World Journal of Pharmaceutical and Life Sciences
WJPLS
www.wjpls.org
SJIF Impact Factor: 6.129

# SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF A NEW MANNICH BASE: 2, 3`-DIHYDROXY-3-PYRROLIDINOMETHYLCHALCONE

**Abdel Karim M.[1]*, Minas M.[2] and Mai Mekki[1]**

[1]Sudan University of Science ans Technology, Faculty of Science.
[2]Intarnational University of Africa, Faculty of Pharmacy.

**Corresponding Author: Abdel Karim M.**
Sudan University of Science ans Technology, Faculty of Science.

**Article Received on 03/03/2022** **Article Revised on 23/03/2022** **Article Accepted on 13/04/2022**

## ABSTRACT

Flavonoids form a family of well known natural products present in most of the plant families. The structural variations of the flavonoids are associated with different biological and pharmacological activities, including anticancer, antioxidant, antibacterial, antifungal and antiviral activity, beside cardiovascular and hepatic protection. The biological activity of Mannich bases is well known. In this study a flavonoid substituted by a Mannich side chain - 2,3`-dihydroxy-3-pyrrolidinomethylchalcone – has been synthesized by the Mannich reaction of 2,3`-dihydroxychalcone with pyrrolidine. The intermediate flavonoid - 2,3`-dihydroxychalcone-was synthesized by the reaction of 3-hydroxyacetophenone with salicylaldehyde. The flavonoid: 2,3`-dihydroxychalcone(I) and its Mannich base:2,3`-dihydroxy-3-pyrrolidinomethylchalcone(II) were evaluated for antimicrobial activity against six standard human pathogens. Compound (I) exhibited significant activity against all test organisms at 200 and 100mg/ml. It also showed significant activity at 50mg/ml against all test organism –except for *Bacillus subtilis*. At a concentration of 200mg/ml, compound (II) showed significant activity against all test organisms with the exception of *Bacillus subtilis*.

**KEYWORDS**: Synthesis, New Mannich base, Antimicrobial Activity.

## INTRODUCTION

Flavonoids form a family of well known natural products present in most of the plant families. More than 8000 different flavonoids have been isolated from their natural source to date. The structural variations of these flavonoids are associated with many different biological and pharmacological activities, including anticancer activity, protection against cancer formation (chemo-protection), antioxidant activity, cardiovascular and hepatic protection, antibacterial, antifungal and antiviral activity. Flavonoids have also been reported to play an important role in hormone-related female diseases, such as breast cancer and menopausal syndrome. Natural flavonoids have therefore been subjected to many chemical modifications in order to improve their activity.[1,2,3]

Flavonoids appear to provide defense against fungal infection.[4] In addition to pollen germination, fertilization, and seed set, flavonoids also function in the attraction of animal pollinators.[5,6] In plants, flavonoids are inducible and constitutive components of the defense mechanism against infection.[7,8] Flavonoids have long been recognized to possess anti-inflammatory, antioxidant, ant-allergic, antimicrobial and hepato-protective properties.[9-13]

## MATERIALS AND METHODS

### Materials

### Test organisms

The synthesized compounds were screened for antibacterial and antifungal activities using the standard microorganisms shown in Table (1).

### Chemicals and Solvents

Analytical grade reagents (Table 2) were used. They were purchased from Sigma – Aldrich company (UK).

### Instruments

The UV spectra were recorded on a Perkin-Elmer Lambda 2 UV-Visible Spectrophotometer. Infra red spectra were measured on a Perkin-Elmer1310 Infra red Spectrophotometer. $^1$HNMR were recorded on EM-360 NMR Spectrophotometer. Mass spectra were measured on a Krates MS 80 RF Mass Spectrometer.

**Table 1: Test organisms.**

| No | Micro organism | Type |
|---|---|---|
| 1 | *Bacillus subtilis* | G+ve |
| 2 | *Staphylococcus aureus* | G+ve |
| 3 | *Pseudomonas aeroginosa* | G-ve |
| 4 | *Escherichia coli* | G-ve |
| 5 | *Aspergillusniger* | fungus |
| 6 | *Candida albicans* | fungus |

**Table 2: Chemicals and solvents.**

| No. | Chemical |
|---|---|
| 1 | Absolute ethanol |
| 3 | Formalin |
| 7 | Pyrrolidine |
| 8 | 2-Hydroxyacetophenone |
| 9 | Salicylaldehyde |

**Methods**

**Synthesis protocols**

**Synthesis of 2,3`-dihydroxychalcone(I)**

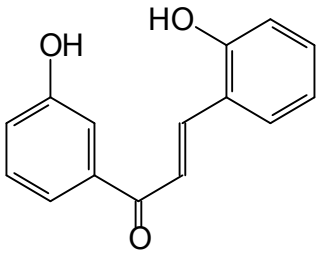
3-Hydroxyacetophenone (2.72g, 20mmol) was added to salicylaldehyde (2.44g,20mmol) in 20% ethanolic sodium hydroxide (20ml). The mixture was stirred for 24 hours at room temperature and left overnight. Removal of the solvent under reduced pressure gave the product.

**Synthesis of 2,3`-dihydroxy-3-pyrrolidinomethylchalcone (II)**

Formalin (0.8g,10mmol) was added dropwise with stirring to a mixture of compound (I) (2.39gm,10mmol) and pyrrolidine (1.7gm,10mmol) in absolute ethanol (10ml) at $0^0$C .The mixture was stirred for 1 hour and left overnight. Removal of the solvent in *vacuo* gave the product.

A Mannich base: 2,3`-dihydroxy-3-pyrrolidinomethylchalcone(II) has been synthesized via a Mannich reaction of 2,3`-dihydroxychalcone with pyrrolidine and formalin. The intermediate flavonoid: 2,3`-dihydroxychalcone(I) was synthesized by the reaction of 3-hydroxyacetophenone with salicylaldehyde. The structures of the Mannich base(II) and the intermediate flavonoid(I) have been characterized by spectral tools (UV,IR ,$^1$HNMR).

**Compound I (2,3`-dihydroxychalcone)**




(I)

The IR spectrum of (I) (Fig. 1) gave $\upsilon$ (KBr) 646,808, 865 (C-H, Ar bending), 1413 (C= C Ar), 1664 (C=O) and 3427 (OH stretching). The UV spectrum of compound (I) showed (Fig.2) $\lambda_{max}$ (MeOH) 322,374 nm which is due to a carbonyl chromophore extended by phenyl and hydroxyl auxochromes.

The $^1$HNMR spectrum of (I) gave(Fig.3) a signal at δ3.45 assigned for two vinylic protons (overlapping the solvent (DMSO) residual water protons).The multiplet at δ6.8 – 7.8 is due to the aromatic protons.The resonance at δ2.50 is due to solvent residual protons.

**RESULTS AND DISCUSSION**

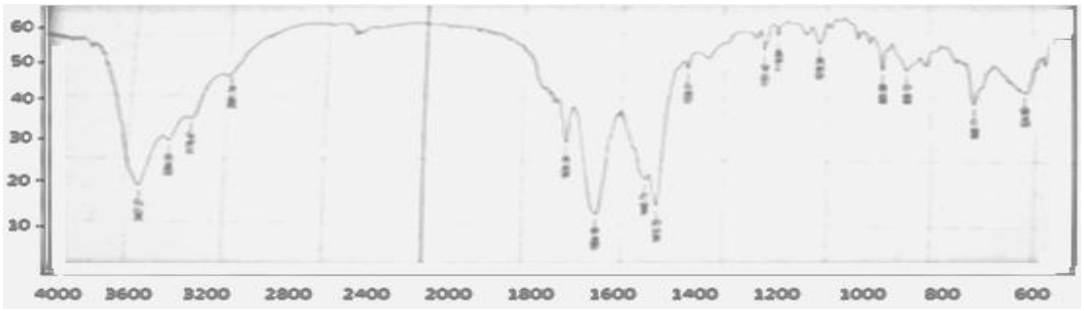



**Fig. 1: IR spectrum of compound (I).**

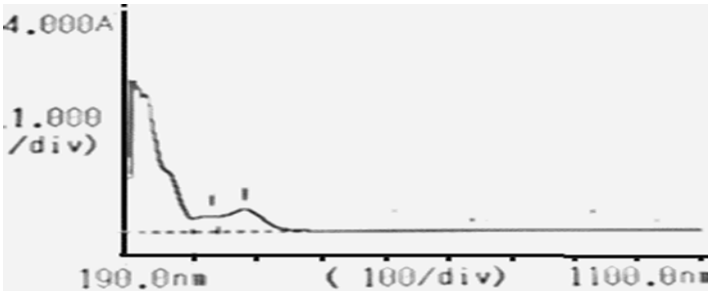
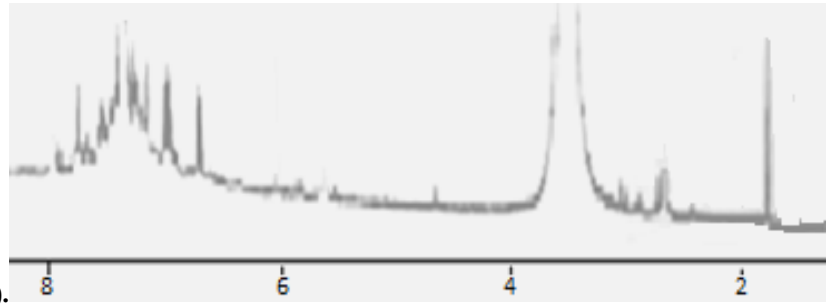


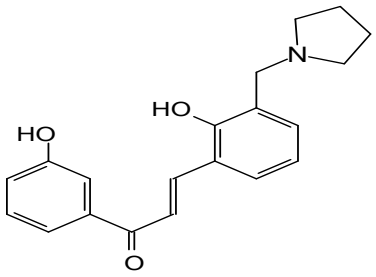

**Fig. 2: UV spectrum of the compound.**




(I).

**Fig. 3: $^{1}$HNMR spectrum of the compound (I).**

**Compound II (2,3`-dihydroxy-3-pyrrolidinoinomethylchalcone)**




**(II)**

The IR spectrum of (II) (Fig. 4) gave $\upsilon$ (KBr) 678,754,864 (C-H, Ar. bending), 1253 (C-N), 1482 (C=C, Ar), 1652 (C=O), and 3386 (OH stretching). The UV spectrum showed (Fig.5) $\lambda_{max}$ (MeOH) 319nm which corresponded to a carbonyl chromophore extended by a phenyl and hydroxyl auxochromes.

The $^{1}$HNMR spectrum of compound II showed (Fig.6) a resonance at δ3.62 assigned for five methylene moieties. The aromatic protons resonated at δ6.2, 7.1 and 7.2ppm. The mass spectrum (Fig. 7) gave the ion m/z 325 corresponding: $M^{+}$+2.

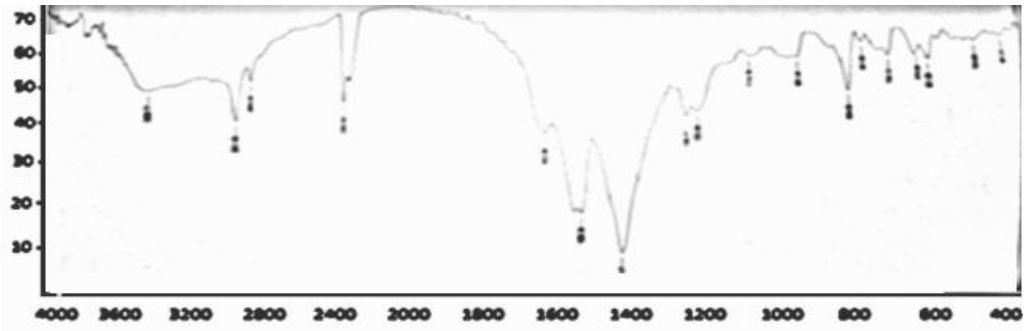



**Fig. 4: IR spectrum of compound (II).**

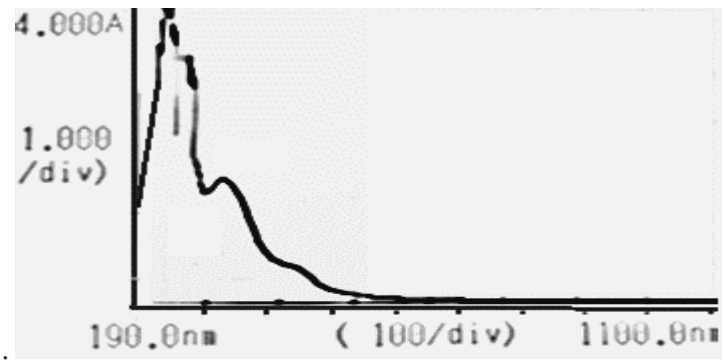



**Fig. 5: UV spectrum of compound II.**

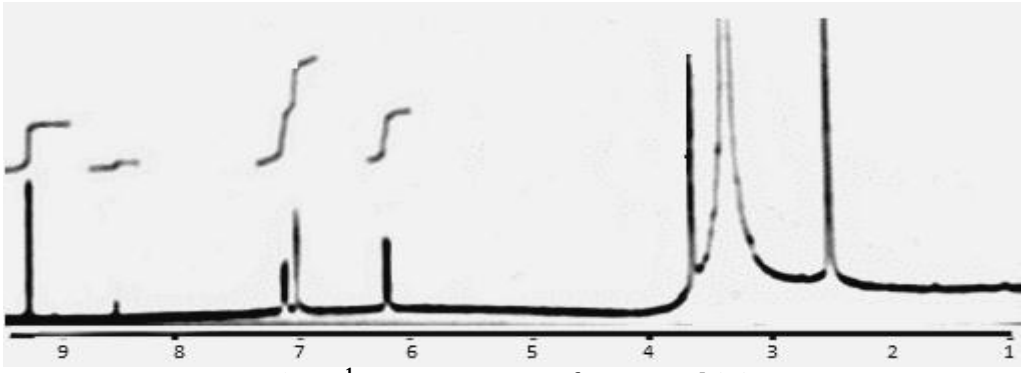



**Fig. 6: $^{1}$HNMR spectrum of compound (II).**

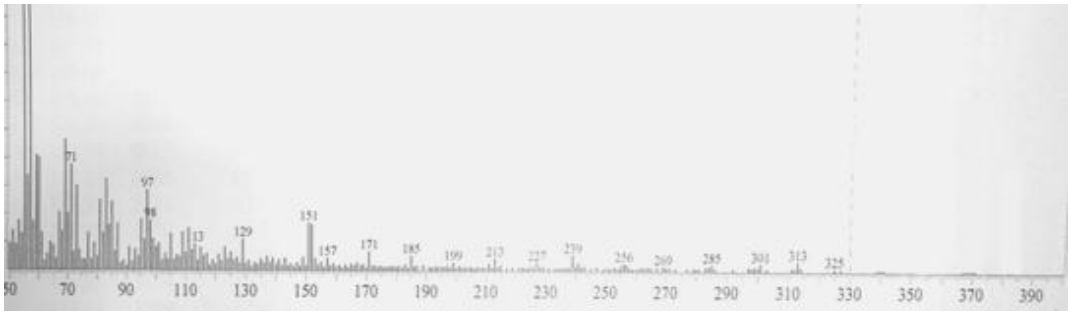



**Fig. 7: Mass spectrum of the compound (II).**

**Antimicrobial activity**

The synthesized compounds (I and II) were screened for their antimicrobial activity against six standard human pathogens. The averages of the diameters of the growth inhibition zones are shown in Table (3).The results were interpreted in commonly used terms: less than 9mm(inactive); 9 -12mm(weak);13-18(active); more than 18(very active).Tables (4) and (5) represent the antimicrobial activity of standard antibacterial and antifungal chemotherapeutic agents against standard bacteria and fungi respectively.

Compound (I) exhibited significant activity against all test organisms at 200 and 100mg/ml. It also showed significant activity at 50mg/ml against all test organism – except for *Bacillus subtilis*. At a concentration of 200mg/ml, compound (II) showed significant activity against all test organisms with the exception of *Bacillus subtilis*.

**Table 3: Inhibition zones (mm/mg sample) of compounds I and II.**

| Sample | Conc. mg/ml | Ec. | Pa. | Bs. | Sa. | Ca. | An. |
|---|---|---|---|---|---|---|---|
| Compound I | 200 | 28 | 24 | 22 | 21 | 25 | 24 |
| | 100 | 25 | 21 | 20 | 19 | 23 | 22 |
| | 50 | 21 | 18 | 16 | 17 | 21 | 18 |
| Compound II | 200 | 17 | 17 | 15 | 17 | 20 | 17 |
| | 100 | 16 | 17 | 15 | 17 | 18 | 15 |
| | 50 | 16 | 16 | 14 | 16 | 16 | 14 |

**Table 4: Inhibition zones (mm/mg sample) of standard antibacterial agents.**

| Drug | Conc. mg/ml | B.s. | S.a. | E.c. | P.a. |
|---|---|---|---|---|---|
| Ampicillin | 40 | 15 | 30 | - | - |
| | 20 | 14 | 25 | - | - |
| | 10 | 11 | 15 | - | - |
| Gentamicin | 40 | 25 | 19 | 22 | 21 |
| | 20 | 22 | 18 | 18 | 15 |
| | 10 | 17 | 14 | 15 | 12 |

**Table 5: Inhibition zones (mm/mg sample) of standard antifungal agent.**

| Drug | Conc. mg/ml | A.n | C.a |
|---|---|---|---|
| Clotrimazole | 30 | 22 | 38 |
| | 15 | 17 | 31 |
| | 7.5 | 16 | 29 |

- S.a*: Staphylococcus aureus*
- E.c*: Escherichia coli*
- P.a: *Pseudomonas aeruginosa*
- A.n: *Aspergillus niger*
- C.a: *Candida albicans*
- S.t: *Salmonella typhi*
- *B.a: Bacillus subtilis*

## REFERENCES


1. Amalesh, S., Gouranga, D., *International Journal of Pharmaceutical Science and Technology*, 2011; 6: 525.
2. Hamdoon, M. "Natural and synthetic flavonoids with potential antioxidant and anticancer activities", Ph.D. Thesis, Qassim University, 2009.
3. Erich, Gr., "The Science of Flavonoids"Vol.11, The Ohio State University Columbus, Ohio, USA, P402, 2006.
4. Mcnally, J., Wurms, V., *Physiological and Molecular Plant Pathology*, 2013; 68: 293.
5. Clegg, M., Durbin, M., *Proceeding of the National Academy Sciences*, 2000; 97: 7016.
6. Thompson, W., Meinwald, J., *American Journal of Science*, 1972; 177: 528.
7. 27) Dixon, R., Paiva, N., *The Plant Cell*, 1995; 7: 1085.
8. Kajiya, K., Hojo, H., *Journalof Agricultural and Food Chemistry*, 2010; 52: 3535.
9. 35) Knekt, R., Jarvinen, A., *British Medical Journal*, 2008; 4: 481.
10. Balant, M., Wermeille, L., *European Journal of Drug Metabolism and Pharmacokinetics*, 2013; 37*:* 1520.
11. Ferenci, B., Dragosics, H., *Journal of Hepatology*, 2012; 60: 1113.
12. Cristián, E., Lamirande, C., *Free Radical Biology and Medicine*, 2012; 41: 528.
13. Cristián, E., Lamirande, C., *Free Radical Biology and Medicine*, 2012; 41: 528.
14. Wardman, P., *Journal of Physical Chemistry Reference Data*, 2016; 118: 6370.
15. Tsajimoto, H., Hashizume, M., *International Journal of Biochemistry*, 2002; 25: 491.