ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 15, Issue 7, 2022

Online - 2455-3891
Print - 0974-2441

Original Article

AN ASSESSMENT OF THE MICROBIOLOGICAL QUALITY OF ANTI-VIRAL CHURNA SOLD IN INDIA BY AYURVEDIC MEDICAL PRACTITIONERS

SARFARAZ KAZI[1]*, SANJAY BAIS[2]

[1]Research Scholar, JJT University, Jhunjhunu, Rajasthan, India. [2]Research Supervisor, JJT University, Jhunjhunu, Rajasthan, India.
Email: sarfarazkazi81@gmail.com

Received: 16 March 2021, Revised and Accepted: 20 April 2022

ABSTRACT

Objectives: The objectives behind of this study are to analyze the unregistered herbal products dispensed by Ayurvedic practitioners, as per the WHO guidelines for determination of potentially hazardous contaminants. To aware the registered medical practitioners regarding contamination of herbal product, the patients are using unregistered herbal products from registered medical practitioners.

Methods: The method employed in this study was a pour plate method. The specific media plates were incubated for 24 h at 37°C. On a colony counter, the plate was placed and observed for the presence of micro-organisms. The media used were nutrient agar medium and cetrimide nutrients, agar, saline agar, and MacConkey agar.

Result: The present study found microbial contamination in herbal products that are widely available across the country. The formulations coded AV5, AV6, and AV7 were found to be contaminated by *Pseudomonas aeruginosa* and AV5 and AV6 were found to be contaminated by *Escherichia coli*, whereas the formulations coded AV1 and AV2 were found to be contaminated by *Staphylococcus aureus* in above permissible limit, posing a risk of infection if consumed.

Conclusion: According to the findings of this study, it shows that few herbal formulations were showing the presence of micro-organism above the permissible of given by the WHO.

Keywords: Herbal formulation, Viral disease, Microbial load.

 DOI: http://dx.doi.org/10.22159/ajpcr.2022v15i7.44679. Journal homepage: https://innovareacademics.in/journals/index.php/ajpcr

INTRODUCTION

Herbal plant preparations and materials with therapeutic value that being used either raw or processed substances are known as herbal medicines. One or more plants, inorganic minerals, or animal products are derived from many herbs, inorganic minerals, or animal products. Herbal medicinal preparations are created and distributed. They are most commonly manufactured and sold on the clinical practice today for the treatment and prevention of illness, thereby promoting global public health [1].

Herbal medications have been utilized as cures and treatments for a variety of ailments since ancient times. Although Western pharmaceuticals play a large part in advanced medicine, herbal medicines is still consumed by about 60% of the people in backward regions [2]. In India, the raw ingredients of herbal medications are susceptible to fungal infections due to unprofessional methods of collection, storage, and transportation. Raw materials are obtained in an unprofessional manner and are frequently exposed to a wide range of microbiological contaminants. Microorganisms may grow on raw materials before harvesting, as well as during handling and storage [3]. The climate and the quality of the raw ingredients employed while manufacturing have an impact on the microbiological quality of medications.

CONTAMINATION SOURCES IN HERBAL PRODUCTS [4]

The microbial contamination may appear from variety of sources, including:

- Medicinal plants are grown or harvested in a variety of environments and situations.
- The conditions in which the goods are stored and the conditions in which they are transported.
- Patients' unsanitary use of medications.
- The manufacturing techniques used to create ready-to-use pharmaceutical medicines (Table 1).

Chronic lung disease

Chronic obstructive pulmonary disease affects a prominent reason of death and morbidity around the world [6,7]. Chronic inflammation of the tiny airways is the hallmark of COPD. A respiratory tract infection is a common cause of acute exacerbation and disease progression [8]. Since ancient times, it was observed that bacteria have been considered the main infectious cause of COPD exacerbations [9]. However, the viral upper respiratory tract infection (URI) is a key risk factor associated with exacerbations of COPD [10]. About 40%–60% of all COPD exacerbations are evidenced with the top respiratory tract infections (URIs) and viral infections (VIRs) proposed as important causes of COPD exacerbations [11]. Indeed, respiratory viruses such as rhinovirus, influenza, and respiratory syncytial virus (RSV) aggravate COPD [12-14].

FORMULATION DETAILS

The herbal formulation mainly contains Swertia chirata, Triphala, Haridra, Kantakari, Brhati, Sunthi, Marica, Murva, Guduchi, Dhanvayasa, Karcura, Daruharidra Katuka, Parpata Pippali, Trayamanag, Nimba (chhal), Puskara, Yasti, Hrivera Kutaja, Yavani, Musta, Indrayava, Bharang, and Sigru that were selected for study in this research.

Table 1: WHO limits for microbial contamination [5]

Microorganism finished product	CFU/g
Escherichia coli	10^1/g
Staphylococus aureus	10^5/g
Pseudomonas aeruginosa	103/g
Salmonella species	Nil

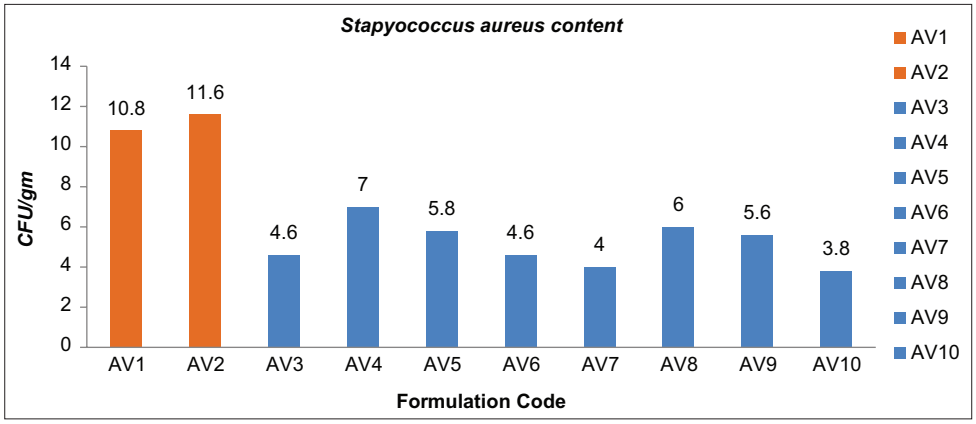

Fig. 1: Summarize the findings

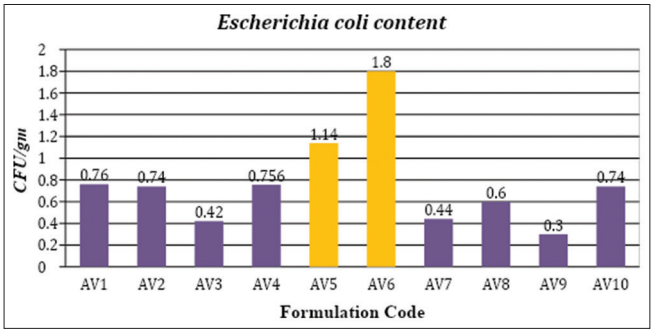

Fig. 2: Summaries the findings

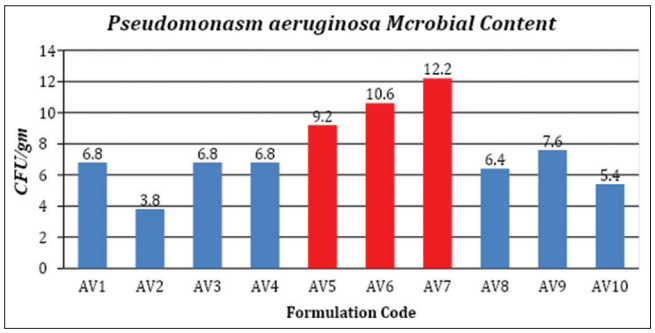

Fig. 3: Summarized the findings

Table 2: Results of Microbial load in Antiviral Churnas dispensed by RMP

Formulation code	*Pseudomonasm aeruginosa* Mean±SD (10^3 CFU/g)	*Escherichia coli Mean±SD* (10^1 CFU/g)	*Staphylococcus aureus* Mean±SD (10^5 CFU/g)	*Salmonelal typhi* Mean±SD NIL CFU/g
AV1	6.8±0.8366	0.76±0.85	10.8±1.4832	---
AV2	3.8±0.8366	0.74±0.1483	11.6±1.1401	---
AV3	6.8±0.8366	0.42±0.3563	4.6±2.0736	---
AV4	6.8±0.8366	0.756±0.1751	7±1.5811	---
AV5	9.2±0.8366	1.14±0.4878	5.8±1.4832	---
AV6	10.6±1.5165	1.8±0.8366	4.6±1.1401	---
AV7	12.2±0.8366	0.44±0.2073	4±1.5811	---
AV8	6.4±1.5165	0.6±0.187	6±1.5811	---
AV9	7.6±1.1401	0.3±0.1581	5.6±2.3021	---
AV10	5.4±2.0736	0.74±0.2701	3.8±0.8366	--

SAMPLE COLLECTION FOR EXPERIMENTAL WORK

The 10 herbal formulations dispensed by registered medical practitioners were gathered from Solapur district India's area.

Materials and procedures

The viability of the samples was tested using serial dilutions.

The pour plate technique was employed. The specific media plates were incubated for 24 h at 37°C. On a colony counter, the plate was placed. The colony morphology was observed. The specific media used were nutrient agar medium and cetrimide nutrients, agar, saline agar, and MacConkey agar [15].

Identifying the pathogen

Staphylococcus aureus identification

The herbal sample was dissolved in 10 mg of tryptic soya broth and incubated for 24 h at 37°C. After that, the herbal sample was streaked on Vogel-Johnson agar and cultured for 24 h at 37°C. Each plate was then replated with a single colony on mannitol salt agar and incubated at 37°C for 24 h. The colony morphology was found after incubation [16]. Triplicate reading of every herbal formulations was evaluated and mean and standard deviation were noted (Fig. 1).

Escherichia coli identification

The pretreatment material was prepared by dissolving 10 g of the sample in lactose broth (pH can be adjusted to 7). The above mentioned homogenized pretreatment material was incubated at 43–45°C for 18–24 h. Pre-treatment material containing 1 g of the sample being studied in 100 ml of MacConkey broth. The presence of *E. coli* was confirmed by the growth of red, non-mucoid, and colonies of Gram-negative rods, surrounded by a crimson zone of precipitation [17]. Triplicate reading of every herbal formulation was evaluated and mean and standard deviation were noted (Fig. 2).

Pseudomonasm aeruginosa identification

On a cetrimide agar plate, the diluted sample was streaked. The colonies were examined having green color for oxidase reactivity and

subcultured into triple sugar iron media after 24 h of incubation at 37°C. Bacterial growth and response outcomes were detected [18]. Triplicate reading of every herbal formulation was evaluated and mean and standard deviation were noted (Fig. 3).

Isolation and identification of Salmonella typhi

The 1.0 g sample was transferred selenite cysteine broth and was incubated for 24 h at 37°C. After incubation, the enhanced culture was streaked on the specific media plates of freshly made bismuth sulfite agar (BSA) and incubated at 37°C for 24 h alone with the control plate of bismuth sulfite agar (BSA). Triple sugar iron (TSI) and lysine iron agar (LIA) assay was employed and the colonies were streaked on the nutrient agar slants for counting the microbial content [19]. Triplicate reading of every herbal formulation was evaluated and mean and standard deviation were noted (Table 2).

RESULTS AND DISCUSSION

The anti-viral herbal formulations were tested for the microbiological quality. We report *Staphylococcus aureus* identification on the basis WHO guidelines. We also report *Escherichia coli* identification and *Pseudomonasm aeruginosa* identification. The some herbal formulations were found to be toxic and formulation modification was thought of in the future. The present study found microbial contamination in herbal products that are widely available across the country. The formulations coded AV5, AV6, and AV7 were found to be contaminated by *Pseudomonas aeruginosa* and AV5 and AV6 were found to be contaminated by Escherichia coli, whereas the formulations coded AV1 and AV2 were found to be contaminated by *Staphylococcus aureus* in above permissible limit, posing a risk of infection if consumed by for decades, herbal formulations have been widely used. According to the findings of this study, the good agriculture practices (GAP) and good laboratory practices (GLP) are need to focused while manufacturing of such products quality and safety.

CONCLUSION

The present work, which was taken, is bonafide and novel work on an assessment of the microbiological quality of anti-viral churna. We have made an attempt in reviewing the literature on the microbiological quality of anti-viral herbal formulations with the help of chemical abstract, journals, and internet surfing. For the microbiological quality of anti-viral herbal formulations, the WHO guidelines were followed. Around 10 samples of herbal formulations were tested. The herbal formulations were observed for their microbiological quality assessment followed by the WHO guidelines. The samples of herbal formulation were found to be toxic in nature. The proposed samples were screened for their microbiological quality against the microbial load in the WHO guidelines. The present work is an attempt in this direction and the efforts have proved to be quite fruitful and promising. It will be worthwhile to concentrate on microbiological quality of anti-viral herbal formulations.

ACKNOWLEDGMENTS

I am very thankful to Dr. Sandip Patil, Managing Director, Biocyte Institute of Research and Development, Sangli, Maharashtra for providing research facilities. I would wish to convey my regards to Dr. S.K. Bais Research Supervisor, JJT University, Jhunjhunu, Rajasthan for his valuable advice and suggestions during research work.

AUTHORS CONTRIBUTION

This research was carried out by Mr. Sarfaraz Kazi and the research data were finalized by Dr. Sanjay Bais.

CONFLICT OF INTEREST

The authors not have conflict of interests regarding publication of this article.

AUTHORS FUNDING

I hereby declare that no funding was provided by any agency for doing this research work.

REFERENCES

1. Choi DW, Kim JH, Cho SY, Kim DH, Chang SY. Regulation and quality control of herbal drugs in Korea. Toxicology 2002;181-182:581-6. doi: 10.1016/s0300-483x(02)00487-0, PMID 12505370
2. Khanyile ZC, Singh N, Smith M, Shode FO, Mngomezulu S, Dewir YH. Camparative assesment of Bacterial contamination in commercial herbal product of *Lessertia futescens*. Am Eur J Agric Environ Sci 2009;5:494-9.
3. Dubey NK, Kumar A, Singh P, Shukla R. Microbial contamination of raw materials: A major reason for the decline of India's share in the global herbal market. Curr Sci 2008;95:25.
4. Chan K. Some aspects of toxic contaminants in herbal medicines. Chemosphere 2003;52:1361-71. doi: 10.1016/S0045-6535(03)00471-5, PMID 12867165
5. Available from: https://www.researchgate.net/publication/320306420_Quality_Control_of_Herbal_Drugs
6. Vestbo J, Hurd SS, Agustí AG, Jones PW, Vogelmeier C, Anzueto A, *et al.* Global strategy for the diagnosis, management, and prevention of chronic obstructive pulmonary disease: Gold executive summary. Am J Respir Crit Care Med 2013;187:347-65. doi: 10.1164/rccm.201204-0596PP, PMID 22878278
7. Mathers CD, Loncar D. Projections of global mortality and burden of disease from 2002 to 2030. PLoS Med 2006;3:e442. doi: 10.1371/journal.pmed.0030442, PMID 17132052
8. Sethi S. Bacterial infection and the pathogenesis of COPD. Chest 2000;117;286S-91. doi: 10.1378/chest.117.5_suppl_1.286s, PMID 10843957
9. Mallia P, Johnston SL. How viral infections cause exacerbation of airway diseases. Chest 2006;130:1203-10.
10. Proud D, Chow CW. Role of viral infections in asthma and chronic obstructive pulmonary disease. Am J Respir Cell Mol Biol 2006;35:513-8. doi: 10.1165/rcmb.2006-0199TR, PMID 16778148
11. Tan WC, Xiang X, Qiu D, Ng TP, Lam SF, Hegele RG. Epidemiology of respiratory viruses in patients hospitalized with near-fatal asthma, acute exacerbations of asthma, or chronic obstructive pulmonary disease. Am J Med 2003;115:272-7. doi: 10.1016/s0002-9343(03)00353-x, PMID 12967691
12. Mallia P, Contoli M, Caramori G, Pandit A, Johnston SL, Papi A. Exacerbations of asthma and chronic obstructive pulmonary disease (COPD): Focus on virus induced exacerbations. Curr Pharm Des 2007;13:73-97. doi: 10.2174/138161207779313777, PMID 17266589
13. Ko FW, Ip M, Chan PK, Chan MC, To KW, Ng SS, *et al.* Viral etiology of acute Exacerbations of COPD in Hong Kong. Chest 2007;132:900-8. doi: 10.1378/chest.07-0530, PMID 17573516
14. Martinello RA, Esper F, Weibel C, Ferguson D, Landry ML, Kahn JS. Human *Metapneu-movirus* and exacerbations of chronic obstructive pulmonary disease. J Infect 2006;53:248-54. doi: 10.1016/j.jinf.2005.11.010, PMID 16412516
15. Mukharji PK, Rai S, Bhattacharya S, Wahile A, Pada Saha B. Marker analysis of poly herbal formulation Triphala-A well-known Indian traditional medicine. Indian J Trad Knowl 2008;7:379-83.
16. Okunlola A, Adewoyin BA, Odeku OA. Evaluation of pharmaceutical and microbial qualities of some herbal medicinal products in South Western Nigeria. Trop J Pharm Res 2007;6:661-70. doi: 10.4314/tjpr.v6i1.14644
17. Waterman RF, Sumner ED, Baldwin JN, Warren FW. Survival of *Staphylococcus aureus* on pharmaceutical oral solid dosage forms. J Pharm Sci 1973;62:1317-20. doi: 10.1002/jps.2600620818, PMID 4725177
18. Vandoorne H, Claushuis EP. The quantitative determination of *Enterobacteriaceae* in pharmaceutical preparations. Int J Pharm 1979;4:119-25. doi: 10.1016/0378-5173(79)90059-0
19. Abba D, Inabo HI, Yakubu SE, Olonitola OS. Contamination of herbal medicinal products marketed in Kaduna metropolis with selected pathogenic bacteria. Afr J Tradit Complement Altern Med 2009;6:70-7.