Phytochemical Screening and Antibacterial Activity of Moringa oleifera Leaf Extract Western Libya

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Abstract

Moringa oleifera (M. oleifera) is one of the Moringaceae family. M. oleifera plant parts retain a precious pharmacological effect. Its leaves are utilized commonly because of having enormous nutrients as nutraceuticals. Moreover, its compresses various effects such as biological and antimicrobial effect as antibacterial or antifungal. Additionally, M. oleifera investigation was carried out either in vivo or in vitro to conclude that their leaves component has pharmacological effect. This current study had confirmed therapeutic effect of Moringa Oleifera leaf extract and its antibacterial activity. There is critical need for more advanced research about use and pharmaceutical effects of Moringa Oleifera leaves in improvement of various drugs and useful foods. This study aimed to investigate phytochemical ingredients and antibacterial effect of Moringa Oleifera leaf extract and the uses of Moringa Oleifera leaf extract either traditionally or pharmacologically concluded.

Keywords: Moringa oleifera, Antibacterial activity, Phytochemical screening, Libya.

INTRODUCTION

The drumstick tree, or Moringa oleifera Lam., is a member of the Moringaceae family and is extensively dispersed in various regions, including tropical and desert nations, as well as in India and Africa. It was primarily used for food and medicine (Al_husnan & Alkahtani, 2016). Moringaoleifera has numerous qualities that may be used for food, medicine, and other industrial and home applications (Falowo et al., 2018; Padayachee & Bajinath, 2020), particularly its leaves, which may be eaten raw, roasted, or dried and kept for a long time without losing any of their nutritional value. In addition to using the leaves for food and feed, they may also be used as functional foods or nutraceuticals since they naturally contain phytochemicals such as flavonoids, carotenoids, and glucosinolates (Amaglo et al., 2010; Sultana et al., 2009). The main phytochemicals found in moringa leaves are astragalin, isoquercetin, and crypto-chlorogenic acid, which have anti-oxidant, anti-hypertension, and anti-inflammation characteristics (Verma et al., 2009; Vongsak et al., 2012).

Based on the bioactive components and their antioxidant activity, several in vitro experiments have largely supported the biological activity of these plant extracts (Leone et al., 2015; Sultana et al., 2009). Its strong antioxidant properties are principally brought on by its high phenolic content. Due to these therapeutic advantages, several pharmaceutical preparations from this plant have been manufactured and sold in both the Indian and global markets.(Mehra et al., 2020; Sahakitpichan et al., 2011) Different sections of M. oleifera have antibacterial potentials that can be used to purify...
water, eliminate biofilm threats, and eradicate pathogenic microbes. The long-term use of *M. oleifera* and its ethno-pharmacological characteristics have proven its safety. *M. oleifera* has also been employed for bio-enhancement and as nanoparticles in medication administration in addition to its antibacterial action (Arora et al., 2013). Fidrianny et al. study mentioned that *M. oleifera* is commonly named Sanjana and drumstick. The name Moringa originated from murungai; a Tamil word that means twisted pod (Dhakad et al., 2019; Fidrianny et al., 2021) study illustrated that the cultivation of Moringa has different reasons according to its nutritional content or medicinal uses (Djemoui et al., 2019). *M. oleifera* has medicinal effects documented by (Djemoui et al., 2019). study, such as hypoglycemic effect, antioxidant, antibacterial, antifungal, and wound healing due to anti-inflammatory activity (Djemoui et al., 2019). (Amaglo et al., 2010) reported that the leaves, in particular, can be used in a salad, roasted, or kept as dry powder for a prolonged time without losing nutritive constituents (Amaglo et al., 2010).

Moringa leaves were reported by (Maheshwari et al., 2014). to have elevated amounts of protein that could be utilized as food. Also, they mentioned that *Moringa oleifera* contains a high amount of provitamin A. (Tshabalala et al., 2020). traced the source of the cardiovascular benefit and body energizing to the omega-3 and omega-6 polyunsaturated fatty acid content of Moringa. It can be used for obese patients due to low calorific value (Kashyap et al., 2022; Maheshwari et al., 2014)

They also illustrated calcium contents and digestible protein that is higher than milk. (Kasolo et al., 2010). mentioned moringa leaves’ high content of potassium, magnesium, and copper. Iron content is 28 times of spinach, proved (Fuglie, 1999; Kasolo et al., 2010). Interestingly, Mbikay approved that fat-soluble vitamins like D and E and water-soluble like folic acid, nicotinic acid, and vitamin care also exist in *M. oleifera* (Mbikay, 2012). Stevens et al. mentioned that leaves are not toxic when taken in large amounts, so it is found to be safer and healthier for consumption (Stevens et al., 2013). Bacterial species develop drug resistance to common antibacterial agents. So, medicinal plants such as Moringa leaves could be used as alternative treatments for infectious diseases (Luqman et al., 2012).

Rios & Recio’s studies reported the need for more studies on the antibacterial activity of essential oils or compounds like alkaloids, flavonoids, diterpenes, and triterpenes to justify the use of medicinal plants (Rios & Recio, 2005), while Abdallah mentioned that metabolites of *Moringa oleifera* leaves, such as alkaloids, flavonoids, and saponins have wonderful pharmacological effects as antibacterial and anti-microbial properties and interestingly as anticancer (Berkovich et al., 2013). study (Abdallah, 2011; Berkovich et al., 2013).

**MATERIALS AND METHODS**

**Collection and Identification of Plant Material**

*Moringa oleifera* tree leaves were collected. The leaves were washed under distilled water for dust and foreign particle removal. After that, the leaves were air-dried in an oven at 60ºC. The dried leaves were added to a mixer to be ground into fine powder. The fine powder was stored in a refrigerator to avoid heat and light exposure and kept for further use.

**Preparation of Plant Material**

About 10g of the fine powdered *Moringa oleifera* leaves were suspended in 100 mL of ethanol and shaken well. The suspensions were kept at room temperature for 24 hours. The suspensions were then filtered. The solvent was removed by heat treatment at 80 ºC to concentrate the extract. The extracts were stored in a refrigerator at 4ºC for further analysis (Abadallah & Ali, 2019).
Phytochemical Screening
The collected plant extracts were subjected to qualitative phytochemical analysis for identification of various bioactive chemical constituents (alkaloids, volatile oils, saponins, flavonoids, steroids, tannins, and terpenoids) conducted using standard laboratory methods as described by (Edeoga et al., 2006; Sofowora, 1993).

Test Isolates
Clinical *Staphylococcus aureus, Streptococcus pyogenes,* and *Pseudomonas aeruginosa* isolates were collected for testing the antibacterial activity of *Moringaoleifera*. As described by Cheesbrough, gram staining and microbiological analysis were used in recognizing the isolates by utilizing differential media and biochemical tests.

Antibacterial activity testing
0.1 ml of each organism culture was inoculated onto the plates’ surface by a sterile swab stick. The plates were incubated for 24 hours at 37ºC, after which the inhibition zones were calculated using a meter ruler.
All the experiments were repeated three times, and the mean was recorded.

RESULTS

Phytochemical Screen
1. Tannins: A green color and gelatinous precipitate resulted upon adding 1 ml of 1% Lead acetate to 1 ml of the moringa leaf extract in a test tube. This result confirmed the existence of tannins.
2. Saponins: Durable white precipitate resulted when 1-3 ml of mercury chloride was added to 5 ml of leaf extract which was judged as an indicator of saponins existence.
3. Flavonoids: A yellow shade that signified flavonoids resulted when moringa leaf extract was added to diluted ammonia and concentrated sulphuric acid.
4. Terpenoids (Salkowskitest): No brown color resulted when 1 ml of leaf extract was added to chloroform, one drop of concentrated sulphuric acid, and one drop of anhydrous acetic acid.
5. Resins: Turbidity resulted when the leaf extract was added to the ethyl alcohol (95%) solution.
6. Volatile oils: A shiny pink layer on filter paper formed after UV light exposure after the filtration of leaf extract, indicating volatile oils’ content.
7. Alkaloids: A grey color confirmed alkaloids’ presence when adding leaf extract to 2 ml of Marquis Reagents.
8. Glycosides: A few drops of Fehling reagent were added to 1 ml of leaf extract.
(Table 1) lists the phytochemical components of moringa leaf extracts.

Table: (1). Illustrate the phytochemical constituent of moringa leaf extract.

<table>
<thead>
<tr>
<th>Phytochemical constituents</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>++</td>
</tr>
<tr>
<td>Saponins</td>
<td>+</td>
</tr>
<tr>
<td>Tannins</td>
<td>+++</td>
</tr>
<tr>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>Terpenoids</td>
<td>-</td>
</tr>
<tr>
<td>Resins</td>
<td>+++</td>
</tr>
<tr>
<td>Volatile oils</td>
<td>+++</td>
</tr>
<tr>
<td>Glycosides</td>
<td>++</td>
</tr>
</tbody>
</table>

Symbol: +=Existence of phytochemical, -=Absence of phytochemical. (+++)high, (++) medium, (+) poor, (-)no found
The results indicate the presence of saponins, resins, tannins, alkaloids, flavonoids, glycosides and volatile oils, and the absence of terpenoids.

**Antibacterial Activity:**
The antibacterial activity of *Moringa Oleifera* leaf extract is summarized in (Table 2). The inhibition zones emphasized that it depends on the type of bacteria and concentration of *Moringa Oleifera* leaf extract. The highest zone of inhibition is demonstrated by *Pseudomonas aeruginosa* (17 mm) in concentrated extract and (14mm) in diluted extract.

Interestingly, the case of *Staphylococcus aureus* and *Streptococcus pyogenes* reported zero results which indicates their negative results and the antibacterial activity of *Moringa Oleifera* leaf extract.

| Table: (2). Demonstrate the antibacterial activity of Moringa Oleifera leaves extract |
|---------------------------------|---------------------------------|
| Name               | Result  | |
| Staphylococcus aureus       | Concentrated | 0 |
|                       | Diluted      | 0 |
| Streptococcus pyogenes     | Concentrated | 0 |
|                       | Diluted      | 0 |
| Pseudomonasaeruginosa     | Concentrated | 17 |
|                       | Diluted      | 14 |

**DISCUSSION**
Chhetri et al. study reported that bioactive constituents can be taken from plant parts like seeds, roots, stems, leaves, and flowers (Chhetri et al., 2008). The results of our study showed the existence of bioactive components in moringa leaf extracts. Our study revealed the presence of alkaloids, volatile oils, saponins, resins, etc., in the ethanol extract of the leaves as in (Table 1). The outcome of the current study is in a parallel line with previous studies that screened the phytochemical composition of moringa leaves. Moringa leaves were proven to include numerous phytochemicals (Edeoga et al., 2006; Madziga et al., 2010).

The phytochemicals have remarkable pharmaceutical effects that are used in drugs for treating sclerosis, diabetes, skin antiseptic, diarrhea, colitis, and cancer. (Aboaba et al., 2011; Madziga et al., 2010). studies illustrated that Alkaloids, for example, are extensively used as anticancer agents, aesthetics, and central nervous stimulants. Alkaloids also have metabolic effects and control development in living systems (Aboaba et al., 2011; Madziga et al., 2010). (Mir et al., 2013). mentioned the role of saponins as one of the phytochemicals in cholesterol lowering. Tannins containing plants are commonly used as mouthwashes, and eye washes, as mentioned by (Abadallah & Ali, 2019; Mir et al., 2013).

They also mentioned that terpenoids are effective as antimicrobial, anti-inflammatory, and immune-modulatory agents and in the case of cancer. (Abadallah & Ali, 2019) explained the result of moringa leaf extracts’ antibacterial activity achieved in our study against the clinical isolates of *Staphylococcus aureus*, *Streptococcus pyogenes*, and *pseudomonas* (Abadallah & Ali, 2019).

The outcome of our study was in line with (Saadabi & Zaid, 2011) results that reported that the extracts of *moringaoleifera* have an impeding effect on pathogenic bacteria like *Pseudomonas aeruginosain* and *Escherichia coli* (Saadabi & Zaid, 2011). The result of this study concludes that ethanol extracts of *moringaoleifera* showed a maximum zone of inhibition (17 mm) against *Pseudomo-*
This conclusion also describes the study by (Al-Bakri & Afifi, 2007) who mentioned moringa leaf extract constraining the growth of E. coli, Klebsiella pneumoniae, and P. aeruginosa (Al-Bakri & Afifi, 2007). Numerous investigations have shown that M. oleifera leaves have antibacterial properties. As an illustration, a study on plant extracts prepared in different solvents found the ether extract of leaves of M. oleifera was the most active ingredient against the ambient and clinical isolates of Proteus mirabilis, a well-known cause of urinary tract infections (Arun & Rao, 2001). Several studies on the antifungal activity of M. oleifera leaves against Trichophytonrubrum, T. mentagrophytes, Epidermophytonfloccosum, and Microsporum documented that the crude extracts as well as essential oils from M. oleifera leaves reveal antifungal activity (Abalaka et al., 2012; Chuang et al., 2007). (Patel et al., 2014) study showed M. oleifera leaf extracts in ethanolic and aqueous forms were effective against the yeast Saccharomyces cerevisiae and the fungus Candida tropicalis, but not against C. albicans. Little is known about the antibacterial activity of M. oleifera roots. Nevertheless, a few investigations have revealed this plant's roots to have anti-microbial properties (Patel et al., 2014). Additionally, in vitro tests on several M. oleifera root bark extracts against S. aureus, E. coli, Salmonella gallinarum, P. aeruginosa, and others showed that ethyl acetate and acetone extracts have the highest activity in comparison to other solvents (Raj et al., 2011). Similar to this, only a small amount of research—a few early reports—has been done on the antibacterial properties of M. oleifera's stem bark. Studies demonstrate the existence of active organic extracts with variable degrees of activity in M. oleifera root extract's antibacterial efficacy against several human diseases (Chhetri et al., 2008).

CONCLUSION

The current study showed that moringa oleifera leaf extracts have an antibacterial activity that impedes bacterial growth. The results express that ethanol extract of moringa oleifera leaf is effective against bacterial strains.

The antibacterial effect may be due to bioactive constituents such as alkaloids, saponins, flavonoids, and tannins. Therefore, the study results emphasized the moringa oleifera leaf components and its therapeutic prospects as an antibacterial agent.

Duality of interest: In relation to this paper's publication, the authors state that they have no conflicts of interest.

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REFERENCES


