PHYTOCHEMICAL ANALYSIS OF SELECTED CURATIVE PLANTS IN MIR ALI SUBDIVISION NORTH WAZIRISTAN, PAKISTAN

Asif Raza, Syed Mukaram Shah, Asad Ullah, Syed Ghias Ali

Centre of Plant Biodiversity, University of Peshawar, Khyber Pakhtunkhwa, Pakistan

Abstract- It is advantageous to apply ethnobotanical and phytochemical research in the search for new drugs. One crucial step in identifying the bioactive components of curative plants used in conventional therapy is screening for phytochemicals. 12 curative plants used in traditional Mir Ali subdivision North Waziristan Pakistan medicine were investigated for the first time. The period of ethnobotanical fieldwork was 2017-2019. Several curative plant species were found and categorized into relevant families. These curative plants were screened using standard protocols to look for the presence of alkaloids, carbohydrates, cardiac glycosides, flavonoids, phenol, phlobatannis, saponins, steroid, tannins, terpenoids and triterpenes. It has been demonstrated that the leaves of all 12 plant species (100%) contained triterpenes, 11 plant species each (91.7%) contained steroid, tannins, flavonoids 10 plant species (83.3%) contained Saponins, 8 plant species each (66.7%) contained Alkaloids, Cardiac glycosides, Phenol and Terpenoide, while 7 plant species each (58.3%) contained Carbohydrates and phlobatannis were the most often used phytochemicals among the curative plants. It has been demonstrated that the leaves of all 12 plant species *i.e., Fagonia indica* Burm. F., *Withania coagulans* (Stocks) Dunal, *Datura stramonium* L., *Olea ferruginea* Wall. ex Aitch, *Nerium oleander* Mill., *Tribulus terrestris* L., *Mentha longifolia* L., *Lepidium draba* L., *Trachyspermum ammi* L., *Verbascum thapsus* L., *Rhazya stricta* L., *Justicia adhatoda* L. contain Triterpenes. The presence of Triterpenes in these curative plants is congruent with their anthropological significance.

Key words: Phytochemicals, curative plants, alkaloids, carbohydrates, cardiac glycosides, flavonoids, phenol, phlobatannis, saponins, steroid, tannins, terpenoids, triterpenes, Mir Ali Subdivision, North Waziristan.

INTRODUCTION

Curative plants are those with naturally occurring active compounds that are known to treat illness and provide pain relief have positive pharmacological effects on people or animals [1]. It is commonly known that traditional medicines and curative plants are used as preventative measures and therapeutic agents for maintaining health, mostly in underdeveloped nations. Nonetheless, at least 25% of medications in the modern pharmacopoeia are derived from plants, and the majority is synthetic analogs based on precursor chemicals that were extracted from plants. In the interest of protecting individual health and well-being and bio prospecting new plant-derived medications, the growing costs of prescription pharmaceuticals have spurred interest in medicinal plants as a resurgent form of health support. There are several factors contributing to the growing acceptance of medicinal plants, one of which is the growing popularity of herbal remedies. Furthermore, it has been observed that industrialized organizations are becoming more and more reliant on the use of these medicinal plants due to the extraction and creation of medications and chemotherapeutics from these plants as well as from traditionally used herbal remedies.

Plant chemistry, often known as phytochemistry, studies the vast array of chemical compounds that are accumulated by vegetation (Harborne, 2011). Certain organic components found in medicinal plants, such as tannins, alkaloids, carbohydrates, flavonoids, terpenoids, and steroids, have defined physiological effects on the human body. These chemical elements found in plant material are beneficial for maintaining health and have the ability to protect the body from harm. Due to extensive screening, the occurrence of these chemical compounds is extremely distinct in terms of biology, taxonomy, and chemistry and is helpful in documenting their aromatic features (Yadav and Agarwala, 2011). It covers these compounds' chemical makeup, biosynthesis, turnover, and metabolism as well as their natural distribution and biological role (Al-Omar et al., 2020).Environmental factors and habitat repercussions associated with desert climate, high salinity, nutrient scarcity, and water scarcity have brought attention to the importance of medicinal plants utilized in pharmaceutical industry development and alternative remedies (Al-Omar et al., 2020) Because of the harsh weather, plants must maintain higher concentrations of chemicals that have defensive properties in order to survive against excessive oxidative stress (Khalaf Allah *et al*.), invasion by animals grazing, and bacterial infection. These kinds of substances are known as plant secondary metabolites (Al-Qahtani et al., 2020; Youssef et al., 2020). The knowledge of using plants

curatively has been passed down for millennia from generation to generation, and it has developed through trial and error, experience, and observation (Karunamoorthi et al., 2013). According to Osman and Abdein (2019), throughout human history, plants have long been used as a source of health (Abdein, 2018). Through trial and error, the diverse therapeutic properties of plants have been passed down through the generations (Mendoza et al., 2018). Different halophytes (Abualreish and Abdein, 2014) have been employed in different contexts (Al-Qahtani et al., 2020). Secondary metabolites generated from plants are utilized as medications to treat human and animal fundamental needs (Osman and Abdein, 2019; Jamshidi-Kia et al., 2018). These plants are generally accepted due to the huge diversity, sustainability, and ease of procurement of natural goods (Abdein, 2018). According to Gurib-Fakim, their acceptability may also be attributed to its accessibility, viability, and safety. Commonly utilized in traditional medicine, plants with demonstrated therapeutic value may include substances that may be candidates for pharmaceutical usage (Rayan et al., 2020; Stevanovic et al., 2019). Furthermore, these compounds are distributed differently in different parts of the plant (Abdel-Mageed et al., 2019). Many plant parts, such as barks, leaves, flowers, stems, fruits, rhizomes, resins, seeds, and roots, are used in traditional medicine. However, people also utilize these herbs for specific goals to address a few major illnesses (Anywar, 2020).

Since ancient times, research into the efficaciousness of therapeutic herbs against various ailments has been necessary. The current era's setup approach for the development of novel medications and evolution is the scientific validation of phytochemicals' bioactivity (Egbuna et al., 2019). Moreover, these compounds give these plants significant nutritional and health-promoting advantages in addition to their medicinal properties (Alqethami et al., 2017). Active compounds differ among plants due to their diversity, and they have different physiological effects on humans (Jithesh et al., 2006). A number of Saudi Arabian regions have ecology of higher salinity affects plant growth and has a significant role in the region's delayed agricultural development. Growing public awareness of the benefits of going "back to nature" for a healthier lifestyle has led to an increase in demand for herbal medications over the past several decades. This study aims to identify secondary metabolites in select traditional medicinal plants and explore the possibility of a relationship between the secondary metabolite content of medicinal plants and their ethnomedicinal usefulness. The phytochemical elements in plants may have anti-oxidant, anti-microbial, and antipyretic activities, which underlie their curative qualities. The World Health Organization states that curative plants are the best source for a wide variety of medications. Therefore, research on these plants is necessary to have a better understanding of their characteristics, safety precautions, and utility. Many of these plant-based curative treatments are found in India's Ayurveda system, and identifying their morphological, pharmacological, or Pharmacognostic characteristics might help to clarify their mode of action and active principle.

MATERIALS AND METHODS

Research area

The study was carried out in the Mir Ali area North Waziristan, Pakistan's (Figure 1). It was situated in the western region of Bannu, encompassing the three tehsils of Mir Ali, Shawa and Spinwam. It is located between latitude $32^{0}59'12''$ and longitude $70^{0}17'21''$. It is located in Koh-e-Suleiman's north and Koh-e-Sufaid's south, where Irano-Turanian plant species predominate (Ali and Qaiser, 1986). Scattered subtropical forests make up this region. The ground is shallow and calcareous. An average of 400 mm of rain falls fall each year.

Phytochemical analysis

Chemical tests for the screening and identification of bioactive chemical constituents in the medicinal plants under study were carried out in extracts as well as powder specimens using the standard procedures as described by Sofowara (1993), Trease and Evans (1989) and Harborne (1973).

Collection and identification of curative plant samples

The native twelve (12) curative plant species were collected from their natural habitat, form Mir Ali Subdivision of North Waziristan, Pakistan. The plant material was identified and authenticated in the Center of Plant Biodiversity University of Peshawar, Pakistan. Leaves were collected *Fagonia indica* Burm. F.; *Withania coagulans* (Stocks) Dunal.; *Datura stramonium* L.; *Olea ferruginea* Wall. ex Aitch.; *Nerium oleander* Mill.; *Tribulus terrestris* L.; *Mentha longifolia* L.; *Lepidium draba* L.; *Trachyspermum ammi* L.; *Verbascum thapsus* L.; *Rhazya stricta* L.; *Justicia adhatoda* L. 12 curative plants (Figure-1 & Table-1).

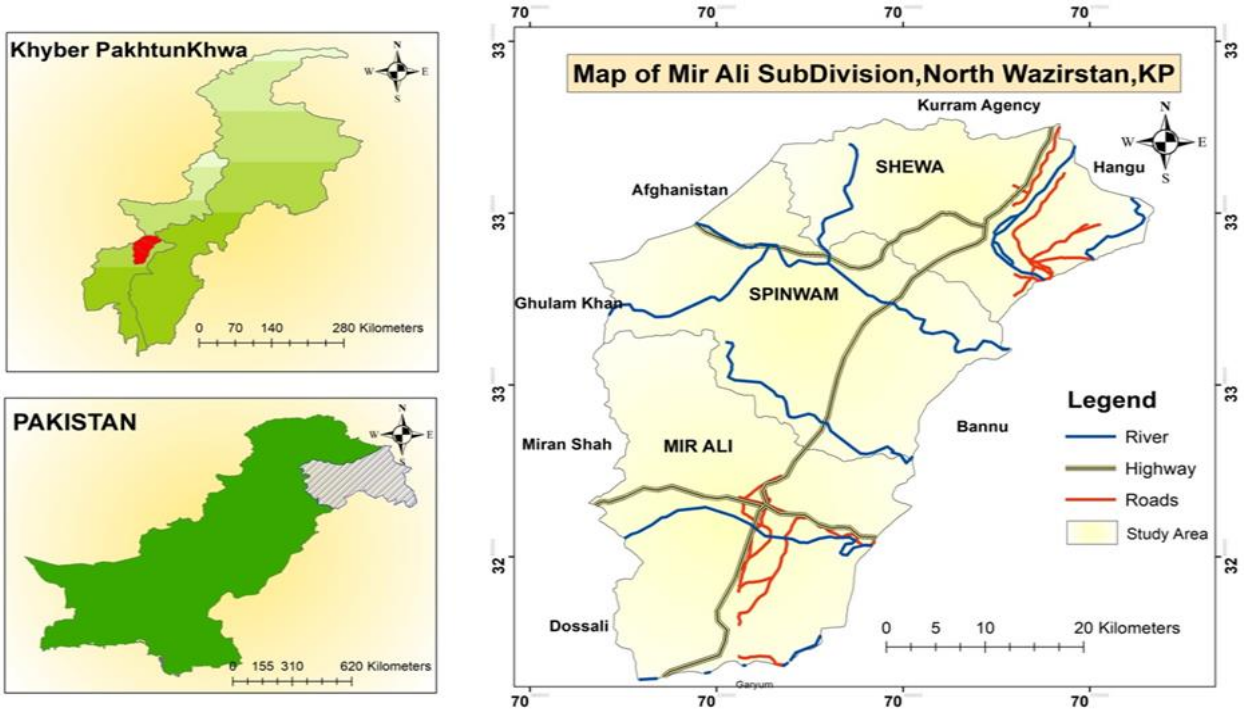

Fig-1. Map of Mir Ali subdivision (Source: GIS Lab. Department of Geography, University of Peshawar).

Table-1: List of curative plants growing in the research area.

Curative Plant	**Local name**	**Family**
Fagonia indica Burm. F.	(Spalahzai)	Zygophyllaceae
Withania coagulans (Stocks) Dunal,	(Shapyanga)	Solanaceae
Datura stramonium L.	(Mangoor Wana)	Solanaceae
Olea ferruginea Wall. ex Aitch	(Shwawan)	Oleaceae
Nerium oleander Mill.	(Gandarai)	Asclepiadaceae
Tribulus terrestris L.	(Markondi)	Zygophyllaceae
Mentha longifolia L.	(Walaniay)	Lamiaceae
Lepidium draba L.	(Bashkay)	Brassicaceae
Trachyspermum ammi L.	(Sperkay)	Apiaceae
Verbascum thapsus L.	(Kharghawag)	Scrophulariaceae
Rhazya stricta L.	(Gandarai)	Apocynaceae
Justicia adhatoda L.	(Baikarh)	Acanthaceae

The collected plants leaves were surface pasteurized by 0.1% mercuric chloride. The collected plant material washed 2-3 times, then allowed to air dry in shade until it reached a steady weight. Unfortunately, a lot of tropical plants haven't had their chemical makeup well investigated recently.

Crude extract preparation

Plants were processed for phytochemical analysis at the Pakistan Council of Scientific and Industrial Research Laboratories Complex (PCSIR), Peshawar, after being chopped, crushed, and shade-dried. The preparation instructions for the crude samples were taken from earlier research (Banso and Adeyemo, 2006). For a full day, the 150 g pulverized sample of the medicinal plant was combined with 500 ml of purified methanol, ethyl acetate, and chloroform in separate conical flasks. The diluted extract was moved to a different flask and filtered the following day. Subsequently, the diluted extract was concentrated at a temperature between 40 and 50 degrees Celsius in a vacuum rotary evaporator with reduced pressure. The solvent, methanol, was then recovered. Three iterations of this procedure were carried out until the extract dried.

Extractive values determination

The resultant extract was heated between 30 and 45 C^0 in a rotary evaporator while being cleared and concentrated under low pressure. The extract was taken out of the round-bottom flask after being reconstituted into a semi-solid state. The product was dried in a water bath set at 45 C^0 using a reweighed china dish. After reweighing the dried extract, the following equation was used to determine the extract's % yield (Banso and Adeyemo, 2006).

$$Extractive\ value\ yield\ \%age = \frac{Weight\ of\ Extract}{Weight\ of\ Ground\ Plant\ Material} \times 100$$

Solvents used

For the phytochemical screening, HPLC-grade methanol (CH_3OH) solvent was utilized.

Phytochemical screening

A color test was used to conduct a phytochemical qualitative screening test in methanol (CH_3OH). Reagents used in this test are changing the extract's color in accordance with the phytochemicals that are present. These 12 different plant species were examined for phytochemical content.

Plant Material Extraction

Following two weeks of air drying at room temperature (26°C), the plant material was milled into a homogeneous powder. 100 g of the dry powdered plant components were soaked in 1 L of ethanol for 48 hours at room temperature to create the ethanol extract. After 48 hours, the extracts were filtered individually using cotton wool and Whatmann filter paper No. 42 (125 mm). Using a rotary evaporator with a water bath set at 40°C, the extracts were concentrated. The extracts' percentage yield varied from 7 to 19% w/w.

Test for alkaloids

Chloroform in order to isolate alkaloids from the chosen plant sample. The alcohol that was boiled, filtered, and acidified produced a variety of extract yield percentages. 2 ml of diluted ammonia was mixed with 5 ml of the filtrate. After a thorough shake and filtration, 10 milliliters of 1% HCl solution were added. Ten milliliters of acetic acid were used to remove the layer of chloroform. This was split into two halves, one of which received the addition of Mayer's reagent and the other of Dragendorff's reagent. When a cream or reddish brown precipitate was formed using Mayer's reagent or Dragendorff's reagent, it was considered that alkaloids were present.

Test for Carbohydrates

One drop of ferric chloride solution was added to two milliliters of glacial acetic acid after 0.5 grams of extract was diluted to five milliliters of water. A milliliter of pure sulfuric acid was applied underneath. A dark ring near the contact suggested that cardenolides have deoxy sugar properties. In the acetic acid layer, a greenish ring formed slightly above the brown ring and eventually expanded throughout the layer, while a violet ring emerged beneath the brown ring. The outcome was the formation of the brick-red precipitate, which suggested the presence of carbohydrates.

Test for cardiac glycosides

Five grams of each extract were combined with 10 ml of glacial acetic acid solution, one drop of 2.0% $FeCl_3$ solution, 10 ml of aqueous plant extract, and 1 ml of H_2SO_4 solution. Consequently, the presence of cardiac glycosides was suggested by the creation of a brown layer in the interfaces.

Test for flavonoids

Three methods were used to test flavonoids.

First, 5 ml of diluted ammonia was combined with a portion of the extract's aqueous filtrate. There was one milliliter (ml) of sulfuric acid administered. The coloration turned yellow while flavonoids were present, but disappeared when the sample stood. The second step involved combining a tiny quantity of the filtrate with a few drops of 1% aluminum solution. The hue turned yellow, indicating the presence of flavonoids. Third, a portion of the extract was cooked in 10 milliliters of ethyl acetate for three minutes in a steam bath. Following the filtering of the combination, 4 ml of the filtrate and 1 ml of diluted ammonia solution were combined. The hue turned yellow, indicating the presence of flavonoids.

Test for phenol

The total phenolic contents of various samples were determined by the method of Makkar et al. (1993). A sample of 0.1 g was combined with 2.8 ml of 10% Na_2CO_3 and 0.1 ml of 2N Folin-ciocalteu reagent. The brownish color indicated the presence of phenolic contents.

Test for Phlobatannins

In order to determine the conformation of phlobatannins in certain plants, 0.2 g of powder from each extract was boiled for 4 minutes in 5 ml of distilled water and then 10 ml of 2% HCl. If there is no outward reaction, there are no phlobatannins.

Test for saponins

Two grams of each extract were heated to boiling after being violently shaken with five milliliters of distilled water to look for saponins in the plants that were chosen. Saponins are present when foam persists.

Test for steroid (Salkowskis test)

An organic molecule with rings structures and a certain shape is called a steroid. Two grams of each extracted sample and two to three drops of concentrated H_2SO_4 were combined in order to examine the steroids in the chosen plants. Consequently, a red coating forms, signifying the presence of steroids.

Test for tannin (Braymer's test)

In a test tube, 0.5 g of the extract was heated in 10 ml of water and subsequently filtered. After adding a few drops of 0.1% ferric chloride ($FeCl_3$) solution, the presence of tannins was determined by looking for a brownish green or blue-black coloration.

Test for Terpenoids (Salkowski Test)

Two milliliters of chloroform were mixed with 0.5 grams of each extract. Carefully, 3 ml of concentrated H_2S0_4 was added to create a layer. The interface's reddish-brown coloration suggested the presence of terpenoids.

Test for Triterpenoids

0.5 mg of dry crude plant extract was diluted in 2 mL of chloroform, and 1 mL of acetic anhydride was added to determine the Triterpenoids in the chosen plants. The mixture was then given a 1 mL concentrated H_2SO_4 addition. Thus, the development of a reddish violet color indicated the existence of Triterpenoids.

RESULTS AND DISCUSSION

Presumably, the selected medicinal plants used in Mir Ali, North Waziristan, are the source of optional metabolites such as flavonoids, starches, glycosides, alkaloids, phenols, and Phytosterols. Because these optional metabolites are nearby, the selected curative plants have a great potential for healing. These phytochemicals give the studied plants' therapeutic estimates.Three distinct solvents ethyl acetate, methanol, and chloroform were used to conduct the phytochemical tests for alkolides, carbohydrates, cardiac glycosides, flavonoids, phenol, phlobatannis, saponins, steroids, tannins, terpenoides, and triterpenes. The findings were displayed as follows (Table-2).

Table-2: Phytochemical screening of some plants ((+) present, (−) absent)) of some curative plants in Mir Ali subdivision, North Waziristan, Pakistan

S.No.	Botanical name	1	2	3	4	5	6	7	8	9	10	11	12
1.	*Fagonia indica* Burm. F.	Leaves	+	+	+++	+	+	+	++	+	+++	+	+
2.	*Withania coagulans* (Stocks) Dunal	Leaves	-	+	+	++	+	-	++	++	+	+	++
3.	*Datura stramonium* L.	Leaves	+++	++	+	-	-	+	+	+	++	-	+
4.	*Olea ferruginea* Wall. ex Aitch.	Leaves	-	-	+	++	++	-	-	++	+++	++	+++
5.	*Nerium oleander* Mill.	Leaves	+	-	-	++	++	+	+++	+	+	+	+
6.	*Tribulus terrestris* L.	Leaves	-	+	-	++	+	+	+	++	++	+	++
7.	*Mentha longifolia* L.	Leaves	-	++	+++	+++	+++	-	++	+++	+++	++	+++
8.	*Lepidium draba* L.	Leaves	++	-	+	+++	-	-	+++	++	++	-	++
9.	*Trachyspermum ammi* L.	Leaves	++	-	++	++	-	+	++	+	+	-	+
10.	*Verbascum thapus* L.	Leaves	++	-	-	++	_	+	+++	+	+	+	+
11.	*Rhazya stricta* L.	Leaves	++	+	++	++	+	+	++	+	-	-	++
12.	*Justicia adhatoda* L.	Leaves	++	+	-	+++	+	-	-	-	+	+	+
Present in concentration (+) %			66.7	58.3	66.7	91.7	66.7	58.3	83.3	91.7	91.7	66.7	100

Keys: 1. Parts used 2. Alkaloids 3. Carbohydrates 4. Cardiac glycosides 5. Flavonoids 6. Phenol 7. Phlobatannis 8. Saponins 9. Steroid 10. Tannins 11. Terpenoide 12. Triterpenes: +Present in small concentration: ++ Present in moderately high concentration: +++ Present in very high concentration: - absent

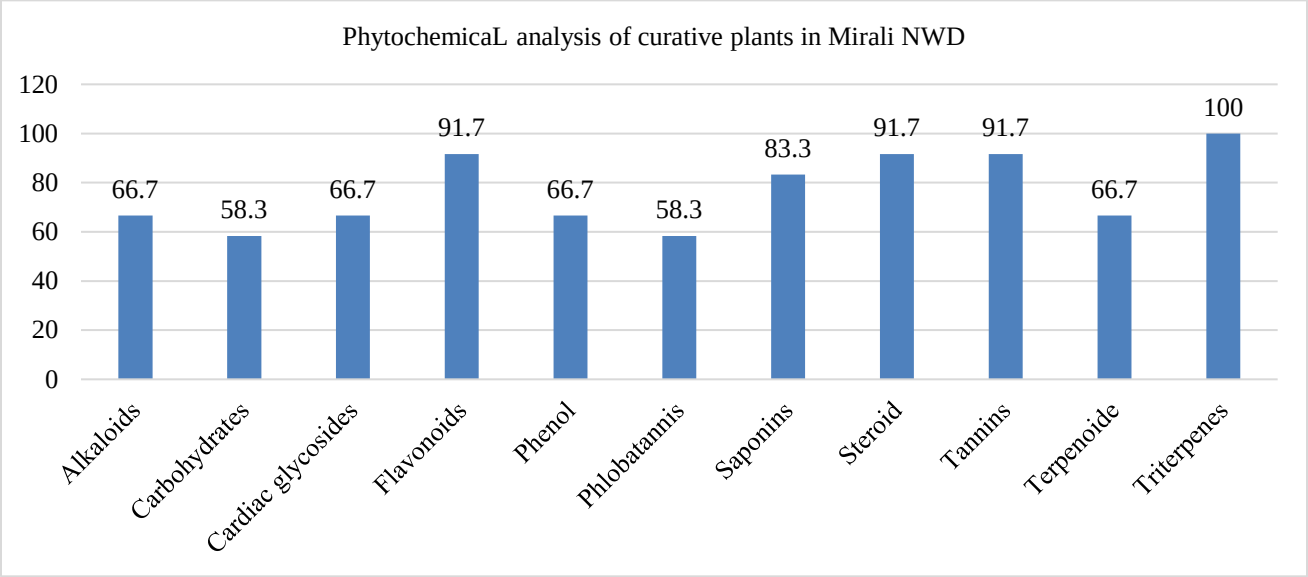

Fig. 2: %age distribution of phytochemicals among screened curative plants

Secondary metabolites such as phenol, phlobatannis, steroid, terpenoide, triterpenes, alkaloids, carbohydrates, tannins, and cardiac glycosides were found in leaves of the *Fagonia indica* plant (Table-2).

The phytochemical analysis of the leaves of *Withania coagulans* revealed the presence of secondary metabolites in a moderately high concentration, including flavonoids, saponins, and steroids; in contrast, the crude extract and its corresponding fractions contained small concentrations of carbohydrates, cardiac glycosides, phenol, tannins, terpenoids, and triterpenes. The crude extract and fractions did not contain alkaloids or phlobatannis (Table-2).

According to the phytochemical results leaves of *Datura stramonium* L. there were secondary metabolites such as alkaloids, which were present in very high concentration, carbohydrates, and tannins, which were present in moderately high concentration; on the other hand, phlobatannis, cardiac glycosides, saponins, steroids, and triterpenes were present in small concentration in the crude extract and its corresponding fractions; flavonoids, phenol, and terpenoids were absent from the crude extract and its corresponding fractions (Table-2).

The leaves of *Olea ferruginea* Wall. ex Aitch. exhibited very high amounts of secondary metabolites such triterpenes and tannins, and moderately high concentrations of flavonoids, terpenoids, phenol, and steroids. Small amounts of cardiac glycosides were discovered in both the crude extract and the appropriate fractions. It was discovered that the crude extract and its fractions lacked alkaloids, polysaccharides, phenolphthalein, and saponins (Table-2).

Nerium oleander Mill. leaves contained very high amounts of secondary metabolites such steroids and saponins, but only moderately high concentrations of flavonoids and phenol. In trace amounts, the crude extract and its corresponding fractions contained alkaloids, phlobatannis, tannins, terpenoids, and triterpenes. It was shown that the crude extract and fractions contained neither cardiac glycosides nor carbohydrates (Table-2).

The leaves of *Tribulus terrestris* L. plant was subjected to a phytochemical analysis, which revealed the presence of secondary metabolites such as flavonoids, steroids, tannins, and triterpenes as well as saponins in a moderately high concentration. In contrast, the crude extract and its corresponding fractions contained small amounts of alkaloids, phlobatannis, tannins, and terpenoids. Alkaloids and cardiac glycosides were found to be absent from crude extract and its fractions (Table-2).

The leaves of *Mentha longifolia* L. revealed the presence of fairly high concentrations of carbohydrates, saponins, tannins, and terpenoids, but very high concentrations of cardiac glycosides, flavonoids, phenol, steroids, and triterpenes. Alkaloids and phlobatannis were found to be absent from crude extract and its fractions (Table-2).

The phytochemical analysis of the leaves of *Lepidium draba* L. was presented in Annexure-22, which demonstrated the presence of secondary metabolites in very high concentrations, including flavonoids and saponins, as well as alkaloids, cardiac glycosides, tannins, steroid and teterapenes. It was shown that phenol, phlobatannis, terpenoids, and carbohydrates were absent from the crude extract and its fractions (Table-2).

The leaves of *Trachyspermum ammi* L. displayed somewhat high concentrations of tannins, teterapenes, phlobatannis, and steroids, but very high concentrations of alkaloids, cardiac glycosides, flavonoids, and saponins. It was demonstrated that phenol, terpenoids, and carbohydrates were absent from the crude extract and its fractions. It was demonstrated that phenol, terpenoids, and carbohydrates were absent (Table-2).

The presence of secondary metabolites in the leaves of *Verbascum thapsus* L. included very high concentrations of saponins and flavonoids, as well as moderately high concentrations of alkaloids, phlobatannis, steroids, tannins, terpenoids, and triterpenes. Carbohydrates, cardiac glycosides, and phenol were found in crude extract and fractions (Table-2).

The results of the phytochemical analysis of the leaves of *Rhazya stricta* L. demonstrates the presence of secondary metabolites, including triterpenes, alkaloids, cardiac glycosides, phenol, and saponins, which are present in very high concentrations, while carbohydrates, phenol, phlobatannis, and steroids are present in moderately high concentrations. It was demonstrated that terpenoids and tannin were present in crude extract and fractions (Table-2).

Secondary metabolites found in *Justicia adhatoda* L. leaves included flavonoids, which were found in very high concentrations, and alkaloids, which were found in moderately high concentrations. Carbohydrates, phenol, steroids, terpenoids, and triterpenes were found in minor concentrations. It was demonstrated that cardiac glycosides, phlobatannis, saponins, and tan were present in the crude extract and fractions (Table-2).

CONCLUSION

Alkolides, carbohydrates, cardiac glycosides, flavonoids, phenol, phlobatannis, saponins, steroids, tannins, terpenoids, and triterpenes were found, according to the phytochemical analysis. Phytochemical analyses aid in identifying different secondary metabolites that regulate fungal infections on medicinal plants. Therefore, at the expense of pesticides, which serve as biological control agents and assist vegetable growers in avoiding synthetic medications, these medicinal plants are playing a crucial role in disease prevention.

ACKNOWLEDGEMENT

We appreciate the assistance with this research provided by the PCSIR, Peshawar biochemistry laboratory. We are also grateful to Dr. Mushtaq Ahmad for his assistance with the manuscript's preparation.

REFERENCES

Al-Omar, M. S., Mohammed, H. A. & Mohammed, S. A. A. (2020) Anti-Microbial, Anti-Oxidant, and α-Amylase Inhibitory Activity of Traditionally- Used Medicinal Herbs: A Comparative Analysis of Pharmacology, and Phytoconstituents of Regional Halophytic Plants' Diaspora. *Molecules*, 25, 5457. https://doi.org/10.3390/molecules25225457

Abdein, M. A. (2018) Genetic Diversity between Pumpkin Accessions Growing in the Northern border Region in Saudi Arabia Based on Biochemical and Molecular Parameters. *Egyptian Journal Botany*, 58, 463-476. https://doi.org/10.21608/ejbo.2018.3612.1171.

Abdein, M. A. & Osman, A. K. E. (2020) Plant Diversity Assessment of Wadi Al-Hilali, Northern Border Region, Saudi Arabia. *International Journal of Botany Studies*, 5, 87-95.

Abdein, M. A., Wrda, H. N. & Osman, A. K. (2020) Genetic Characterization of Genus Tephrosia Pers. Based on Molecular Markers in KSA. International Journal of Botany Studies, 5, 203-209.

Abdel-Mageed, A. M., Osman, A. K. E., Awad, N .S. & Abdein, M. A. (2019) Evaluation of Antidiabetic Potentiality of Truffles and Balanites Aegyptiaca among Streptozotocin Induced Diabetic Rats. *International Journal of Pharmaceutical Research & Allied Sciences*, 8, 53-58.

Abualreish, M. J. A. & Abdein, M. A. (2014). The Analytical Applications and Biological Activity of Hydroxamic Acids. *Journal of Advances in Chemistry*, 10, 2118- 2125. https://doi.org/10.24297/jac.v10i1.5587.

Ali, S. I. & M. Qaiser, M. (1986). A phytogeographical Analysis of Phanerogams of Pakistan and Kashmir. *Proc. of Royle Soc. Edinburgh*, 89B: 89-101.

Allah, K., Rehab, M., Yousef, M., et al. (2019) Utilization of Black Berry Juice to Reduce the Oxidative Stress in Rats Treated with Acrylamide. *Asian Journal of Biological Sciences*, 12, 9-16. https://doi.org/10.3923/ajbs.2019.9.16

Al-Qahtani, H., Alfarhan, A.H. and Al-Othman, Z.M. (2020) Changes in Chemical Composition of *Zilla spinosa* Forssk. Medicinal Plants Grown in Saudi Arabia in Response to Spatial and Seasonal Variations. *Saudi Journal of Biological Sciences*, 27, 2756-2769. https://doi.org/10.1016/j.sjbs.2020.06.035

Alqethami, A., Hawkins, J.A. and Teixidor-Toneu, I. (2017) Medicinal Plants Used by Women in Mecca: Urban, Muslim and Gendered Knowledge. *Journal of Ethnobiology and Ethnomedicine*, 13, 62. https://doi.org/10.1186/s13002-017-0193-4.

Anywar, G. (2020) Historical Use of Toxic Plants. In: Mtewa, A.G., et al., Eds., Poisonous Plants and Phytochemicals in Drug Discovery, Wiley, Hoboken, 1-17. https://doi.org/10.1002/9781119650034.ch1.

Egbuna, C., Kumar, S. & Ifemeje, J. C. (2019) Phytochemicals as Lead Compounds for New Drug Discovery. Elsevier, Amsterdam.

Gurib-Fakim, A. (2006) Medicinal Plants: Traditions of Yesterday and Drugs of Tomorrow. *Molecular Aspects of Medicine*, 27, 1-93. https://doi.org/10.1016/j.mam.2005.07.008.

Harborne J. B. (2011). Phytochemical methods: A guide to modern techniques of plant analysis, Springer International 3rd edition 1.

Jamshidi-Kia, F., Lorigooini, Z. and Amini-Khoei, H. (2018) Medicinal Plants: Past History and Future Perspective. Journal of Herbmed Pharmacology, 7, 1-7. https://doi.org/10.15171/jhp.2018.01.

Jithesh, M. N., Prashanth, S. R., Sivaprakash, K. R. & Parida, A. K. (2006). Antioxidative Response Mechanisms in Halophytes: Their Role in Stress Defence. Journal of Genetics, 85, 237. https://doi.org/10.1007/BF0293534.

Karunamoorthi, K., Jegajeevanram, K., Vijayalakshmi, J. & Mengistie, E. (2013) Traditional Medicinal Plants: A Source of Phytotherapeutic Modality in Resource Constrained Health Care Settings. *Evidence-Based Complementary and Alternative Medicine*, 18, 67-74. https://doi.org/10.1177/2156587212460241

Makkar, H. P. S., Blummel, M., Borowy, N. K. & Becker, K. (1993). Gravimetric determination of tannins and their correlations with chemical and protein precipitation methods. *J. Sci. Food Agric*., 61:161-165.

Mendoza, N. & Escamilla, E. (2018). Introduction to Phytochemicals: Secondary Metabolites from Plants with Active Principles for Pharmacological Importance. In: Asao, T. and Asaduzzaman, M., Eds., Phytochemicals: Source of Antioxidants and Role in Disease Prevention, IntechOpen, London, 25-47. https://doi.org/10.5772/intechopen.78226.

Osman, A. K. & Abdein, M. A. (2019). Karyological and Molecular Studies between Six Species of *Plantago* in the Northern Border Region at Saudi Arabia. *Journal of Taibah University for Science*, 13, 297-308. https://doi.org/10.1080/16583655.2019.1571400.

Osman, A. K. E. & Abdein, M. A. (2019) Floristic Diversity of Wadi Ar'ar, Saudi Arabia. Journal of Taibah University for Science, 13, 772-789. https://doi.org/10.1080/16583655.2019.1634177.

Rayan, A. M., Abdein, M. A. & Ibrahim, A. A. (2020). Associated Weeds of Some Agro ecosystems in the Northern Border Region, KSA. *International Journal of Botany Studies*, 5, 345-351.

Sofowara, A. (1993). Medicinal plants and traditional medicine in Africa. Spectrum books, Ibadan 150.

Stevanovic, Z. D., Stankovic, M. S., Stankovic, J., et al. (2019) Use of Halophytes as Medicinal Plants: Phytochemical Diversity and Biological Activity. In: Halophytes and Climate Change: Adaptive Mechanisms and Potential Uses, CABI, Wallingford, 343. https://doi.org/10.1079/9781786394330.0343.

Yadav, R. N. S. & Agarwala, M. (2011). Phytochemical analysis of some medicinal plants. *J. of Phytol.*, 3(12): 10-14.

Youssef, S. M., Sayed S. N. & A. M. M. Mahmoud. (2020) Response of Stressed China Aster (*Callistephus chinensis*) cv. Kamini Plants to Foliar Application of Benzyladenine (BA) and Cycocel (CCC). *The Seybold Report*, 15, 30-42.https://doi.org/10.5281/zenodo.7350717.

AUTHORS

First Author- Asif Raza, PhD Scholar, Centre of Plant Biodiversity, University of Peshawar, Khyber Pakhtunkhwa, Pakistan.
Second Author- Syed Mukaram Shah, Ph.D. Lecturer Centre of Plant Biodiversity, University of Peshawar, Pakistan
Third Author- Asad Ullah, Ph.D. Assistant Professor & Director Centre of Plant Biodiversity, University of Peshawar, Pakistan
Fourth Author- Syed Ghias Ali, Ph.D. Assistant Professor Centre of Plant Biodiversity, University of Peshawar, Pakistan
Correspondence Author – Asad Ullah, Ph.D. Assistant Professor & Director Centre of Plant Biodiversity, University of Peshawar, Pakistan,