Research Article

Assessment of physicochemcial properties and microbial contamination of marketed preparations

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ABSTRACT

Aim: The study aimed to evaluate the physicochemical properties and bacterial contamination of herbal medicinal preparations sourced from identified herbal retail outlets and herbal shops in distinct regions of Meerut. **Methodology:** The assessments of physicochemical properties and contamination of the herbal products were carried out using standard procedures. **Results:** Total solid content, alcohol- and water-soluble extractive values, loss of drying, ph assessment, and moisture level are found to be as per pharmacopeia standards. Further total yeast count and total aerobic viable count are also found to be near the standard values. No any herbal preparation contacting *Escherichia coli*, *Salmonella* Typhi, and *Staphylococcus aureus*. **Conclusion:** Hence, herbal medications in Meerut area are not likely to be contaminated with potentially pathogenic bacteria. The quality assurances of these products are according to pharmacopeia standards.

Keywords: Alcohol- and water-soluble extractive values, bacterial contamination, herbal preparations, Meerut, pathogenic bacteria

Introduction

In the written record, the study of herbs dates back over 5000 years to the Sumerians, who created clay tablets with lists of hundreds of medicinal plants such as myrrh and opium. The use of herbs to treat disease is almost universal among non-industrialized societies. Development of chemical and phytochemical analysis has led to the increasing use of herbal medicine for the treatment of human diseases. Many of the pharmaceuticals currently available to physicians have a long history of use as herbal remedies, including opium, aspirin, digitalis, and quinine. The World Health Organization (WHO) estimates that 80% of the populations of some Asian and African countries presently use herbal medicine for some aspect of primary healthcare.[1]

Ayurveda deals with a preventive and curative measure for the well-being of creature. One of the unique approaches of this traditional science is to treat each human individually. The treatment is planned by physician's own vision and wisdom (Yukti) with the proper administration of therapies in accordance of patient individualistic Dosha (humors), Prakriti (constitution), and Vikriti (disease condition). Each body is believed to determine the unique combination of physical, physiological, and psychological features of a creature. Acharya Charaka states; "a single drug may have many applications due to its diverse actions just as a man is able to perform various actions." For thousands of years, ingredients from Ayurvedic medicine have been connected to efficacy in a human being. Ayurveda does not follow the organ-oriented anatomy and physiology of conventional medical science. Ayurveda adopts its own function-oriented approach through its alternative theories of Panchamahabhuta (five basic elements, namely, Akasha, Vayu, Agni, Prathvi, and Jala), Tridosha (three humors, namely, Vata, Pitta, and Kapha).[2]

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Modified herbal medicines

The herbal medicines under this category has the same description as the medicines under 1 and 2 categories, except that the medicines this

category altered in certain way – either form or shape comprising dose, mode of administration, dosage form, medical indications, methods of preparation, and herbal medicinal ingredients. The medicines should meet the national regulatory board of herbal medicines requirement of efficacy and safety. All the imported herbal medicines come under this category which includes the products and raw materials.

The pharmaceutical industry is one of the pillar industries for economic development worldwide. In 2006, global spending on prescription drugs topped US$ 643 billion and the USA accounted for almost half of the global pharmaceutical market, with US$ 289 billion in annual sales. Nowadays, humans cannot live well without medicine, particularly in the developed countries. Although drug discovery has been driven by a variety of technology platforms, which can also expedite the development of therapeutic agents from herbal medicines, drug development remains a lengthy process with a low rate of success and huge capital investment. On average, it takes about 10–15 years for a newly synthesized compound to become a marketable therapeutic agent, and the cost in 2006 was approximately C 1 billion. In 2005, Big Pharma spent about US$ 50 billion, which is more than double the amount spent in 1996. However, in that year only 22 new drugs were approved by the Food and Drug Administration (FDA) in USA, whereas 53 new drugs reached the market in 1996. Despite the protracted time course of development, only one or two in 10,000 of such chemical compounds have proven to be clinically efficacious and safe for approval by regulatory agencies. In fact, about half of all drug candidates fail in the late stages of clinical trials. Furthermore, soon after their approval, some new drugs have to be withdrawn from the market due to severe side effects and clinical risks that are not detected in Phase III trials.[3]

Modification strategies of natural products

Maintaining the structure-activity relationship (SAR) is absolutely necessary. The long-known alkaloid cocaine was historically useful as a topical anesthetic in eye and nasal surgery, yet the major disadvantages of this use are cocaine's apostrophe intense vasoconstrictor activity and potential for cardiovascular toxicity. In the absence of information on target structures, classical medicinal chemistry methods are normally applied to the modification of natural products. SARs or quantitative SAR are explored to reveal and assign the pharmacophores, which guide the design of novel compounds with simplified or different scaffolds. Removal of unnecessary chiral centers provides an increase of activity strength and selectivity because of the appropriate binding to sterically-complementary and asymmetrical targets. The negative side is the difficulty in synthesis, separation, and resolution of single eutomers.[4]

Contamination of herbal formulation

Contaminants from source materials, such as microorganisms, microbial poisons, natural toxins or heavy metals, must check in herbal and conventional medicines. The assembly of completed products must be as per the good manufacturing practices (GMPs); with post-shown quality confirmation observation. The assessment of toxicity and antagonistic drug response of herbal planning was neglected territory, as herbs are considered normal and safe along these lines. This lack of data creates it intricate to look at the potential hazard profile of herbal medicines. In addition, for the vast majority of drugs, the correlation of common medicines with current drugs with relative adequacy has not been established.

Herbal medicines are claimed and commonly recognized to be valuable; conversely, there is been reports of intense and constant inebriation due to their use. The prevalence and accessibility of conventional cures have given rise to concerns about the well-being, viability, and duty of professionals using conventional cures. Heavy metals are considered to be a contaminant or an adulterant of such common remedies. Asian and Indian herbal remedies were statement to enclose elevated amounts of arsenic, lead, and mercury and lower levels of gold, and so forth.[5]

About 20% of 70 Ayurvedic herbal medicinal products carrying possibly dangerous amounts of poisonous heavy metals (American Medical Association 2004) in Ayurvedic medicinal products. Theory assigns an significant therapeutic function to metals like mercury; arsenic; and lead 35–40% of Ayurvedic medicinal products deliberately produce at least one metal. The plants with medicinal value are attributed to a diverse contamination of microbes such as fungal spores and bacteria (Fungi and bacterial endospores). In addition, the varied diversity of viruses, fungal cells, and bacteria could be identified either within the plant material or on it. Among the different micro-organisms, pathogens tend to limit the usage of such plants. Pesticides are concoction mixtures that can be used to monitor or eliminate disturbances. Which are assigned pest sprays, fungicides, nematocides, herbicides, rodenticides, and others (e.g., ascaricides and molluscicides) on the basis of coordinated usage. They are gathered as organochlorine pesticides (hexachlorocyclohexanes or benzene hexachlorides, lindane, and dichlorodiphenyl trichloroethane), organophosphorus pesticides (chlorpyrifos and methylchlorpyrifos, coumaphos, dichlorvos, ethion, fenchlorphos, Malathion, and parathion), and nitrogen-containing pesticides. An actual object contaminates the food is known as physical contamination. At times, such physically contaminated food will results in the contamination of biologicals. The physical contamination could bring hazardous bacteria. Fingernail is an example of physical contamination. The physical contamination may occur from various sources such as hair, glass or metal, pests, jewellery, dirt, and finger nails.[6] Some number of contacts toward the ionizing radiation is unavoidable because there exist various sources which encompasses radionuclides taking place natural in the atmosphere and ground as well. Hazardous contamination can be the impact of accident.[7]

Negative health impact of herbal preparations on human health

Each type of medicine has many strengths and weaknesses. To protect and improve our health, it is important to become an informed medical consumer. Herbal drugs are used widely for preventive and therapeutic purposes. The manufacturers of these products are not required to submit proof of safety and efficacy before marketing, so the adverse effects associated with remedies are largely unknown. Furthermore, herbal products are not regulated for purity and potency. Thus, some of adverse effects reported could be caused by impurities or batch to batch variability. The potency of herbal products

may increase the possibility of adverse effects. This paper highlights potential benefits and possible risks associated with consumption of herbal product so that conventional treatments can be made more safe and effective.[8] The toxicities include nephrotoxicity, hepatotoxicity, cardiotoxicity, neurotoxicity, and skin toxicity.

As the global use of herbal medicinal products continues to grow and many more new products are introduced into the market, public health issues, and concerns surrounding their safety are also increasingly recognized. Although some herbal medicines have promising potential and are widely used, many of them remain untested and their use also not monitored. This makes knowledge of their potential adverse effects very limited and identification of the safest and most effective therapies as well as the promotion of their rational use more difficult.

Assessment of quality control of herbal medicines

There is no gainsaying the fact that the requirements as well as the research protocols, standards, and methods needed for the evaluation of the safety and efficacy of herbal medicines are much more complex than those required for conventional or orthodox pharmaceuticals. A single herbal medicine or medicinal plant may contain hundreds of natural constituents, and a mixed herbal medicinal product may contain several times that number. Suppose every active ingredient was to be isolated from individual herb from which the herbal medicine is formulated or produced, the time and resources required would be tremendous. Such an analysis may practically be impossible especially where an herbal product is a mixture of two or more herbs.

The quality of the source materials used in the production of herbal medicines determines to a large extent the safety and efficacy of these herbal remedies. In general, the quality of source materials is dependent not only on intrinsic (genetic) factors but also on extrinsic factors such as environmental conditions, good agricultural, and good collection practices for medicinal plants, including plant selection and cultivation. It is the combination of these factors that make it difficult to perform quality controls on the raw materials of herbal medicines. According to GMP, correct identification of species of medicinal plants, special storage, and special sanitation and cleaning methods for various materials is important requirements for quality control of starting materials. One of the major challenges often encountered in the quality control of finished herbal medicinal products, especially mixture herbal products, is the difficulty in ascertaining the inclusion of all the plants or starting materials. Thus, the general requirements and methods for quality control of finished herbal products remain far more complex than for other pharmaceuticals. To ensure safety and efficacy of herbal medicines, therefore, WHO continues to recommend the institution of quality assurance and control measures such as national quality specification and standards for herbal materials, GMP for herbal medicines, labeling, and licensing schemes for manufacturing, import, and marketing, in countries where herbal medicines are regulated.

Herbal therapy is one of the best practices to overcome the illness. The quality assessment of herbal formulations is very important to justify their acceptance in modern system of medicines. It is thus mandatory that the microbiological limit tests of herbal medicinal preparations be done to ensure that the product is free from risk. Due to all these reasons standardization of herbal formulations is essential to assess the quality of the drug.[9]

Methodology

The investigations for the limit for microbes are intended to carry out the quantitative and qualitative evaluations of particular reliable microorganisms existing in the experimental samples. It comprises tests for the overall reliable count (fungi and bacteria) and *Escherichia coli*. The care must be mainly carried out to perform the tests, in order that the contamination of microbes from the outside might be neglected.

Study area and sampling

An overall five distinct preparations of herbal plants were randomly purchased from examined retail outlets and herbal shops in distinct regions of Meerut. Oral fluids includes for the evaluation of micro-organism quality. Bresol Syrup, Evacare Syrup, Joshina herbal remedy, Masturin, and Saduri syrup were taken to assess the microbial contamination.

Determination of physical as well as physiochemical properties

Physical evaluations were performed and this includes total solid content, alcohol- and water-soluble extractive values, and alcohol content has been analyzed and estimated based on the Indian Pharmacopoeia method.[10,11]

Total solid content

Determination of % of solid contents 4 g of herbal formulations was placed in a previously clean, dry, and weighed evaporating dish. The dish and herbal formulation was weighed again to confirm the exact weight of the herbal formulation. The liquid portion of the herbal formulation was evaporated by placing the evaporating dish on the hot plate. The weight and thus % of the solid contents of herbal formulation left after complete drying were calculated.[12]

Determination of extractive value

Accurately weighed herbal formulation was taken and macerated with 100 ml of 95% alcohol for 24 h in an air tight container. The contents were frequently shaken during the first 6 h and allowed to remain for 18 h. After 24 h, the extracts were filtered and filtrate was evaporated finally the extract was dried at 105°C to a constant weight and extractible value was calculated as % (w/w) with reference to air dried drug.[13]

Loss on drying

Weigh accurately about 1.5 g of the powdered drug in a tared porcelain dish and dried at 105°C in hot air oven to get constant weight and then weighed. From the difference in weight, the percentage loss on drying with reference to the air-dried substance was calculated.[14]

pH assessment

The pH meter Hanna microprocessor was the machine used to find out the pH of the herbal products according to the method proposed by Norris and Ribbons in the year 1970. The sample is suspended it in 100 ml sterilized distilled water in a beaker and thus the sample solution of 10% was produced as a homogenous solution. The pH meter as mentioned above was used to check it.[15]

Moisture level assessment

Mettler Toledo HR73 Halogen moisture analyzer was used to find out the content of moisture in the herbal preparations. One gram of sample was taken in a pan and the machine itself automated the entire process, that is, analysis and reading.[16]

Yeast and mold (fungus) contamination assessment

Ten grams of the sample were suspended in 100 ml of phosphate buffer with pH 7.2. Following this, 1 ml of the prepared mixture was then added to the 15 ml of liquefied potato dextrose agar medium as two partitions in Petri dishes. This was later incubated for 7 days at 25°C. Following this, the dishes were then monitored, and count for total colonies was taken.[17]

Number of total aerobic microbes

Ten grams of the sample were suspended in 100 ml of buffered sodium chloride-peptone solution with pH 7. Subsequently, polysorbate 80 of 0.1% w/v was added to assist the suspension of poorly wet table substances. Following this, about 15 ml of the liquefied casein soyabean digest agar and 1 ml of the preparation were added to two Petri dishes incubated at 30–35°C for 4 days and kept at not more than 45°C. Following this, the petri dishes were monitored, and colonies count was taken.[18]

E. coli assessment

In a sterile screw capped jar, 10 g sample was taken and then suspended it in a total of 100 ml of buffered lactose broth through vigorous shaking. Polysorbate 80 in 0.1% w/v was added. After this, the solution was transferred into a sterile container that can be capped with a screw and added 50 ml of nutrient broth. After shaking, the mixture was incubation for a total of 1 day at 37°C. Then, the dishes were evaluated for the availability of acid as well as gas according to established protocol.[19]

Salmonella assessment

In a sterile screw capped container, 1 g of sample was suspended in 100 ml of nutrient broth and allowed it untouched for a total of 240 min and after shaking it was incubation at a temperature of 35°C not more than 37°C for a time period of 1 day. From the improved culture, took 1 ml and added it to two cylinders that were already loaded with 10 ml of selenite F stock as well as tetrathionate bile-brilliant green broth. These were kept under incubation at 36°C to a maximum temperature of 38°C for 2 days. Further sample was cultured in brilliant green agar as well as in bismuth sulfite agar. After this process, these plates were kept under incubation at 37°C for 1 day. These plates were kept under observation for the appearance of pink or black-green colonies.[20]

Staphylococcus aureus assessment

Ten grams of the sample were suspended in 100 ml of nutrient broth and kept untouched for a period of 240 h and then shaken which is further incubation at 37°C for 1 day. From this, 1 ml was taken to Soyabean-Casein Digest media and was assessed for the growth availability. Part of the medium was then streaked in the plates containing Vogel-Johnson agar and Mannitol-salt agar (MSA) medium. These Petri plates were also kept under incubation at 37°C for 18 h. Appearances of yellow as well as black colonies identified as *Staphylococcus* and were assessed.[21]

Isolation and identification of *S. aureus*

One gram of the sample with peptone water (sterilized) in McCartney bottle and kept under incubation for about 18 h at 37°C. Isolation of *S. aureus* was done through streaking method, that is, streaking of the pre-enriched culture present in the peptone water in a chosen variant of agar plate that had MSA in it. The MSA was developed according to the rules and regulations of the product manufacturer. After this, the Petri plates were kept under incubation at 37°C for a 1 day under aerobic conditions. Observation of yellow color and completely colorless colonies identified as *Staphylococcus*. Such colonies were taken out and made to undergo biochemical tests, for instance, catalase, slide, and tube coagulase to confirm *S. aureus*.

Isolation and identification of *E. coli*

One gram of sample with sterile McCartney bottle that had sterilized nutrient broth in it and this was kept under incubation at 37°C. Then cultivated in variant *E. coli* media and kept under incubation at a temperature of 44°C. Streaking of inoculum with *E. coli* media was performed in eosin methylene blue agar and incubated. Identification of metallic sheen denotes the availability of lactose fermenters which need to be assured by *E. coli,* that is, IMVIC assessments as put forth according to Prescott *et al*.

Isolation and identification of *Salmonella* Typhi

One gram of sample with 10 ml of sterile cysteine broth was taken and incubated at 37°C. The upgraded culture was then cultivated in the alternative plates of freshly developed bovine serum albumin (BSA), and incubated with the control plate that had BSA. Black and green colonies denotes the availability of *Salmonella* which need to be assured by nutrient agar slants through next step- biochemical checking with the help of two assessments names triple sugar iron as well as lysine iron agar tests.

Results

Contamination in Brisol syrup

Total solid content in Brisol syrup was found to be 52 ± 0.52, alcohol- and water-soluble extractive values was found to be 0.158, pH was

observed as 5.95 ± 0.06, total yeast and mold count the standard value is 103 CFU/g and observed value is 101 CFU/g, total aerobic viable count the standard value is 105 CFU/g and the observed value is 104 CFU/g, assessment for *E. coli*, assessment for *S.* Typhi and assessment for *S. aureus*, from the above conducted study it was found that the values for all the evaluated parameters were normal or were found significant compared with the standard values as per Indian Pharmacopoeia [Table 1]. Microbial growth was indicated in Petri dish [Figure 1].

Contamination in Evecare syrup

Total solid content of Evecare Syrup was found to be 49 ± 0.32, alcohol- and water-soluble extractive values was found to be 0.132, pH was observed as 6.29 ± 0.06, total yeast and mold count the standard value is 103 CFU/g and observed value is 103 CFU/g, total aerobic viable count the standard value is 105 CFU/gm and the observed value is 105 CFU/gm, assessment for *E. coli*, assessment for *S.* Typhi, and assessment for *S. aureus*, from the above conducted study it was found that the values for all the evaluated parameters were normal or were found significant compared with the standard values as per Indian Pharmacopoeia [Table 2]. Microbial growth was indicated in Petri dish [Figure 2].

Table 1: Microbial analysis of (Brisol syrup)

Microbial analysis	Standard value	Observed value
Total solid content	As per I.P.	Found optimum
Alcohol- and water-soluble extractive values	As per I.P.	Found optimum
Loss on drying	As per I.P.	Found optimum
pH assessment	As per I.P.	Found optimum
Moisture level assessment	As per I.P.	Found optimum
Total yeast and mold count	103 CFU/g	101 CFU/g
Total aerobic viable count	105 CFU/g	104 CFU/g
Assessment for *E. coli*	Not present	Not present
Assessment for *S.* Typhi	Not present	Not present
Assessment for *S. aureus*	Not present	Not present

E. coli: *Escherichia coli*; *S.* Typhi: *Salmonella* Typhi; *S. aureus*: *Staphylococcus aureus*

Table 2: Microbial analysis of (Evecare syrup)

Microbial analysis	Standard value	Observed value
Total solid content	As per I.P.	Found optimum
Alcohol- and water-soluble extractive values	As per I.P.	Found optimum
Loss on drying	As per I.P.	Found optimum
pH assessment	As per I.P.	Found optimum
Moisture level assessment	As per I.P.	Found optimum
Total yeast and mold count	103 CFU/g	103 CFU/g
Total aerobic viable count	105 CFU/g	105 CFU/g
Assessment for *E. coli*	Not present	Not present
Assessment for *S.* Typhi	Not present	Not present
Assessment for *S. aureus*	Not present	Not present

E. coli: *Escherichia coli*; *S.* Typhi: *Salmonella* Typhi; *S. aureus*: *Staphylococcus aureus*

Contamination in Joshina herbal remedy

Total solid content of Joshina herbal a cough and cold remedy was found to be 51 ± 0.45, alcohol- and water-soluble extractive values were found to be 0.143, pH was observed as 6.10 ± 0.02, total yeast and mold count the standard value is 103 CFU/g and observed value is 100 CFU/g, total aerobic viable count the standard value is 105 CFU/g and the observed value is 104 CFU/g, assessment for *E. coli*, assessment for *S.* Typhi and assessment for *S. aureus*, from the above conducted study it was found that the values for all the evaluated parameters were normal or were found significant compared with the standard values as per Indian Pharmacopoeia [Table 3]. Microbial growth was indicated in Petri dish [Figure 3].

Contamination in Masturin

Total solid content of Masturin was found to be 50 ± 0.35, alcohol- and water-soluble extractive values were found to be 0.128, pH was observed as 5.43 ± 0.04, total yeast and mold count the standard value is 103 CFU/g and observed value is 103 CFU/g, total aerobic viable count the standard value is 105 CFU/g and the observed

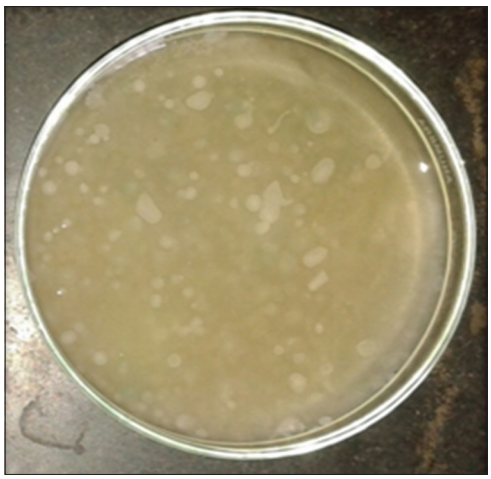

Figure 1: Microbial growth in Brisol syrup

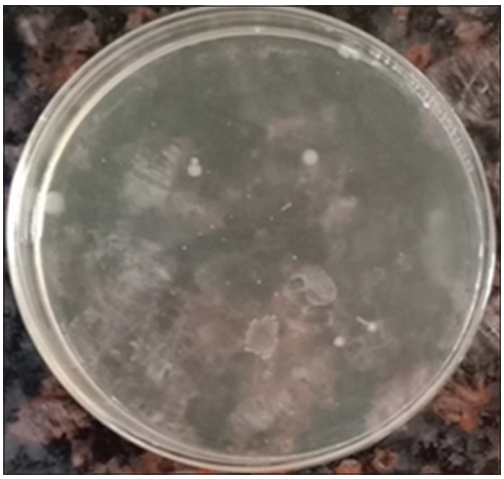

Figure 2: Microbial growth in Evecare syrup

value is 104 CFU/g, assessment for *E. coli*, assessment for *S.* Typhi and assessment for *S. aureus*, from the above conducted study it was found that the values for all the evaluated parameters were normal or were found significant compared with the standard values as per Indian Pharmacopoeia [Table 4]. Microbial growth was indicated in Petri dish [Figure 4].

Contamination in Saduri syrup

Total solid content of Saduri syrup was found to be 53 ± 0.25, alcohol- and water-soluble extractive values was found to be 0.167, pH was observed as 6.47 ± 0.04, total yeast and mold count the standard value is 103 CFU/g and observed value is 103 CFU/g, total aerobic viable count the standard value is 105 CFU/g and the observed value is 103 CFU/g, assessment for *E. coli*, assessment for *S.* Typhi and assessment for *S. aureus*, from the above conducted study it was found that the values for all the evaluated parameters were normal or were found significant compared with the standard values as per Indian Pharmacopoeia [Table 5]. Microbial growth was indicated in Petri dish [Figure 5].

Discussion

The results of the moisture content showed that there was remarkable variation among the different herbal preparations sampled. European Agency for the Evaluation of Medicinal products (1998) suggested that water content should be included in the list of comprehensive specifications for herbal medicinal products especially the powdered forms. The maximum moisture content limit of 8%/g of herbal preparations is satisfactory according to National Agency for FDA

Table 3: Microbial analysis of (Joshina herbal remedy)

Microbial analysis	Standard value	Observed value
Total solid content	As per I.P.	Found optimum
Alcohol- and water-soluble extractive values	As per I.P.	Found optimum
Loss on drying	As per I.P.	Found optimum
pH assessment	As per I.P.	Found optimum
Moisture level assessment	As per I.P.	Found optimum
Total yeast and mold count	103 CFU/g	100 CFU/g
Total aerobic viable count	105 CFU/g	104 CFU/g
Assessment for *E. coli*	Not present	Not present
Test for *S.* Typhi	Not present	Not present
Assessment for *S. aureus*	Not present	Not present

E. coli: *Escherichia coli*; *S.* Typhi: *Salmonella* Typhi; *S. aureus*: *Staphylococcus aureus*

Table 4: Microbial analysis of (Masturin)

Microbial analysis	Standard value	Observed value
Total solid content	As per I.P.	Found optimum
Alcohol- and water-soluble extractive values	As per I.P.	Found optimum
Loss on drying	As per I.P.	Found optimum
pH assessment	As per I.P.	Found optimum
Moisture level assessment	As per I.P.	Found optimum
Total yeast and mold count	103 CFU/g	103 CFU/g
Total aerobic viable count	105 CFU/g	104 CFU/g
Assessment for *E. coli*	Not present	Not present
Assessment for *S.* Typhi	Not present	Not present
Assessment for *S. aureus*	Not present	Not present

E. coli: *Escherichia coli*; *S.* Typhi: *Salmonella* Typhi; *S. aureus*: *Staphylococcus aureus*

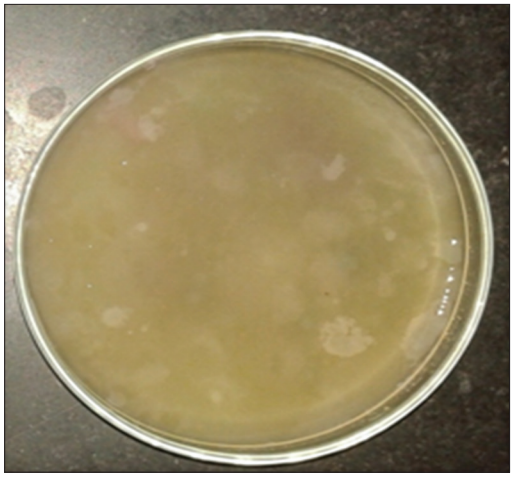

Figure 3: Microbial growth in Joshina herbal remedy

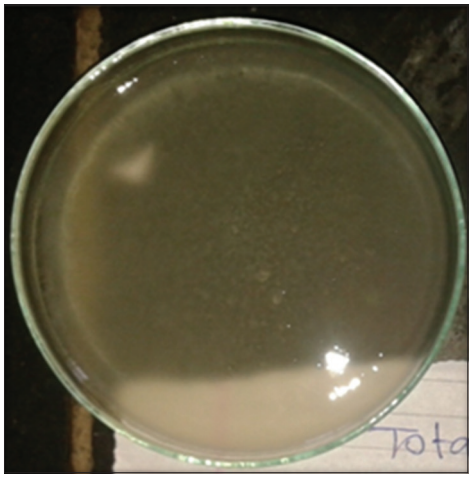

Figure 4: Microbial growth in Masturin

Figure 5: Microbial growth in Saduri syrup

Table 5: Microbial analysis of (Saduri syrup)

Microbial analysis	Standard value	Observed value
Total solid content	As per I.P.	Found optimum
Alcohol and water soluble extractive values	As per I.P.	Found optimum
Loss on drying	As per I.P.	Found optimum
pH assessment	As per I.P.	Found optimum
Moisture level assessment	As per I.P.	Found optimum
Total yeast and mold count	103 CFU/g	103 CFU/g
Total aerobic viable count	105 CFU/g	103 CFU/g
Assessment for *Escherichia coli*	Not present	Not present
Assessment for *Salmonella* Typhi	Not present	Not present
Assessment for *Staphylococcus aureus*	Not present	Not present

and Control.[22] In the current study shown that herbal products were within the limit stated according to IP.

Based on the pH values determined, five herbal products that were used in the study had acidic properties. When the pH value is low (acidic), the bacterial count was observed to be equally low, but at neutral or higher pH the level of contamination of the herbal preparations was observed to be higher. This suggests that a neutral or alkaline pH favored high contamination levels of the herbal preparations. This agrees with the observation that bacterial growth is optimal at more or less neutral pH, around pH 5–8.5.[23]

The limits of bacterial contamination given various pharmacopoeias are: Total aerobic bacteria (10^5 CFU/g), Enterobacteria and other Gram-negative organisms (10^3 CFU/g).[24] E. coli and Salmonella should be absent. The herbal products under study meet these specifications in most cases.

Conclusion

Various herbal formulations are assessed for various parameters and shown total solid content, alcohol- and water-soluble extractive values, pH assessment, total yeast and mold count, and total aerobic viable count in a permissible level. Further assessment for pathogenic bacteria such as *E. coli*, *S.* Typhi, and *S. aureus* is shown absence. Finally, all herbal preparations were found good at quality levels when compared with the pharmacopoeia standards.

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