

In-vitro Evaluation of Antimicrobial Branded Herbal Formulations for their Efficacy

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Abstract

In the present research work forty six herbal antimicrobial branded products were investigated for their antibacterial/antifungal activities using well diffusion method. Those herbal products were collected from local market which were manufactured by well-known herbal manufacturer and have clear recommendations against various microbial infections. These antimicrobial products were tested on the basis of pathogenesis caused by different microorganisms as indicated on the label. Among the tested branded formulations, only nine herbal preparations exhibited better antimicrobial activities, while rest of herbal formulations were devoid of antimicrobial effect. B34 and B30 demonstrated antibacterial activity when tested against *Salmonella paratyphi* B18 was the only product, which showed activity against *Pseudomonas aeruginosa*. The pathogenic fungi *Candida albican* that causes various infections was sensitive to herbal products like B42, B41, B43 and B14 as like to their sensitivity to antifungal drug (Griseofulvin). The results of the present study indicated the poor quality of herbal products being sold in the market with very high label claims. Only 9 out of 46 sampled products with antimicrobial label claimed qualify to some extent for medicinal use. As any health authority (DRA/MOH) in Pakistan does not regulate the herbal products, there was no control or check on the manufacturers and prescriber of these medicines. All the tested products contained herbs, which had proven data for their antimicrobial activity, still most of them failed to produce any pharmacological results.

Keywords: Antimicrobial; Branded herbal formulations; Antibacterial and antifungal

Introduction

In last few years exploring of herbs used for medicinal purposes, which may be safe, economical and easily available to the consumers as well as resistance of disease, causing organisms should be minimum or no development of resistance. The efficacy of more plant species has been determined through scientific validation but most herbal remedies used may cause serious toxic effects and also drug-drug interaction because these herbal remedies may not undergo careful scientific evaluation. Ongoing researches are required to explain the pharmacological activities of many herbal remedies being used [1]. As the environmental conditions, such as the type of soil and cultivation, methods may affect the natural composition of the constituents in a plant so the ingredient that may cause a specific antimicrobial effect is not known. The antimicrobial activity of most plants have been determined but majority of them have not been evaluated properly [2]. A source of antimicrobial activity has been reported from many plants therefore these plants may be used as a source of effective and powerful drugs in different countries of the world [3]. Ointments containing Aloe vera, Neem and Turmeric exhibited broad spectrum anti-bacterial and anti-fungal activities against *E. coli*, *S. aureus*, *Pseudomonas aeruginosa*, *B. subtilis*, *A. varis*, *A. niger* and *P. notatum*

[4]. Medicinal plant parts may be collected either in small quantities for use by the local communities and folk healers or in larger quantities for many herbal manufacturers for extracts as raw drugs because they possess different medicinal properties [5]. The main objectives of the current study were, to evaluate the antimicrobial branded herbal formulations for their efficacy and awareness of the public about blind use of these herbal formulations.

Materials and Methods

This research study was performed at the Department of Microbiology, Hazara University, Mansehra, Pakistan. All the experimental equipment and materials were supplied by the said department.

Samples collection

Forty six herbal branded formulations were purchased from the local market.

Apparatus used

Pre sterilized glass petri dishes, Metallic borer, volumetric flask, Pyrex A (Germany), Sanyo labo autoclave, MLS-3780, S.NO-2Y0301, Phase, Company Sanyo electric co, Ltd Made in Japan, Streamline

Horizontal laminar flow cabinet, ESCO. En 1822.1 class H 13. HEPA filters, ISO 14644.1 Class 4. IEC 61010-1, US Federal standard 209E Class 10, Uni-Bloc SHIMADZU. Capacity Maximum 220 g and Minimum 10mg. SHIMADZU corporation made in Japan, Irmeco hybridization Oven. Digital constant temperature tank (China). Item-Model VRN-360. RPM 60-230 +10, Gemmy industrial corp (Taiwan).

***In vitro* antimicrobial studies of selected herbal branded formulations**

The herbal branded formulations were tested by Agar Well Diffusion Techniques as described by [6]. Different concentrations of the extract were used for testing. For the agar well diffusion method, Mueller Hinton agar plates were seeded with standard inoculation of the microorganisms being tested. Herbal branded formulations were studied for their antimicrobial activity against the selected experimental microorganisms.

Applications of herbal medicines

Wells of 6 mm diameter were prepared in the plates with a sterile metal borer and five, 10, 15, 20, 25 and 100 microlitres of the already prepared test samples were pipette directly into the wells. These plates were incubated overnight at 37°C [7]. The antimicrobial potential of test compound was determined on the basis of diameter of zone of inhibition around the wells [8]. The test was carried out in triplicate and the mean zones of inhibition were calculated. For each bacterial strain, controls were included that comprised of sterile water (in the case of the aqueous extract) or ethanol (in the case of the ethanolic extract). Other wells were supplemented with 10 µg ciprofloxacin. Griseofulvin antifungal discs were used as standard drug. The zones of inhibition were then measured after 24 hrs incubation period. All the experiments were conducted in triplicate [9]. In case, ethanol was used as solvent, calculated the effective zone size of the test samples the zone size of the negative control (ethanol) was subtracted from the total zone of inhibition size.

Results

Antibacterial effects against *Escherichia coli*

Twenty-eight herbal products were tested for their antibacterial activity against *E. coli*. All the tested products were clinically used against all pathogenic condition caused by *E. coli*. The manufacturer of these tested herbal formulations claim that these herb are used for various infections. It is clear that the drops of B14 has maximum inhibitory activity against this gram negative bacterium with zone of inhibition 25 mm. The syrup B8 showed weak activity against *E. coli* with mean zone of inhibition 7 mm. B11 and B41 also showed activity against the tested bacterium with zone of inhibition 15 and 13 mm respectively. The antibacterial effects of these products were compared with standard broad spectrum antibacterial drug (ciprofloxacin) having 26 mm zone size. It was very surprising to note that twenty four tested products failed to show antibacterial activity although the label of all these products indicated that these products were being used in various bacterial infections (Table 1).

S.No	Test samples	Resistant	Sensitive	Zones of inhibition
1	B1 (syrup)	R	-	-
2	B2 (syrup)	R	-	-
3	B14 (drops)	-	S	25 ± 1.23 mm
4	B3 (syrup)	R	-	-
5	B32 (syrup)	R	-	-
6	B18 (syrup)	R	-	-
7	B33 (syrup)	R	-	-
8	B26 (syrup)	R	-	-
9	B28 (syrup)	R	-	-
10	B5 (syrup)	R	-	-
11	B6 (syrup)	R	-	-
12	B11 (syrup)	-	S	15 ± 2.13 mm
13	B27 (syrup)	R	-	-
14	B23 (syrup)	R	-	-
15	B7 (syrup)	R	-	-
16	B8 (syrup)	-	S	7 ± 1.34 mm
17	B9 (syrup)	R	-	-
18	B44 (syrup)	R	-	-
19	B24 (syrup)	R	-	-
20	B12 (syrup)	R	-	-
21	B43(Oil)	R	-	-
22	B29 (Powder)	R	-	-
23	B11 (Tablet)	R	-	-
24	B12 (Ointment)	R	-	-
25	B38 (Ointment)	R	-	-
26	B45 (Ointment)	R	-	-
27	J B11 (Ointment)	R	-	-
28	B41 (Ointment)	-	S	13 ± 2.87 mm
	Ciprofloxacin (standard)	-	S	26 ± 0.09 mm

Table 1: Antibacterial potential of various herbal branded formulations against *Escherichia coli*.

Antibacterial effects against *Staphylococcus aureus*

Twenty-seven herbal formulations were tested against *S. aureus*. Out of tested samples only B11 exhibited antibacterial effects against *S. aureus* with zones of inhibition 20 mm and rest of samples were devoid of antibacterial effect (Table 2).

S.No	Test samples	Resistant	Sensitive	Zones of inhibition
01	B1 (syrup)	R	-	-
02	B16 (syrup)	R	-	-
03	B32 (syrup)	R	-	-
04	B17 (syrup)	R	-	-
05	B18 (syrup)	R	-	-
06	B19 (syrup)	R	-	-
07	B22B21 (syrup)	R	-	-
08	B28(syrup)	R	-	-
09	B5 (syrup)	R	-	-
10	B6 (syrup)	R	-	-
11	B11 (syrup)	-	S	20 ± 2.87 mm
12	B24 (syrup)	R	-	-
13	B7(syrup)	R	-	-
14	B8 (syrup)	R	-	-
15	B9 (syrup)	R	-	-
16	B44 (syrup)	R	-	-
17	B10 (syrup)	R	-	-
18	B32 (syrup)	R	-	-
19	B23 (syrup)	R	-	-
20	B43(Oil)	R	-	-
21	B21 (Ointment)	R	-	-
22	B37 (Cream)	R	-	-
23	B38 (Ointment)	R	-	-
24	B45 (Ointment)	R	-	-
25	B24(Ointment)	R	-	-
26	B2(Ointment)	R	-	-
27	B41 (Ointment)	R	-	-
	Ciprofloxacin (standard)	-	S	26 ± 0.08 mm

Table 2: Antibacterial potentials of various herbal branded formulations against *Staphylococcus aureus*.

Antibacterial effects against *Salmonella paratyphi*

Five herbal products were tested against *S.paratyphi* as the labels of these products indicated that these products are recommended in the treatment of *S. paratyphi* infections. Out of tested products *B34* and *B30* exhibited significant effect against tested bacteria. The zone of inhibition of *B34* and *B30* are 19 and 17 mm, respectively (Table 3).

S.No	Test samples	Resistant	Sensitive	Zones of inhibition
01	B30 (syrup)	-	S	17 ± 2.87 mm
02	B31 (syrup)	R	-	-
03	B32 (syrup)	R	-	-
04	B33 (syrup)	R	-	-
05	B34 (syrup)	-	S	19 ± 1.34 mm
	Ciprofloxacin (standard)	-	S	24 ± 0.07 mm

Table 3: Antibacterial potentials of various herbal branded formulations against *S. paratyphi*.

Antibacterial effects against *Shigella dysenteriae*

Fifteen herbal branded formulations were tested for their antibacterial effects against *Shigella dysenteriae*. Among the tested products only B11 showed significant activity had mean zones of inhibition 19 mm and rest of antimicrobial products showed resistance. The activity of the test samples were compared with Standard anti-bacterial drug with zone of inhibition 20 mm (Table 4).

S.No	Test samples	Resistant	Sensitive	Zones of inhibition
1	B1 (syrup)	R	-	-
2	B1 (syrup)	R	-	-
3	B3 (syrup)	R	-	-
4	B18 (syrup)	R	-	-
5	B26 (syrup)	R	-	-
6	B5 (syrup)	R	-	-
7	B6 (syrup)	R	-	-
8	B11 (syrup)	-	S	19 ± 1.23 mm
9	B27 (syrup)	R	-	-
10	B7(syrup)	R	-	-
11	B8 (syrup)	R	-	-
12	B29 (Powder)	R	-	-
13	B9 (syrup)	R	-	-
14	B10 (syrup)	R	-	-
15	B45 (tablet)	R	-	-
	Ciprofloxacin (standard)	-	S	20 ± 0.06 mm

Table 4: Antibacterial potentials of various herbal branded formulations against *Shigella dysenteriae*.

Antibacterial effect against *Pseudomonas aeruginosa*

Twenty-five herbal formulations were tested against *P. aeruginosa*. These formulations are indicated against *P. aeruginosa* infections. Among the tested samples only B18 was active with mean zone of inhibition 12 mm (Table 5).

S.No	Test samples	Resistant	Sensitive	Zones of inhibition
01	B1 (Syrup)	R	-	-
02	B15 (syrup)	R	-	-
03	B16 (syrup)	R	-	-
04	B17 (syrup)	R	-	-
05	B18 (Syrup)	-	S	12 ± 1.25 mm
06	B19 (syrup)	R	-	-
07	B20 (syrup)	R	-	-
08	B21 (syrup)	R	-	-
09	B33 (Syrup)	R	-	-
10	B26 (syrup)	R	-	-
11	B28 (syrup)	R	-	-
12	B27 (syrup)	R	-	-
13	B24 (syrup)	R	-	-
14	B44 (syrup)	R	-	-
15	B34 (syrup)	R	-	-
16	B23 (syrup)	R	-	-
17	B25 (syrup)	R	-	-
18	B25 (syrup)	R	-	-
19	B43 (Oil)	R	-	-
20	B29 (Powder)	R	-	-
21	B33 (Ointment)	R	-	-
22	B37 (Cream)	R	-	-
23	B41 (Ointment)	R	-	-
24	B12 (Ointment)	R	-	-
25	B41 (Ointment)	R	-	-
	Ciprofloxacin (standard)	-	S	14 ± 0.14 mm

Table 5: Antibacterial potential of various herbal branded formulations against *Pseudomonas aeruginosa*.

Antibacterial effects against *Salmonella typhimurium*

None of the tested formulations were effective against *Salmonella typhimurium* (Table 6).

S.No	Test samples	Resistant	Sensitive	Zones of inhibition
1	B1 (Syrup)	R	-	-
2	B11 (Syrup)	R	-	-
3	B7 (Syrup)	R	-	-

S.No	Test sample	Resistant	Sensitive	Zone of inhibition
1	B23	R	-	-
2	B35	R	-	-
3	B14	-	S	26 ± 1.23 mm
4	B37	R	-	-
5	B38		S	5 ± 2.14 mm
6	B45 Marham	R	-	-
7	B46	R	-	-
8	B40	R	-	-
9	B43		S	4 ± 0.82 mm
10	B41		S	6 ± 1.23 mm
	Griseofulvin (standard)		S	17 ± 0.11 mm

Table 6: Antibacterial potential of various herbal branded formulations against *Salmonella typhimurium*.

Antifungal activity against *Candida albicans*

Ten herbal branded formulations were tested for the determination of their antifungal activity. Out of the tested products four products showed activity against *Candida albicans* with zone of inhibition 26 mm, 5 mm, 4 mm and 6 mm respectively. Surprisingly B14 showed better result than Griseofulvin the standard anti-fungal drug (Table 7).

S.No	Test sample	Resistant	Sensitive	Zone of inhibition
1	B23	R	-	-
2	B35	R	-	-
3	B14	-	S	26 ± 1.23 mm
4	B37	R	-	-
5	B38		S	5 ± 2.14 mm
6	B45 Marham	R	-	-
7	B46	R	-	-
8	B40	R	-	-
9	B43		S	4 ± 0.82 mm
10	B41		S	6 ± 1.23 mm
	Griseofulvin (standard)		S	17 ± 0.11 mm

Table 7: Antifungal activity against *Candida albican*.

Discussion

The scope of present study is to evaluate the antimicrobial activity of various herbs in the form of final dosage form. Herbal products are available in different dosage form like syrups, capsules, tablets, powder, ointments, emulsions and herbal teas. A number of herbal manufacturing companies have surfaced and established well equipped manufacturing units throughout Pakistan. Some of them are enjoying a very good business position while others are struggling. Botanical medicine has become a topic of increasing global importance, with both medicinal and economic implications. The numerous reports of adverse effects and widespread sale of adulterated products and misleading health claims of these products demand proper regulations on botanical medicine [10]. Depending on the particular country and existing legislation, herbal products used for diagnosis, cure, mitigation, treatment, or prevention of diseases are normally regulated as drugs. However, in some countries, including the United States, botanical products are marketed as "dietary supplement". Other countries treat the herbal preparations as drugs, and to be registered these products need to be tested to prove their safety and clinical efficacy. However, so far, few programs have been established to study the safety and efficacy of herbal medicines as

originally proposed by the WHO Guidelines for the assessment of herbal medicines [11,12]. Currently, no organization or government body regulates the manufacture or certifies the labeling of herbal preparations. This means there is no assurance that the amount of the herb contained in the bottle or even from dose to dose, is the same as what is stated on the label. However, it is still important to ask companies that are making standardized herbal products the basis for their product's guarantee. It is still important to consult a doctor or an expert in herbal medicine for the recommended doses of any herbal products [13]. Drug regulatory agency (DRA) of Pakistan has recently been established and is in process of completing its structure. The tasks assigned to DRA were previously carried out by Ministry of Health of Pakistan (MOH), which has been dissolved. Affairs related to drugs/ medicines are governed by Drug Act of Pakistan 1976 with few amendments. But Drug Act of Pakistan 1976 only addresses the Allopathic medicines, while the Herbal/ Unani/ Homeopathic medicines do not qualify the definition of drug as per Drug Act. A

drug may be any substance or mixture of substance that is manufactured, sold, stored, offered, for sale or represented for internal or external use in the treatment, mitigation, prevention or diagnosis of disease, an abnormal physical state or the symptoms thereof in human being or animals or the restoration, correction or modification of organic function in human being or animals. Not being a substance exclusively used or prepared for use in accordance with the Ayurvedic, Unani, Homeopathic or Biochemical system of treatment except those substances and in accordance with such condition as may be prescribed [14,15]. Tables 8-12 shows list of herbal products purchased from the market and their ingredients (herbs). Literature review in Table 1 shows all these herbs have a strong research data for their pharmacological activities. Almost every sample product contains one or more herbs having reported antimicrobial activity supporting the label claims of these products. But the present study shows that only few products have shown promising results against the selected strains.

Medicine name	Manufacturer	Ingredients used
B1	Aftab Qarshi	Pomegranate bark, <i>Clitoria ternatea</i> , <i>Curcuma zerumbet</i> , <i>Foeniculum vulgare</i>
B2	Dawakhana Hakim Ajmal Khan	<i>Rosa damascene</i> mil, <i>Foeniculum vulgare</i> , <i>Polygonumbistora</i> , <i>Helicteres isora</i> , <i>Phyllanthus</i> , <i>Amomum subulatum</i>
B3	Falcon Herbal Lab	<i>Rosa damascene</i> , <i>Cinnamomum zeylanicum</i> , <i>Hyssopus Officinalis</i> , <i>Kamoon sufaid</i> , <i>Mangifera indica</i> , <i>Danheel</i> , <i>Grape seed</i> , <i>Menthasylvestris</i> , <i>Coriandrum sativum</i> , <i>Sodium bicarbonate</i> , <i>Satelimon</i> , <i>Turang</i> , <i>Sirkadesi</i> , <i>Punica granatum</i> , <i>Pruns domestica</i> , <i>Foeniculum vulgare</i> , <i>Sodium chloride</i> , <i>QandSufaid</i> .
B4	Qarshi	<i>Acacia arabica</i> wild, <i>Punica granatum</i> , <i>Foeniculum vulgare</i> , <i>Myrtus communis</i>
B5	Wasay Lab	<i>Mentha sylvestris</i> , <i>Peucedanum graveolens</i> , <i>Foeniculum vulgare</i> , <i>Cinnamomum zeylanicum</i> , <i>Rawand khatai</i> , <i>Myrtus communis</i> , <i>Polygonumbistora</i>
B6	Al-Majid Herbal	As in B5
B7	Mumtaz Dawakhana	<i>Ptychotis ajowan</i> , <i>Punica granatum</i> , <i>Wrightia tinctoria</i> , <i>Bombax malabaricum</i> , <i>Cypreuss carious</i> , <i>Plantago major</i> , <i>Mentha sylvestris</i> , <i>Myrtus communis</i> , <i>Punica granatum</i> , <i>Ptychotis ajowan</i> , <i>Foeniculum vulgare</i> , <i>Tajqalme</i> , <i>Ocimum gratissim</i> , <i>Tamarix gallica</i> , <i>Polygonumbistora</i>
B8	Mumtaz Dawakhana	<i>Mentha sylvestris</i> , <i>Carcuma cassia</i> , <i>Post taranj</i> , <i>Amomum subulatum</i> , <i>Rosa damascene</i> , <i>Samakdana</i>
B9	Master Unani	<i>Amomum subulatum</i> , <i>Hyssopus officinalis</i> , <i>Mentha sylvestris</i> , <i>Punica granatum</i> , <i>Namaksiah</i> , <i>Ptychotis ajowan</i> , <i>Cinnamomum zeylanicum</i> , <i>Myristica fragrans</i> , <i>Valeriana officinalis</i> , <i>Mentha sylvestris</i> , <i>Camphor</i> .
B10	Ashraf Lab	<i>Acacia arabica</i> , <i>Punica granatum</i> , <i>Myrtus communis</i> , <i>Psidium guajava</i> , <i>Cydonia oblonga</i> , <i>Pyrus malus</i> .
B11	Qarshi	<i>Punica granatum</i> , <i>Mentha sylvestris</i> , <i>Amomum subulatum</i> , <i>Citrus limonum</i> <i>Valeriana officinalis</i> , <i>Rosa damascene</i> , <i>Coriandrum sativum</i> , <i>Pistacia vera</i>
B12	Falcon	<i>Foeniculum vulgare</i> , <i>Ptychotis ajowan</i> , <i>Mentha sylvestris</i> , <i>Coriandrum stivum</i> , <i>Cinnamomum tamala</i> , <i>Amomum subulatum</i> , <i>Apium graveolens</i> , <i>Mentha sylvestris</i> , <i>Qandsufaid</i>
B13	(Al-majid herbal pharma)	<i>Glycyrrhiza glabra</i> , <i>Ocimum gratissim</i> , <i>Ispaghul husk</i> , <i>Pistacia lentiscum</i> , <i>Tokhm-e bartang</i> , <i>Phyllanthus</i> , <i>Ptychotis ajowan</i>

Table 8: List of herbal formulations used for the treatment of GIT disorders. (Colitis due to dysentery, diarrhea, gastritis and enteritis, acute and chronic dysentery, useful for intestinal ulcers, Cholera. For watery and bloody diarrhea and gastrointestinal weakness and cramps, gastralgia, peptic ulcer).

Medicine name	Manufacturer	Ingredients used
B14	Qarshi	<i>Camphora officinarum</i> , <i>Mentha pipriata</i> oil, <i>Ptychotis ajowan</i> oil, <i>Foeniculum vulgare</i> oil, <i>Pistacia terebinthus</i> oil, <i>Populus euphratica</i> oil

B15	Ashraf Lab	<i>Ptychotis ajowan, Curcuma Longa, Urginea indica, Foenicullum vulgare, Mentha arvensis, Ephedra vulgaris, Adhatoda vasica, Calotropis gigantea, Lobelia, Polygala senega, Papaver somniferum, Glycyrrhiza glabra, Rhus succedanea, Linum usitatissimum, Piper cubeba, Piper longum, Zingiber officinale.</i>
B16	Qarshi	<i>Ephedra vulgaris, Ammoniiichloridum, Glycyrrhiza glabra, Adhatoda vasica</i>
B17	Ashraf lab	<i>Glycyrrhiza glabra, Cichorium intybus, Tinospora cordifolia, Hyssopus officinalis, Viola odorata, Polygala senega, Ephedra vulgaris, Scilla serrata, Sisymbrium irio, Ficus carica, Ptychotis ajowan, Bombyx mori</i>
B18	Qarshi	<i>Ephedra vulgaris, Papaver somniferum, Glycyrrhiza glabra, Adhatoda vasica, Rhus succedanea, Achyranthus aspera, Mentha piperita</i>
B19	AftabQarshi	<i>Adhatoda vasica, Glycyrrhiza glabra, Adiantum capillus-veneris, Ptychotis ajowan, Ephedra vulgaris.</i>
B20	Riazdawkhana	<i>Adhatodavasica, Calotropis, Curcuma longa, Ephedra, Eucalyptus, Foeniculumvulgare, Foeniculumvulgare, Lyceum barbarum, Menthe arvensis, Papaversomniferum, Pimpinellaanisum, PtychotisajowanScilla, Trigonellafoenumgraecum.</i>
B22	Ashraf Lab	<i>Ephedra vulgaris, Glycyrrhizaglabra, Polygala senega, Adhatodavasica, Plumbicarbonas, Papaversomniferum, Menthaarvensis, Foeniculumvulgare, Lobelia nicotianaefolia.</i>
B11	New Pak	<i>Adhatodavasica, Hyssopusofficinalis, Althoeraofficinalis, Zizyphussativus, Ficuscarica, Adiantumcapillus-veneris, Piper betle, Glycyrrhizaglabra, Viola odorata, Malvasylvestris, Sande, Cordiamyxa, Ephedra vulgaris.</i>
B23	AftabQarshi	<i>Onosmabrcateatum, Hyssopusofficinalis, Viola odorata, Rhus succedanea, Cordiamyxa, Ephedra vulgaris, Glycyrrhizaglabra, Zizyphussativus, B25B24, Foeniculumvulgare.</i>
B25	AftabQarshi	<i>Morusnigra.</i>
B25	Falcon	<i>Morusnigra (extract), Glycyrrhizaglabra.</i>

Table 9: List of herbal formulations used for Respiratory Tract Infections (RTIs).

B26	Otogen	<i>Small caltrops, Sanataium white, Coriandrum sativum, Carbonate of potash, Postassinitras, Tinosporiacordifolia, Cucumber.</i>
B27	MumtazDawkhana	<i>B27 juice, Sugarcane vinegar, Potassium carbonate, Peter Salt, Helmintholithusjudiacus, Ptychotisajowan</i>
B28	Ashraf lab	<i>Emblicaofficinalis, Terminaliachebula (raw), Terminaliablerica, Vitexnegundo, Artemisia absinthium, Meliaazadirach , Alhagimaurorum, Azadirachtaindica, Azadirachtaindica, Centaureasolsitalis, Citrulluscolocynthis, Sphaeranthusindicus, Tephrosapurpurea, Fagoniacretica, Smilexofficinalis, Swertiachirata, Fumariaofficinalis, Achilleamillefolium, Zizyphussativus</i>
B29	Qarshi	<i>Calcined oyster shell, Datura alba, Rheum emodi, Zingibarofficinale 34.4mg.</i>

Table 10: List of herbal formulations used for Urinary Tract Infections (UTIs)

B30	AftabQarshi	<i>Caesalpinabonducella, Foeniculumvulgare, Sisymbriumirio, Glycyrrhizaglabra, Vicisvinifera, Fumariaofficinalis, Ficuscarica, Ammonium chloride, Tinsporacardifolia, Miers, Alum, Ptychotisajowan.</i>
B31		
B32	Ashraf	<i>Tinosporiacardifolia, Cichoriumintybus, Nymphaea alba , Azadiracchtaindica, Fumariaofficinalis, Pterocarpussantalinus, Glycyrrhizaglabra, Foeniculumvulgare, Sphaeranthushirtus, Ext glycyrrhiza, Berbreisaristata, Ammonium chloride.</i>
B33	Qarshi	<i>Tinosporacardifolia, Meliaazadirachta, Swertiachirata, Glycyrrhizaglabra, Sisymbriumirio , Ptychotisajowan, Fumariaofficinalis, Solanumnigram</i>
B34	Qarshi	<i>Sisymbriumirio, Zizyphus Vulgaris, Glycyrrhizaglabra, FicusCarica, AmomumSublatum, Oyster shell, Zinc Murakab, Foeniculumvulgare.</i>

Table 11: Medicines used for treating different type of temperatures.

Aafaqee	Al-shifadawkhana.	Graphites, Calendula, Canthris, Hydrastis, Sulphur, Carbovrg, Glycerin, Vasline.
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B35	Kent Homoeopathic Pharmacy	Herb. Equif, <i>Calendula</i> , <i>Matricaria</i> , <i>recutita</i> , <i>Echinacea angustifolia</i> , <i>Emblica officinalis</i> , <i>Hydrocotyle asiatica</i> , Kali brom, <i>Thuja occidentalis</i> .
B37	Q-Sons	<i>Hydrocotyle asiatica</i> , <i>Echinacea angustifolia</i> , <i>Emblica officinalis</i> , <i>Calendula</i> , <i>Thuja occidentalis</i> .
B38	Unanifakiri	Sat loban, <i>Camphor</i> , Borax, <i>Plumbimonoxydum</i> , Kashgharisufaida, <i>Menthasylvestris</i> , Kanreet, Gadalfaza, <i>Ptychotisajowan</i> .
B46B39	Hamdard	<i>Camphora officinarum</i> , <i>Oleum eucalypti</i> , <i>Thymol</i> , <i>Oleum turpentine</i> , <i>Ferula gummosa</i> , Chlorophyll col
B40	Jalil Jildi Unani Dawakhana	Sulphur, <i>Psoralea corylifolia</i> , Copper Sulphate, Hydrargyri subchloridum, Sulsufaid.
B41	AL-Chemist	Jastphool, Boric acid, <i>Styrax benzoin</i> , <i>Ptychotisajowan</i> , <i>Menthasylvestris</i> , <i>Cinnamomum camphora</i> , Nees, Vasline.
Musaffi	Aftab Qarshi	<i>Swertiachirata</i> , <i>Fumaria officinalis</i> , <i>Sphaeranthus hirtus</i> , <i>Zizyphus vulgaris</i> , <i>Fagonia arbica</i> , <i>Artemisia absinthium</i> , <i>Melia azadirachta</i> , <i>Lowsonia alba</i>
B24	Qarshi	<i>Tinosporacardifolia</i> , <i>Fumaria officinalis</i> , <i>Swertiachirata</i> , <i>Sphaeranthus hirtus</i> , <i>Artemisia vulgaris</i> , <i>Terminalia chebula</i> , <i>Tephrosia purpurea</i> , <i>Zizyphus vulgaris</i> , <i>Melia azadirachta</i> , <i>Lycopodium clavatum</i>
B44	Ashraf Lab	<i>Pterocarpus Santalinus</i> , <i>Azadirachta indica</i> , <i>Fumaria indica</i> , <i>Swertiachirata</i> , <i>Smilax china</i> , <i>Smilax officinalis</i> , <i>Sphaeranthus indicus</i> , <i>Tephrosia purpurea</i> , <i>Cassia tora</i> (seeds), <i>Prunus domestica</i> , <i>Ipomoea turpethum</i> , <i>Terminalia chebula</i> (raw), <i>Cassia angustifolia</i> , <i>Cuscuta reflexa</i> , <i>Zizyphus sativus</i> , <i>Artemisia absinthium</i>

Table 12: Medicines used for different types of skin disease

Conclusion

The results of the present study indicated the poor quality of herbal products being sold in the market with very high label claims. Only 9 out of 46 sampled products with antimicrobial label claimed qualify to some extent for medicinal use. As any health authority (DRA/MOH) in Pakistan does not regulate the herbal products, there was no control or check on the manufacturers and prescriber of these medicines. All the tested products contained herbs, which had proven data for their antimicrobial activity, still most of them failed to produce any pharmacological results. We can conclude any of the following reasons.

1. These products had not been produced by the professional people.
2. They had not been processed properly.
3. Selection of herbal ingredients was not proper. Quality assurance played important role to monitor such shortcomings.
4. Common extraction processes for herbal product manufacturing were decoction, distillation, infusion, maceration and percolation. These processes need trained employees and appropriate equipment. Any deficiency in provision of both of these may lead to poor quality.

Recommendations

It is necessary to identify and rectify these problems immediately. Following recommendations were made based on present study:

1. Proper legislation must be done to regulate the manufacturing and prescription of Herbal/ Unani/Homeopathic medicines.
2. Only qualified pharmacists must be allowed to supervise the manufacturing of such products. The role of Botanist/Phytochemist is necessary for proper identification of herbs and their extraction process.
3. The herbal manufacturing industries should be issued manufacturing licenses as is done in case of pharmaceutical industries.

a. Herbal manufacturing industries should be forced to establish well equipped quality control labs to ascertain the efficacy and safety of their products.

4. Quality assurance is required to monitor all the activities related to identification, collection, drying, transportation, storage, and extraction of herbs, manufacturing of herbal product, stability of formulation, quality control and marketing.

5. As the properties of soil to soil vary it is impossible to procure or collect herbs with similar phytoconstituents every time, every industry must have its own Research and Development department (R&D).

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