ₂) |
| | MS (m/z) | 246 [M] ⁺ , 218 [M-C ₂ H ₄] ⁺ , 174 [M-C ₄ H ₈ O] ⁺ , 160 [M-C ₄ H ₈ NO] ⁺ , 147 [C ₈ H ₅ NO ₂] ⁺ , 146 [M-C ₂ H ₁₀ NO] ⁺ , 132 [C ₈ H ₄ O ₂] ⁺ , 100 [C ₅ H ₁₀ NO] ⁺ , 104 [C ₇ H ₄ O] ⁺ , 86 [C ₄ H ₈ NO] ⁺ . |
| 4b | IR (cm ⁻¹): | 3214 (C-H str., Ar-H), 2903 (C-H str., CH ₂), 1716, 1790 (C=O str.), 1203 (C-N str.), 749 (Ar-H) |
| | ¹ H NMR (δ): | 8.03- 7.56 (m, 4H, ArH), 4.31 (s, 2H, N-CH ₂ -N), 3.68 (t, 4H, CH ₂ -N-CH ₂), 2.80 (s, 3H, NCH ₃), 2.31 (t, 4H, CH ₂ -N(Me)CH ₂). |
| | MS (m/z) | 259 [M] ⁺ , 231 [M-C ₂ H ₄] ⁺ , 230 [M-NCH ₃] ⁺ , 202 [M-C ₃ H ₇ N] ⁺ , 174 [M-C ₅ H ₁₁ N] ⁺ , 160 [M-C ₅ H ₁₁ N ₂] ⁺ , 147 [C ₈ H ₅ NO ₂] ⁺ , 132 [C ₈ H ₄ O] ⁺ , 113 [C ₆ H ₁₃ N ₂] ⁺ , 140 [C ₇ H ₄ O] ⁺ . |
| 6a | IR (cm ⁻¹): | 3300 (N-H str.), 3086 (C-H str., Ar-H), 2850 (C-H str., CH ₂), 1725 (C=O str.), 1081 (C-O str.), 760 (Ar-H bend.). |
| | ¹ H NMR (δ): | 7.84 - 7.68 (m, 4H, ArH), 9.12 (s, 1H, NH), 4.30 (CH ₂ - N-CH ₂), 3.75 (t, 4H, CH ₂ -O-CH ₂), 2.95 (t, 4H, CH ₂ -N-CH ₂). |
| | MS (m/z) | 247 [M] ⁺ , 246 [M-H] ⁺ , 219 [M-C ₂ H ₄] ⁺ , 175 [M-C ₄ H ₈ O] ⁺ , 161 [M-C ₄ H ₈ NO] ⁺ , 147 [M-C ₅ H ₁₀ NO] ⁺ , 146 [M-C ₂ H ₁₀ NO] ⁺ , 119 [C ₇ H ₅ NO] ⁺ , 100 [C ₅ H ₁₀ NO] ⁺ , 86 [C ₄ H ₈ NO] ⁺ . |
| 6b | IR (cm ⁻¹): | 3321 (N-H str.), 3021 (C-H str., Ar-H), 2831 (C-H str., CH ₂), 1710 (C=O str.), 1213 (C-N str.), 757 (Ar-H bend.). |
| | ¹ H NMR (δ): | 7.72- 7.23 (m, 4H, ArH), 9.15 (s, 1H, NH), 4.38 (s, 2H, N-CH ₂ -N), 3.71 (t, 4H, CH ₂ -N-CH ₂), 2.72 (s, 3H, NCH ₃), 2.37 (t, 4H, CH ₂ -N(Me)-CH ₂). |
| | MS (m/z) | 260 [M] ⁺ , 259 [M-H] ⁺ , 232 [M-C ₂ H ₄] ⁺ , 231 [M-NCH ₃] ⁺ , 203 [M-C ₃ H ₇ N] ⁺ , 175 [M-C ₅ H ₁₁ N ₂] ⁺ , 161 [M-C ₅ H ₁₁ N ₂] ⁺ , 147 [C ₈ H ₅ NO ₂] ⁺ , 132 [C ₈ H ₄ O] ⁺ , 119 [C ₇ H ₅ NO] ⁺ , 113 [C ₆ H ₁₃ N ₂] ⁺ . |
| 8a | IR (cm ⁻¹): | 3056 (C-H str. Ar-H), 1777 (C=O str.), 1595 (C=N str.), 1122 (C-O str.), 738 (Ar-H bend.). |
| | ¹ H NMR (δ): | 4.55 (s, 2H, CH ₂), 4.25 (s, 2H, N-CH ₂ -N), 375 (t, 4H, CH ₂ -O-CH ₂), 2.43 (t, 4H, CH ₂ -N-CH ₂), 1.88 (s, 3H, CH ₃). |
| | MS (m/z) | 197 [M] ⁺ , 182 [M-CH ₃] ⁺ , 169 [M-CO] ⁺ , 156 [M-C ₂ H ₃ N] ⁺ , 125 [M-C ₄ H ₈ O] ⁺ , 111 [M-C ₆ H ₈ NO] ⁺ , 100 [C ₅ H ₁₀ NO] ⁺ , 97 [M-C ₅ H ₁₀ NO] ⁺ , 86 [C ₄ H ₈ NO] ⁺ . |
| 8b | IR (cm ⁻¹): | 3070 (C-H str., Ar-H), 1750 (C=O str.), 1610 (C=N str.), 1225 (C-N str.), 745 (Ar-H bend.). |
| | ¹ H NMR (δ): | 4.55 (s, 2H, CH ₂), 4.31 (s, 2H, N-CH ₂ -N), 3.80 (t, 4H, CH ₂ -N-CH ₂), 2.67 (s, 3H, CH ₃), 2.30 (t, 4H, CH ₂ -N(Me)CH ₂). |
| | MS (m/z) | 210 [M] ⁺ , 195 [M-CH ₃] ⁺ , 182 [M-CO] ⁺ , 181 [M-NCH ₃] ⁺ , 169 [M-C ₂ H ₃ N] ⁺ , 153 [M-C ₃ H ₇ N] ⁺ , 113 [C ₆ H ₁₃ N ₂] ⁺ , 111 [M-C ₅ H ₁₁ N ₂] ⁺ . |



EXPERIMENTAL SECTION

General Procedure

All the reactions were carried out in a domestic microwave oven (Kenstar, Model No. OM-26 EGO, Power-1200W). Melting points were determined in open capillaries. Reactions were monitored by thin layer chromatography using silica gel-G as adsorbent. IR spectra (KBr pellets) were recorded on Perkin-Elmer 1800 (FTIR) spectrometer. ¹H NMR spectra (CDCl₃) were taken on a Bruker DRX spectrometer (300 MHz FTNMR) using TMS as internal standard and chemical shift are expressed in *d* (ppm). Mass spectra were taken on JEOL SX-102/PA-6000 (EI) spectrometer.

1H- benzotriazole (1):

o-phenylenediamine (0.01 mol) was dissolved in a mixture of glacial acetic acid (0.2 mol) and water (30.0 mL). The solution was cooled to 15°C and then a solution of sodium nitrite (0.11 mol) in water (15.0 mL) was added in lot to the stirred solution of *o*-phenylenediamine. The reaction mixture was irradiated in microwave oven for 20-30 sec. It attains temperature of 80-85 °C. It was cooled at room temperature and chilled in ice water for 30 min. The

resulting solid was separated, filtered and washed with ice water, dried and recrystallized from benzene (m.p.100 °C).

1-[(Morpholin-4-yl/4-methylpiperazin-1-yl)methyl]-1H-benzotriazole (2a & 2b):

Compound (1) (0.001 mol) was dissolved in ethanol and acidic alumina (5.0 g) was added to it. The slurry formed was kept in an oven at 60°C for 1 hour. After cooling, formaldehyde (0.003 mol) and morpholine (0.001 mol) were added to the reaction mixture. It was exposed to microwave for 9 min. Single spot in TLC indicated completion of reaction and product was separated from alumina using methanol. It was recrystallized from methanol to give compound (2a). Compound (2b) was synthesized using N-methylpiperazine under similar reaction conditions.

1 H - Isoindol- 1,3- (2H)- dione (3):

The mixture of phthalic anhydride (0.01 mol) and urea (0.01 mol) was grinded thoroughly in mortar. The mixture was transferred to beaker and few drops of DMF were added to it. The reaction mixture was irradiated in microwave oven for 15 seconds. After cooling of reaction mixture, 50 mL

cold water was added to it. A white crystalline solid was formed. It was filtered, washed with water, dried and recrystallized from ethanol (m.p. 235 °C).

2-[(Morpholin-4-yl/4- methylpiperazin-1-yl) methyl]-1-H- isoindol-1,3 (2H) - dione (4a & 4b):

Compound (3) (0.001 mol) was dissolved in absolute alcohol and acidic alumina (5.0 g) was added to it. It was kept in oven at 60°C. After cooling, formaldehyde (0.003 mol) and morpholine (0.001 mol) were added to it. The reaction mixture was exposed under microwave irradiation for 2 minutes. The target product was separated from acidic alumina using mixture of methanol and conc. HCl. It was recrystallized from methanol to give compound (4a). Compound (4b) was synthesized by similar method using N-methylpiperazine in place of morpholine.

3,4-Dihydroquinoxalin-2- (1H)- one (5):

Chloroacetic acid (0.001 mol) neutralized with 1N NaOH solution was added to o-phenylenediamine (0.001 mol) solution and the reaction mixture was irradiated for 2.30 min. in microwave oven. The reaction mixture was cooled for 4-5 hrs at room temp., brown colored solid was separated which was recrystallized from ethanol (m. p. 75 °C)

4-[Morpholin-4-yl/4-methylpiperazin-1-yl)methyl]-3,4-dihydroquinoxalin-2- (1H)- one (6a & 6b):

Compound (5) (0.001 mole) was dissolved in ethanol and acidic alumina (5.0 g) was added to it. It was kept in oven for 1 hour. Then, formaldehyde (0.003 mol) and morpholine (0.001 mol) were added to it and exposed to microwave irradiation for 4 minutes. When single spot appeared on TLC plate, the product was separated from alumina using boiling water. It was recrystallized from hot water to give compound (6a). Compound (6b) was synthesized by similar method using N-methylpiperazine in place of morpholine.

5- Methyl-2, 4-dihydro-3H-pyrazol-5-one (7):

A mixture of ethyl acetoacetate (0.04 mol) and hydrazine hydrate (0.04 mol) was irradiated in microwave oven for 2 seconds. The reaction mixture was cooled and ether (10 mL) was added to it. The mixture was stirred to give a solid product, which was filtered, washed with ether and crystallized from ethanol (m.p. 216 °C).

5-Methyl-2-(morpholin-4-yl/4-methylpiperazin-1-yl) methyl-2, 4-dihydro-3H-pyrazole-3-one (8a & 8b):

Compound (7) (0.01 mol) was dissolved in ethanol and acidic alumina (5.0 g) was added to it. The slurry was kept in oven at 60°C for 1 hour and cooled. Then, formaldehyde (0.03 mol) and morpholine (0.01 mol) were added to it and the reaction mixture was exposed to microwaves. Completion of the reaction was observed by TLC technique, the product formed was separated using mixture of methanol and conc. HCl to give compound (8a). Compound (8b) was synthesized similarly using N-methylpiperazine in place of morpholine.

BIOLOGICAL SCREENING

All synthesized Mannich bases were screened *in vitro* for their antimicrobial activities against two strains of bacteria (*Bacillus cereus* and *Escherichia coli*) and two strains of

fungi (*Candida albicans* and *Aspergillus fumigatus*) by the Cup or Well²¹ using DMF as a solvent at various dilutions of synthesized compounds. Commercial antibacterial drug Ciprofloxacin and antifungal drug Amphotericin - B were also screened under similar conditions for comparison. The diameter of inhibition zones (mm) were measured and recorded. The results have been tabulated in the form of minimum inhibitions concentration of drug in Table (3). From the data reported in Table (3) it could be concluded that the synthesized compounds exhibit excellent activity against both bacterial strains. Evaluation of MIC values lead us to conclude that among synthesized Mannich bases **2a**, **4b**, **6b**, and **8a** exhibit excellent activity against both *B. cereus* and *E. Coli*, while others exhibit moderate to good activity. Among the all synthesized Mannich bases **2a**, **4a**, **4b**, **6b** and **8a** exhibits good activity against both *C. albicans* and *A. fumigatus*, while others exhibit moderate to good activity.

Table 3: MIC (u g / m L) of synthesized compounds

| Compound | Antibacterial activity | | Antifungal activity | |
|-----------------------|------------------------|----------------|---------------------|---------------------|
| | <i>B. cereus</i> | <i>E. coli</i> | <i>C. albicans</i> | <i>A. fumigatus</i> |
| 2a | 0.08 | 0.017 | 0.53 | 1.29 |
| 2b | 0.10 | 0.019 | 0.96 | 1.09 |
| 4a | 0.18 | 0.026 | 0.69 | 1.28 |
| 4b | 0.06 | 0.015 | 0.60 | 1.09 |
| 6a | 0.08 | 0.037 | 1.36 | 1.28 |
| 6b | 0.05 | 0.021 | 0.58 | 1.48 |
| 8a | 0.07 | 0.012 | 0.57 | 1.44 |
| 8b | 0.08 | 0.060 | 0.67 | 1.67 |
| Ciprofloxacin | 0.06 | 0.010 | | |
| Amphotericin-B | - | - | 0.50 | 1.00 |

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BIBLIOGRAPHY

1. (a) Mingas D. P. and Baghurst D. R., *Chem. Soc. Rev.*, **20**, 1, (1991).
(b) Abrakmvoich R. A., *Org. Prep Proceed. Int.*, **23**, 685 (1991).
2. Denon J. J., Turner R., Neir W. B. and Lawson V. A., *J. Am. Chem. Soc.*, **71**, 2048, (1949).
3. Burckhalter J. P. and Johnson S. H., *J. Am. Chem. Soc.*, **73**, 4835, (1951).
4. Mannich C. and Lammering D., *Chem. Ber.*, **55**, 3510, (1922).
5. Levvy G. A. and Nisbet H. B., *J. Chem. Soc.*, 1053, (1938).
6. Blanton C. D. and Nobles W. L., *J. Pharma Sci.*, **51**, 878 (1962).
7. Rogers F. C. and Nobles W. L., *J. Pharma Sci.*, **51**, 273 (1962).
8. Caldwell H. C. and Nobles W. L., *J. Am. Pharma Asst. Sci. Ed.*, **43**, 644 (1954).
9. Hellmann H. and Loschmann I., *Chem Ber.*, **87**, 1664, (1954).
10. Moore M. B. and Rapela R. T., *J. Am. Chem. Soc.*, **68**, 1657, (1946).
11. Atkinson R. O., *J. Chem. Soc.*, 1329, (1954).
12. Ger. Pet., **575**, 114, (1954).
13. Swaminathan S. and Narsimhan K., *Ber.*, **99**, 889, (1966).
14. Burekhulter J. H., Wells J. N. and Mayer W. J., *Tetrahedron Lett.*, 1353, (1964).
15. Bryant G. and Heisey L. V., *J. Am. Chem. Soc.*, **68**, 2486, (1946).
16. Parmar S. S., Mishra R. S., Chaudheri A. and Gupta T. K., *J. Pharm. Sci.*, **61**, 1322, (1972).
17. El-Marry A. H., Fahmy H. H. and Abdelwahed S. H., *Molecules*, **5**, 1429, (2000).
18. Tramontini M., *Synthesis*, **12**, 707, (1973).
19. Pascal C., Giron C. and Froissant J., *Fr. Pat. Appl.*, 2593181, (1973).
20. Dianov V. M., Chikaeva I. G. and Timirkhanov G. A., *Khim. Farm. Zh.*, **28**, 21, (1994).
21. Collee G. J., Fraser G. A., Mamion P. B. and Suninous A., *Practical Medical Microbiology*, 14th ed.; Churchill Luingstone, Edinburgh: **Vol. II**, 163, (1996).