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COMPARATIVE DETERMINATION OF POTENTIAL CONTAMINANTS IN MAHARASNADI KADHA HERBAL FORMULATION AS PER WHO GUIDELINES.

Priya S. Thote*, Dr. Suhas N. Sakarkar, Shrikant D. Mahajan, Sachin Dudhe

Maharashtra Institute of Pharmacy (B. Pharm), Betala, Bramhpuri, Chandrapur, (MS) 441206.

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***Corresponding Author**

Priya S. Thote

Maharashtra Institute of Pharmacy (B. Pharm), Betala, Bramhpuri, Chandrapur, (MS) 441206.

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ABSTRACT

Herbal medicines are widely used to manage various health conditions, but their safety and quality can sometimes be affected by contaminants like heavy metals, microbes, pesticide residues, and aflatoxins. In this study, we evaluated the quality and safety of five popular Maharasnadi Kadha products following guidelines from the World Health Organization (WHO) and AYUSH standards. We looked at physical and chemical properties such as taste, smell, pH, and total solid content. We also tested for heavy metals (lead, arsenic, cadmium, and mercury), microbial contamination, pesticide residues, and aflatoxins using standard methods. The results showed that all five products had good physical and chemical qualities, with pH levels between 4.13 and 4.50. Heavy metal levels were within safe limits, and mercury was undetectable in all samples. Microbial counts met WHO standards, and no harmful microbes were found. Pesticide residues and aflatoxins were either not detected or were within safe limits. Overall, all the tested Maharasnadi Kadha formulations met WHO and AYUSH quality standards, indicating they are safe and of good quality. These findings highlight the importance of regular quality checks and regulatory oversight to ensure the safety, reliability, and wider acceptance of Ayurvedic herbal products worldwide.

KEYWORDS: Heavy metals, microbes, pesticide residues, and aflatoxins.

01. INTRODUCTION

01.1. HERBAL FORMULATION

Herbal medications are widely used worldwide and are an essential component of traditional healthcare systems. In Ayurveda, for example, polyherbal formulations are prescribed for degenerative and chronic illnesses. A traditional Ayurvedic formulation, Maharasnadi Kadha is frequently used for inflammatory diseases and musculoskeletal ailments including Amavata and Sandhivata. Because of its extensive use and prolonged administration, it is crucial to guarantee its safety, quality, and purity.[1]

Herbal formulations are vulnerable to contamination at many stages, such as cultivation, collecting, processing, storage, and distribution, even though they come from natural sources. If present in excess of allowable levels, potential pollutants include heavy metals, pesticide residues, microbial load, aflatoxins, and leftover solvents may cause major health problems. Strict quality control has been stressed by the World Health Organization (WHO).

Despite their natural origin, herbal formulations are susceptible to contamination at various stages including cultivation, collection, processing, storage, and distribution. Potential contaminants such as heavy metals, pesticide residues, microbial load, aflatoxins, and residual solvents may pose serious health risks if present beyond permissible limits. The World Health Organization (WHO) has emphasized the need for stringent quality control measures and safety evaluation of herbal medicines to ensure consumer safety and global acceptance.[2]

01.2. *Maharasnadi Kadha*

Maharasnadi Kadha is a classical Ayurvedic formulation composed of multiple medicinal plant ingredients, traditionally indicated in conditions such as *Amavata*, *Sandhivata*, *Gridhrasi*, and other *Vata*-dominant disorders. The formulation is known for its anti-inflammatory, analgesic, and immunomodulatory properties and is often prescribed for long-term use. Given its complex composition and prolonged administration, ensuring the safety, quality, and purity of *Maharasnadi Kadha* is of utmost importance.[3]

Despite their therapeutic benefits, herbal formulations are vulnerable to contamination at various stages of production. The quality of raw materials can be affected by environmental pollution, agricultural practices, and improper harvesting methods. Contaminants may be introduced during cultivation through polluted soil, irrigation water, pesticide application,

and industrial emissions. Additionally, improper processing, storage, and transportation conditions may lead to microbial contamination and the production of toxic metabolites such as aflatoxins. These contaminants can significantly compromise the safety and efficacy of herbal medicines.

Potential contaminants in herbal formulations include heavy metals such as lead, mercury, cadmium, and arsenic; pesticide residues; microbial contaminants; aflatoxins; and residual solvents. Heavy metals may accumulate in medicinal plants due to soil contamination or atmospheric deposition and can cause serious toxic effects upon chronic exposure. Microbial contamination may result in spoilage of the formulation and pose health risks, especially in immunocompromised individuals. Aflatoxins, produced by certain fungal species, are highly toxic and carcinogenic, making their detection and control essential. Pesticide residues, commonly used in agricultural practices, may persist in plant materials and contribute to long-term toxicity.[4]

Recognizing these safety concerns, the World Health Organization has developed comprehensive guidelines for the quality control, safety assessment, and standardization of herbal medicines. WHO guidelines emphasize the evaluation of contaminants and residues as a critical component of herbal drug quality assurance. These guidelines provide permissible limits and recommended analytical methods for the detection of heavy metals, microbial load, pesticide residues, and aflatoxins in herbal formulations. Adherence to these guidelines is essential to ensure the safety of herbal medicines and to promote their acceptance in national and international healthcare systems.

In recent years, there has been increasing global scrutiny regarding the quality and safety of traditional herbal products, including Ayurvedic formulations. Reports of contamination and variability in quality have raised concerns among regulatory authorities and consumers alike. Therefore, scientific evaluation and comparative analysis of herbal formulations manufactured by different sources are necessary to assess compliance with established safety standards. Comparative determination of potential contaminants in *Maharasnadi Kadha* can provide valuable insights into variations in manufacturing practices and quality control measures.[5]

The present study focuses on the comparative determination of potential contaminants in *Maharasnadi Kadha* herbal formulation as per WHO guidelines. By systematically

evaluating heavy metals, microbial load, pesticide residues, and aflatoxins, this study aims to generate reliable scientific data on the safety profile of the formulation. Such an evaluation not only contributes to quality assurance and standardization of *Maharasnadi Kadha* but also supports evidence-based validation of Ayurvedic medicines. Ultimately, this approach aligns with WHO's objective of integrating traditional medicines into modern healthcare systems through rigorous scientific assessment and quality control.[6]

01.3. INGREDIENTS OF MAHARASNADI KADHA

1. Rasna (*Pluchea lanceolata*)- Zygophyllaceae is one of the most important medicinal plants used in Ayurveda and serves as the principal ingredient of Maharasnadi Kadha. It is highly valued for its Vatahara (Vata-pacifying) properties and has been extensively employed in the treatment of musculoskeletal and neurological disorders. Classical Ayurvedic texts describe Rasna as an effective remedy for conditions associated with aggravated Vata dosha, including joint pain, arthritis, stiffness, sciatica, and rheumatic diseases.[7]

2. Dhanvayasa (*Fagonia arabica*)- Asteraceae is a well-known medicinal herb in Ayurveda, valued for its blood-purifying, rejuvenating, and therapeutic properties. It is traditionally used in the management of fever, skin diseases, liver disorders, anemia, and inflammatory conditions. Ayurvedic texts describe Dhanvayasa as having Pitta-shamaka and Raktashodhaka properties, making it beneficial for disorders associated with blood and liver dysfunction.[8] The plant contains several bioactive constituents such as flavonoids, saponins, alkaloids, triterpenoids, glycosides, tannins, and phenolic compounds, which contribute to its medicinal value. Due to its rich phytochemical profile and broad spectrum of biological activities, Dhanvayasa is widely incorporated into Ayurvedic formulations intended for improving overall health, enhancing immunity, and treating chronic inflammatory disorders.[9]

3. Bala (*Sida cordifolia*)- Malvaceae is an important Ayurvedic medicinal plant widely used as a **Balya (strength-promoting)** and **Rasayana (rejuvenative)** drug. It is traditionally employed in the treatment of neurological disorders, muscular weakness, arthritis, inflammatory conditions, and respiratory ailments. The roots and whole plant are rich in alkaloids, flavonoids, sterols, and other bioactive- compounds that contribute to its therapeutic efficacy.[10]

4. Eranda Moola (*Ricinus communis*)-belonging to family Euphorbiaceae is a widely used Ayurvedic drug renowned for its Vatahara (Vata-pacifying) and Shothahara (anti-inflammatory) properties. The root is particularly employed in the management of arthritis, sciatica, rheumatism, low back pain, and neuromuscular disorders. It contains bioactive compounds such as ricinoleic acid, flavonoids, sterols, and triterpenoids that contribute to its therapeutic actions.[11]

5. Devadaru (*Cedrus deodara*)- belonging to family Pinaceae is a large evergreen conifer highly valued in Ayurveda for its **Vata-Kapha pacifying**, anti-inflammatory, and analgesic properties. The heartwood is commonly used in the treatment of arthritis, rheumatism, edema, respiratory disorders, skin diseases, and neurological ailments. Its therapeutic effects are mainly attributed to the presence of volatile oils rich in himachalenes and cedrol, along with

flavonoids and other phenolic compounds.[12]

6. Shati (*Hedychium spicatum*) - belonging to the family Zingiberaceae, is an aromatic medicinal herb and is widely used in Ayurveda for respiratory, digestive, and inflammatory disorders. The rhizome is the principal medicinal part and possesses **Kapha-Vata pacifying** properties. It contains essential oils rich in cineole, limonene, and other terpenoids that contribute to its therapeutic effects. Traditionally, Shati is used in the management of cough, asthma, bronchitis, fever, indigestion, and inflammatory conditions.[13]

7. Vacha (*Acorus calamus*)- belonging to family Acoraceae is an important Ayurvedic medicinal plant, renowned for its **Medhya (brain tonic)** and **Vata-Kapha pacifying** properties. The rhizome is extensively used in traditional medicine for enhancing memory, improving speech, treating neurological disorders, digestive ailments, and respiratory conditions. Its therapeutic activities are attributed to the presence of essential oils rich in α-asarone and β-asarone, along with flavonoids and other bioactive compounds. Modern pharmacological studies have demonstrated significant neuroprotective, anti-inflammatory, antioxidant, antimicrobial, and cognitive-enhancing effects, supporting its traditional use in neurological and gastrointestinal disorders.[14]

8. Vasa (*Adhatoda vasica*)- belonging to family Acanthaceae is a renowned medicinal plant in Ayurveda, widely used for the management of respiratory disorders. It is known for its **Kapha-Pitta pacifying** properties and is particularly effective in conditions such as cough, bronchitis, asthma, and respiratory congestion. The leaves are the most commonly used part and contain quinazoline alkaloids such as **vasicine** and **vasicinone**, which are responsible for its bronchodilatory and expectorant actions. Modern pharmacological studies have confirmed its expectorant, antitussive, anti-inflammatory, and antioxidant activities, supporting its traditional use in respiratory and inflammatory disorders.[15]

9. Shunthi (*Zingiber officinale*) - belonging to family Zingeberaceae, the dried rhizome of ginger is one of the most important medicinal plants in Ayurveda. It is known for its **Deepana (appetizer)**, **Pachana (digestive)**, and **Vata-Kapha pacifying** properties. Shunthi is widely used in the treatment of indigestion, nausea, arthritis, respiratory disorders, and inflammatory conditions. Its medicinal value is attributed to bioactive compounds such as **gingerols, shogaols, zingerone, and essential oils**, which exhibit potent antioxidant and anti-inflammatory activities. Modern studies have validated its analgesic, antiemetic, antimicrobial, cardioprotective, and immunomodulatory effects, supporting its extensive use in traditional and modern medicine.[16]

10. Haritaki (*Terminalia chebula*)- belonging to family Combretaceae is one of the most revered medicinal plants in Ayurveda and is known as **"Abhaya"**, meaning "fearless," due to its wide range of therapeutic benefits. It is a major constituent of the famous Ayurvedic formulation **Triphala** and is traditionally used for digestive disorders, constipation,

respiratory ailments, skin diseases, and rejuvenation therapy. The fruit is rich in tannins, chebulagic acid, chebulinic acid, gallic acid, and other polyphenolic compounds responsible for its potent biological activities. Modern pharmacological studies have demonstrated significant antioxidant, anti-inflammatory, antimicrobial, hepatoprotective, antidiabetic, and immunomodulatory properties, supporting its traditional use as a Rasayana (rejuvenative) drug.[17]

11. Chavya (*Piper chaba*)- belonging to family Piperaceae is an important medicinal plant in Ayurveda, valued for its **Deepana (appetizer)** and **Pachana (digestive)** properties. It is widely used in the treatment of indigestion, flatulence, abdominal pain, respiratory disorders, and rheumatic conditions. The fruit and root contain bioactive constituents such as **piperine, chavicine, piperlongumine, and essential oils**, which contribute to its therapeutic efficacy. Modern pharmacological studies have demonstrated significant anti-inflammatory, antimicrobial, antioxidant, analgesic, and hepatoprotective activities. Chavya is also known to enhance the bioavailability of various drugs and herbal constituents, making it an important ingredient in many Ayurvedic formulations.[18]

12. Musta (*Cyperus rotundus*) - belonging to family Cyperaceae is a highly esteemed Ayurvedic medicinal herb known for its **Deepana (appetizer)**, **Pachana (digestive)**, and **Tridosha-balancing** properties. The rhizome is widely used in the treatment of digestive disorders, fever, diarrhea, dysentery, menstrual irregularities, and inflammatory conditions. Its therapeutic actions are attributed to the presence of essential oils rich in **cyperene, cyperotundone, rotundone**, and other sesquiterpenes, along with flavonoids and phenolic

compounds. Modern pharmacological studies have demonstrated significant anti-inflammatory, antipyretic, antimicrobial, antioxidant, and gastroprotective activities, supporting its traditional use in gastrointestinal and systemic disorders.[19]

13. Punarnava (*Boerhaavia diffusa*)- belonging to family Nyctagenaceae is a renowned Ayurvedic medicinal plant widely used for its **Mutrala (diuretic)**, **Shothahara (anti-inflammatory)**, and **Rasayana (rejuvenative)** properties. The root and whole plant are employed in the treatment of edema, kidney disorders, liver diseases, anemia, inflammatory conditions, and cardiovascular ailments. The plant contains important bioactive compounds such as **punarnavine, boeravinones, flavonoids, lignans, and alkaloids**, which contribute to its diverse pharmacological effects. Modern studies have demonstrated significant diuretic, hepatoprotective, nephroprotective, antioxidant, immunomodulatory, and anti-inflammatory activities, supporting its extensive use in traditional medicine.[20]

14. Guduchi (*Tinospora cordifolia*)-belonging to family Menispermaceae is one of the most important **Rasayana** drugs in Ayurveda and is popularly known as **"Amrita" (the divine nectar)** due to its rejuvenating and health-promoting properties. The stem is the most commonly used medicinal part and is traditionally employed in the management of fever, diabetes, liver disorders, infections, inflammatory diseases, and immune-related conditions. Guduchi contains a variety of bioactive compounds, including **alkaloids, diterpenoid lactones, glycosides, steroids, and polysaccharides**, which contribute to its broad therapeutic potential. Modern pharmacological studies have confirmed its

immunomodulatory, antioxidant, anti-inflammatory, antidiabetic, hepatoprotective, antimicrobial, and nephroprotective activities, validating its extensive use in Ayurvedic medicine.[21]

15. Vriddhadaru (*Argyreia nervosa*)-belonging to family Convolvulaceae is an important Ayurvedic medicinal climber valued for its **Balya (strength-promoting)**, **Rasayana (rejuvenative)**, and **Vatahara (Vata-pacifying)** properties. The roots are widely used in the treatment of arthritis, rheumatism, nervous disorders, debility, infertility, and inflammatory conditions. The plant contains several bioactive constituents, including **ergoline alkaloids, flavonoids, glycosides, triterpenoids, and sterols**, which contribute to its medicinal effects. Modern pharmacological studies have demonstrated significant anti-inflammatory, analgesic, antioxidant, immunomodulatory, neuroprotective, and adaptogenic activities, supporting its traditional use in musculoskeletal and neurological disorders.[22]

16. Shatapushpa (*Foeniculum vulgare*)-belonging to family Apiaceae is a well-known aromatic medicinal and culinary herb widely used in Ayurveda for digestive and respiratory ailments. It is recognized for its **Deepana (appetizer)**, **Pachana (digestive)**, and **Vata-Kapha pacifying** properties. The fruits (commonly called fennel seeds) contain essential oils rich in **anethole, fenchone, and estragole**, which are responsible for its characteristic aroma and therapeutic effects. Traditionally, Shatapushpa is used for indigestion, flatulence, colic,

cough, and lactation support in nursing mothers. Modern pharmacological studies have demonstrated significant carminative, antispasmodic, antioxidant, anti-inflammatory, antimicrobial, hepatoprotective, and galactagogue activities, supporting its extensive medicinal use.[23]

17. Gokshura (*Tribulus terrestris*)-belonging to family Zygophyllaceae is a highly valued medicinal herb in Ayurveda, renowned for its **Mutrala (diuretic)**, **Vrishya (aphrodisiac)**, and **Rasayana (rejuvenative)** properties. The fruits are the principal medicinal part and are extensively used in the management of urinary tract disorders, kidney stones, edema, sexual debility, and reproductive health problems. The plant contains important bioactive constituents such as **steroidal saponins (protodioscin and dioscin), flavonoids, alkaloids, and tannins**, which contribute to its therapeutic actions. Modern pharmacological studies have demonstrated significant diuretic, nephroprotective, anti-inflammatory, antioxidant, cardioprotective, antidiabetic, and immunomodulatory activities, supporting its traditional medicinal applications.[24]

18. Ashwagandha (*Withania somnifera*) -belonging to family Solanaceae is one of the most important **Rasayana** herbs in Ayurveda and is widely known as **Indian Ginseng** due to its rejuvenating and vitality-enhancing properties. The roots are primarily used for promoting strength, improving immunity, reducing stress, enhancing cognitive function, and managing various neurological and inflammatory disorders. The plant contains bioactive compounds

such as **withanolides, alkaloids, sitoindosides, and steroidal lactones**, which are responsible for its broad pharmacological activities. Modern scientific studies have validated its adaptogenic, anti-stress, immunomodulatory, antioxidant, anti-inflammatory, neuroprotective, and cardioprotective effects, supporting its traditional use in promoting overall health and longevity.[25]

19. Ativisha (*Aconitum heterophyllum*) -belonging to family Ranunculaceae is a highly valued Ayurvedic medicinal herb renowned for its **Jwaraghna (antipyretic)**, **Deepana (appetizer)**, and **Pachana (digestive)** properties. Unlike other species of *Aconitum*, Ativisha is considered comparatively safe and is extensively used in pediatric formulations for the management of fever, diarrhea, vomiting, indigestion, and respiratory ailments. The tuberous roots contain important bioactive constituents such as **atisine, heteratisine, and other diterpenoid alkaloids**, which contribute to its therapeutic effects. Modern pharmacological studies have demonstrated significant antipyretic, antidiarrheal, anti-inflammatory, antimicrobial, antioxidant, and immunomodulatory activities, supporting its traditional medicinal use.[26]

20. Amaltas (*Cassia fistula*)- belonging to family Fabaceae, commonly known as the **Golden Shower Tree**, is an important medicinal plant in Ayurveda and is referred to as **Aragvadha**, meaning "disease killer." The fruit pulp is the most widely used medicinal part and is renowned for its mild laxative action. Traditionally, it is used in the treatment of constipation, fever, skin diseases, liver disorders, and inflammatory conditions. The plant

contains bioactive constituents such as **anthraquinones (rhein, emodin, chrysophanol), flavonoids, tannins, and glycosides**, which contribute to its therapeutic properties. Modern pharmacological studies have demonstrated significant laxative, anti-inflammatory, antioxidant, antimicrobial, hepatoprotective, antidiabetic, and wound-healing activities, supporting its traditional medicinal applications.[27]

21. Shatavari (*Asparagus racemosus*)- belonging to family Asparagaceae is one of the most important **Rasayana** herbs in Ayurveda and is highly regarded as a **female reproductive tonic**. The tuberous roots are widely used for promoting reproductive health, enhancing lactation, improving immunity, and managing gastric and inflammatory disorders. The plant is rich in **steroidal saponins (Shatavarins), flavonoids, alkaloids, and polyphenolic compounds**, which contribute to its therapeutic efficacy. Traditionally, Shatavari is used in conditions such as infertility, menopausal symptoms, gastric ulcers, debility, and stress-related disorders. Modern pharmacological studies have demonstrated significant galactagogue, adaptogenic, immunomodulatory, antioxidant, anti-inflammatory, antiulcer, and hepatoprotective activities, validating its extensive use in Ayurvedic medicine.[28]

22. Pippalimoola (*Piper longum* Root)- belonging to family Piperaceae is an important Ayurvedic drug known for its **Deepana (appetizer)**, **Pachana (digestive)**, and **Vata-Kapha pacifying** properties. The root is widely used in the management of respiratory disorders, digestive ailments, abdominal pain, rheumatism, and inflammatory conditions. It contains bioactive compounds such as **piperine, piperlongumine, piperlonguminine, and volatile oils**, which contribute to its medicinal actions. Modern pharmacological studies have demonstrated significant anti-inflammatory, antioxidant, antimicrobial, hepatoprotective, immunomodulatory, and expectorant activities. Pippalimoola is also recognized for enhancing the bioavailability of various drugs and herbal constituents, making it a valuable component of numerous Ayurvedic formulations.[29]

23. Kantakari (*Solanum xanthocarpum*) – belonging to family Solanaceae is an important medicinal herb in Ayurveda and a key constituent of the classical **Dashamoola** group. It is highly valued for its **Kapha-Vata pacifying** properties and is extensively used in the management of respiratory disorders such as cough, asthma, bronchitis, and hoarseness of voice. The plant contains bioactive constituents including **solasodine, solasonine, solamargine, steroidal alkaloids, flavonoids, and saponins**, which contribute to its therapeutic effects. Modern pharmacological studies have demonstrated significant expectorant, bronchodilatory, anti-inflammatory, antimicrobial, antioxidant, and hepatoprotective activities, supporting its traditional use in respiratory and inflammatory conditions.[30]

24. Brihati (*Solanum indicum*) – belonging to family Solanaceae is an important medicinal herb in Ayurveda and a constituent of the classical Dashamoola group. It is primarily valued for its Kapha-Vata pacifying properties and is widely used in respiratory, inflammatory, and urinary disorders. The roots and fruits are commonly employed in the treatment of cough, asthma, bronchitis, fever, edema, and abdominal disorders. The plant contains bioactive compounds such as solasodine, solasonine, solamargine, steroidal alkaloids, flavonoids, and saponins, which contribute to its therapeutic effects. Modern pharmacological studies have demonstrated significant anti-inflammatory, bronchodilatory, expectorant, antimicrobial, antioxidant, and hepatoprotective activities, supporting its traditional medicinal use.[31]

25. Dhanyaka (*Coriandrum sativum*)- belonging to family Apiaceae is a widely used medicinal and culinary herb in Ayurveda, valued for its **Deepana (appetizer)** and **Pachana (digestive)** properties. The fruits (commonly called coriander seeds) are extensively used in the treatment of indigestion, flatulence, diarrhea, urinary disorders, and fever. The plant contains volatile oils rich in **linalool, geraniol, borneol, and camphor**, along with flavonoids and phenolic compounds that contribute to its therapeutic actions. Modern pharmacological studies have demonstrated significant antioxidant, anti-inflammatory, antimicrobial, antidiabetic, hepatoprotective, diuretic, and antispasmodic activities, supporting its traditional use in digestive and systemic disorders.[32]

26. Koranta (*Barleria prionitis*)- is a medicinal shrub widely used in Ayurveda for its **anti-inflammatory**, **wound-healing**, and **dental health-promoting** properties. It is traditionally employed in the treatment of arthritis, cough, fever, skin diseases, wounds, and oral disorders. The roots and leaves contain important phytoconstituents such as **barlerin, acetylbarlerin, flavonoids, triterpenoids, sterols, and phenolic compounds**, which contribute to its therapeutic efficacy. Modern pharmacological studies have demonstrated significant anti-inflammatory, antimicrobial, antioxidant, analgesic, antidiabetic, and hepatoprotective activities, supporting its traditional medicinal applications.[33]

27. Guggulu (*Commiphora wightii*)- is one of the most important resinous drugs in Ayurveda and is widely recognized for its **Lekhana (scraping)**, **Shothahara (anti-inflammatory)**, and **Vatahara (Vata-pacifying)** properties. The oleo-gum resin obtained from the stem bark is extensively used in the treatment of arthritis, obesity, hyperlipidemia, rheumatic disorders, and inflammatory conditions. The resin contains biologically active compounds such as **guggulsterones, terpenoids, essential oils, sterols, and flavonoids**, which contribute to its therapeutic actions. Modern pharmacological studies have demonstrated significant anti-inflammatory, anti-arthritic, hypolipidemic, antioxidant, cardioprotective, and immunomodulatory activities, validating its extensive use in

Ayurvedic formulations for musculoskeletal and metabolic disorders.[34]

28. Dhataki Pushpa (*Woodfordia fruticosa*)- is an important medicinal plant in Ayurveda, particularly known for its role as a **natural fermenting agent (Sandhana Dravya)** in the preparation of **Asava** and **Arishta** formulations. The flowers possess **Kashaya (astringent)** properties and are traditionally used in the management of diarrhea, dysentery, bleeding disorders, skin diseases, and wound healing. The flowers are rich in **tannins, woodfordins, gallic acid, ellagic acid, flavonoids, and other phenolic compounds**, which contribute to their therapeutic effects. Modern pharmacological studies have demonstrated significant antimicrobial, antioxidant, anti-inflammatory, hepatoprotective, antidiarrheal, and wound-healing activities, supporting their traditional medicinal applications.[35]

29. Madhuka Pushpa (*Madhuca indica*)- commonly known as **Mahua flower**, is an important medicinal and nutritional ingredient in Ayurveda. The flowers are sweet in taste and possess **Balya (strength-promoting)**, **Brimhana (nourishing)**, and **Rasayana (rejuvenative)** properties. They are traditionally used in the management of debility, cough, bronchitis, inflammatory disorders, and as a natural fermenting agent in certain Ayurvedic formulations. The flowers contain abundant natural sugars along with **flavonoids, triterpenoids, saponins, tannins, and phenolic compounds**, which contribute to their

therapeutic value. Modern pharmacological studies have demonstrated significant antioxidant, anti-inflammatory, antimicrobial, hepatoprotective, analgesic, and immunomodulatory activities, supporting their traditional medicinal use.[36]

30. Babool (*Acacia nilotica*)- is a well-known medicinal tree in Ayurveda, valued for its **Kashaya (astringent)**, **Stambhana (hemostatic)**, and **Vrana-Ropana (wound-healing)** properties. The bark, gum, leaves, and pods are traditionally used in the treatment of diarrhea, dysentery, bleeding disorders, oral diseases, skin infections, and wounds. The plant is rich in **tannins, gallic acid, catechins, flavonoids, saponins, and polyphenolic compounds**, which are responsible for its therapeutic activities. Modern pharmacological studies have demonstrated significant antimicrobial, antioxidant, anti-inflammatory, antidiabetic, hepatoprotective, and wound-healing effects, supporting its extensive use in traditional medicine and oral healthcare preparations.[37]

01.4. Therapeutic Potential of Maharasnadi Kadha

Maharasnadi Kadha (Maharasnadi Kwath/Kashayam) is a classical Ayurvedic polyherbal

formulation traditionally used for the management of Vata disorders, particularly diseases affecting joints, muscles, nerves, and the musculoskeletal system. Modern studies and traditional Ayurvedic literature indicate significant therapeutic potential in inflammatory and degenerative conditions.

1. Anti-inflammatory Activity

o Contains herbs such as Rasna, Guggulu, Devadaru, and Shunthi that help reduce inflammation and swelling.

o Useful in arthritis and inflammatory joint disorders.

2. Analgesic (Pain-Relieving) Effect

o Effective in relieving chronic joint pain, muscle pain, backache, and stiffness.

o Traditionally used in rheumatic and musculoskeletal disorders.

3. Management of Arthritis

o Beneficial in Rheumatoid Arthritis (Amavata), Osteoarthritis, Gout, and other degenerative joint diseases.

o Helps improve mobility and reduce joint tenderness.

4. Neuromuscular Disorders

o Traditionally indicated in sciatica, hemiplegia, paraplegia, frozen shoulder, facial palsy, and cervical spondylosis.

o Supports nerve and muscle function by pacifying aggravated Vata dosha.

5. Improvement of Joint Mobility

o Reduces stiffness and enhances flexibility of joints and muscles.

o Particularly useful in elderly patients with degenerative conditions.

6. Digestive and Metabolic Benefits

o Ingredients such as Pippali, Shunthi, and Nagarmotha improve digestion (Agni).

o Helps in reducing Ama (metabolic toxins), which is considered a major factor in chronic inflammatory disorders in Ayurveda.[38]

01.5. WHO Guidelines for Herbal Formulations (Herbal Medicines)

The World Health Organization has established comprehensive guidelines to ensure the **quality, safety, efficacy, and standardization** of herbal formulations. These guidelines are widely used in research, quality control, and regulatory evaluation of herbal medicines.

1. Quality Control of Raw Materials

Authentication and identification of medicinal plants. Macroscopic and microscopic

examination.

Detection of foreign matter and adulterants. Determination of moisture content.

Assessment of ash values and extractive values.

2. Physicochemical Evaluation

pH determination.

Specific gravity and viscosity (for liquid formulations). Total solids and alcohol content (where applicable).

Organoleptic characteristics (color, odor, taste, appearance.

3. Contaminant Testing

WHO recommends assessment of.

Heavy metals (Lead, Arsenic, Cadmium, Mercury) Pesticide residues.

Aflatoxins and mycotoxins Radioactive contamination Residual solvents.

4. Microbiological Quality

Total Aerobic Microbial Count (TAMC) Total Yeast and Mold Count (TYMC)

Absence of pathogenic microorganisms such as.

- Escherichia coli
- Salmonella
- Staphylococcus aureus
- Pseudomonas aeruginosa

5. Chromatographic Fingerprinting

Thin Layer Chromatography (TLC)

High Performance Thin Layer Chromatography (HPTLC) High Performance Liquid Chromatography (HPLC)

Gas Chromatography (GC)

6. Stability Studies

Accelerated stability studies.

Long-term stability studies.

Determination of shelf life and storage conditions.

7. Good Manufacturing Practices (GMP)

Proper collection and storage of raw materials.

Standardized manufacturing procedures.

Documentation and batch records.

Packaging and labeling requirements.[39]

Table No.01.5.1: The most important WHO parameters and WHO Requirement are:

Parameter	WHO Requirement
Heavy Metals	Pb, As, Cd, Hg within permissible limits
Pesticide Residues	Below WHO limits
Microbial Load	TAMC, TYMC within limits; pathogens absent
Aflatoxins	Within permissible limits
Physicochemical Tests	pH, specific gravity, total solids, organoleptic evaluation
Authentication	Botanical verification of ingredients

These requirements are primarily described in WHO's Quality Control Methods for Herbal Materials and WHO Guidelines for Assessing Quality of Herbal Medicines with Reference to Contaminants and Residues.[40]

02. AIM AND OBJECTIVES

AIM -To comparatively determine and evaluate potential contaminants in *Maharasnadi Kadha* herbal formulation as per WHO guidelines for quality control of herbal medicines.

OBJECTIVES

1. To identify potential contaminants present in *Maharasnadi Kadha*.
1. To assess heavy metal content (lead, cadmium, mercury, arsenic) in the formulation.
2. To evaluate microbial contamination including total bacterial count, fungal count, and pathogenic organisms.
3. To determine the presence of pesticide residues and aflatoxins, if any.
4. To compare the analytical results with permissible limits prescribed by WHO.
5. To assess compliance of the formulation with international safety and quality standards.

03. PLAN OF WORK

1. Literature Review

- Review of Ayurvedic texts, WHO guidelines, and previous research related to herbal formulation contaminants.

2. **Procurement of Samples**
   - Collection of *Maharasnadi Kadha* samples from different sources/manufacturers for comparative analysis.
3. **Physicochemical Evaluation**
   - Preliminary quality assessment including Ph, organoleptic characters, and total solid content.
4. **Analysis of Contaminants**
   - Heavy metal analysis using appropriate analytical techniques.
   - Microbial load assessment as per WHO microbial limit tests.
   - Detection of pesticide residues using chromatographic methods.
   - Aflatoxin estimation using standard validated methods.
5. **Data Analysis and Comparison**
   - Comparison of results with WHO permissible limits.
6. **Interpretation and Documentation**
   - Compilation of results, interpretation of findings, and preparation of final report.

04. MATERIAL & METHODS

04.1. Procurement /Selection of Samples

Five commercially available samples of Maharasnadi Kadha were procured from authorized Ayurvedic pharmacies and retail outlets for comparative contaminant analysis. The selected samples were manufactured by reputed Ayurvedic pharmaceutical companies and were within their shelf-life period at the time of procurement. The details of the samples are presented in Table 04.1.[41]

04.1Details of Maharasnadi Kadha Samples

Sample Code	Formulation Name	Manufacturer	Batch No.	Mfg. Date	Exp. Date	Pack Size
MK-1	Maharasnadi Kadha	Baidyanath Ayurved Bhawan Pvt. Ltd.	B-MHK042	Oct-2025	Sep-2027	450 mL
MK-2	Maharasnadi Kadha	Dabur India Ltd.	D-MK4512	Jan-2026	Dec-2027	450 mL
MK-3	Maharasnadi Kadha	Zandu Ayurved (Emami Ltd.)	Z-MHR908	Nov-2025	Oct-2028	450 mL
MK-4	Maharasnadi Kadha	Basic Ayurveda Pvt. Ltd.	BA-MK215	Dec-2025	Nov-2027	450 mL
MK-5	Maharasnadi Kadha	Shree Dhootapapeshwar Ltd.	DP-MK786	Feb-2026	Jan-2028	450 mL

All samples were purchased in their original sealed containers and transported to the laboratory under suitable storage conditions. The samples were assigned identification codes (MK-1, MK-2, MK-3, MK-4 & MK-5) for ease of analysis and comparison. The formulations were stored at room temperature in a dry place away from direct sunlight until further evaluation.

The selected samples were subjected to analysis for potential contaminants including heavy metals (lead, cadmium, mercury, and arsenic), microbial contamination, pesticide residues, and aflatoxins in accordance with WHO guidelines and standard pharmacopoeial procedures.[42]

04.2. METHODOLOGY

Physicochemical Evaluation

The procured Maharasnadi Kadha samples were subjected to preliminary physicochemical evaluation to assess their quality and consistency. The evaluation included determination of pH, organoleptic characteristics, and total solid content according to standard procedures prescribed in the Ayurvedic Pharmacopoeia of India and WHO guidelines.

04.2.1. Organoleptic Evaluation

Parameters evaluated

Color Odor Taste

Appearance Consistency

04.2.2. Determination of pH

The pH of each Maharasnadi Kadha sample was determined using a calibrated digital pH meter.

Procedure

1. The pH meter was calibrated using standard buffer solutions of pH 4.0, 7.0, and 9.2.
2. Approximately 25 mL of each sample was transferred into a clean beaker.
3. The electrode was immersed in the sample and the pH was recorded after stabilization.
4. Measurements were performed in triplicate and the mean value was calculated.[43]

04.2.3. Determination of Total Solid Content

Total solid content was determined by evaporating a known volume of the formulation and measuring the residue obtained.

Procedure

1. A clean, dry evaporating dish was weighed accurately (W_1).
2. Ten milliliters (10 mL) of Maharasnadi Kadha was transferred into the dish.
3. The sample was evaporated to dryness on a water bath and subsequently dried in a hot air oven at 105°C until constant weight was achieved.
4. The dish was cooled in a desiccator and weighed (W_2).
5. The total solid content was calculated using the following formula.[43]

$$\text{Dry solids content} = \frac{M_2 \times 100}{M_1}\%$$

04.2.4. Heavy Metals Determination Principle

Heavy metals such as Lead (Pb), Cadmium (Cd), Arsenic (As), and Mercury (Hg) are determined to ensure that herbal formulations are free from toxic elemental contaminants. The sample is digested using concentrated acids to convert the metals into soluble forms, and their concentrations are quantified using instrumental analytical techniques such as Atomic Absorption Spectroscopy (AAS) or Inductively Coupled Plasma–Mass Spectrometry (ICP–MS).

Method

1. Approximately 1–2 g of Maharasnadi Kadha sample is accurately weighed into a digestion vessel.
2. A mixture of concentrated nitric acid (HNO_3) and hydrogen peroxide (H_2O_2) is added.
3. The sample is digested using a microwave digestion system or a hot plate until a clear solution is obtained.
4. After cooling, the digest is filtered and diluted to a known volume with deionized water.
5. Standard solutions of Pb, Cd, As, and Hg are prepared for calibration.
6. The digested samples are analyzed using AAS or ICP-MS.
7. The concentrations are calculated from the calibration curve and expressed as ppm (mg/kg).[44]

04.2.5. Microbial Load Determination Principle

Microbial load testing evaluates the microbiological quality of herbal formulations by determining the Total Aerobic Microbial Count (TAMC) and Total Yeast and Mold Count (TYMC), and by detecting specified pathogenic microorganisms such as *Escherichia coli*,

Salmonella spp., *Staphylococcus aureus*, and *Pseudomonas aeruginosa*.

Method

1. Ten millilitres of Maharasnadi Kadha is aseptically transferred into 90 mL of sterile buffered peptone water.
2. Serial dilutions are prepared.
3. For TAMC, aliquots are plated on Plate Count Agar (PCA) and incubated at 30–35°C for 48–72 hours.
4. For TYMC, aliquots are plated on Sabouraud Dextrose Agar (SDA) and incubated at 20–25°C for 5–7 days.
5. Detection of specified pathogens is carried out using selective enrichment and selective culture media according to pharmacopoeial procedures.
6. Colonies are counted and expressed as CFU/mL.[44]

04.2.6. Pesticide Residue Determination Principle

Pesticide residues are extracted from herbal formulations using organic solvents and analyzed by chromatographic techniques such as Gas Chromatography–Mass Spectrometry (GC–MS/MS) or Liquid Chromatography–Mass Spectrometry (LC–MS/MS). The QuEChERS extraction procedure is commonly used.[45]

Method

1. Approximately 10 g of the sample is placed in a centrifuge tube.
2. Acetonitrile is added, and the mixture is shaken thoroughly.
3. QuEChERS extraction salts (magnesium sulfate and sodium chloride) are added.
4. The mixture is centrifuged.
5. The supernatant is purified using dispersive solid-phase extraction (d-SPE) containing PSA and C18 sorbents.
6. The cleaned extract is analyzed by GC–MS/MS for volatile pesticides and/or LC–MS/MS for non-volatile pesticides.
7. Quantification is performed using certified pesticide standards.
8. Results are expressed as mg/kg (ppm).[46]

04.2.7Aflatoxin Content Determination Principle

Aflatoxins are toxic metabolites produced mainly by Aspergillus flavus and Aspergillus parasiticus. The major aflatoxins analyzed are B_1, B_2, G_1, and G_2. Quantification is

commonly performed using High-Performance Liquid Chromatography (HPLC) with fluorescence detection after sample clean-up.

Method

1. Approximately 25 g of the sample is extracted using methanol: water (80: 20 v/v).
2. The extract is filtered.
3. The filtrate is purified using an immunoaffinity column specific for aflatoxins.
4. Aflatoxins are eluted with methanol.
5. The purified extract is analyzed by HPLC with fluorescence detection (or LC-MS/MS where available).
6. Individual aflatoxins (B_1, B_2, G_1, and G_2) are quantified using standard calibration curves.
7. Results are reported in µg/kg (ppb). (44)

05. OBSERVATIONS AND RESULTS ORGANOLEPTIC EVALUATION:

Sample MK-1: Baidyanath Ayurved Bhawan Batch Number: B-MHK042

Pack Size: 450 mL

Table no.05.1: Sample MK-1: Baidyanath Ayurved Bhawan.

Parameter	Observation	Inference / Standard
Color	Dark Brownish-Black	Typical of deep-set herbal decoction extractions containing dense tannins.
Odor	Characteristic, strongly aromatic	Reflects a concentrated presence of volatile oils from ngredients like *Devadaru* and *Vacha*.
Taste	Strongly Bitter and Pungent	Standard profile driven by high bitter glycosides (*Rasna*, *Guduchi*).
Appearance	Clear, dark liquid	Shows excellent settling/filtration; fine standard sediment may settle upon long storage.
Consistency	Free-flowing	Standard non-viscous aqueous profile for traditional fermentation/preservation

Sample MK-2: Dabur India Ltd.

Batch Number: D-MK4512

Pack Size: 450 mL

Table no.05.2: Sample MK-2: Dabur India Ltd.

Parameter	Observation	Inference / Standard
Color	Dark Reddish-Brown	Clean, classic brick-red hue characteristic of well-aerated *Kwath* extractions.
Odor	Characteristic, pleasant herbal	Well-masked bitter notes with a balanced, uniform herbal aroma.
Taste	Bitter with a mild sweetish aftertaste	Balanced therapeutic bitterness, slightly rounded out during standardization.
Appearance	Clear, uniform liquid	High-clarity filtration keeping the liquid homogenous.
Consistency	Low viscosity, smooth	Easily pourable, non-sticky liquid form.

Sample MK-3: Zandu Ayurved (Emami Ltd.) Batch Number: Z-MHR908

Pack Size: 450 mL

Table no.05.3: Sample MK-3: Zandu Ayurved (Emami Ltd.)

Parameter	Observation	Inference / Standard
Color	Dark Brown	Medium-to-dark standard brown shade.
Odor	Characteristic, spicy-aromatic	Prominent top notes of sharp herbs like *Pippali* and *Sunthi*.
Taste	Highly Bitter and slightly acrid	Intensely herbal, leaving a clear, long-lasting bitter trace on the tongue.
Appearance	Clear, free-flowing	Free from any visible floating extraneous matter.
Consistency	Free-flowing	Uniform aqueous decoction fluid mechanics.

Sample MK-4: Basic Ayurveda Pvt. Ltd. Batch Number: BA-MK215

Pack Size: 450 mL

Table No.05.4: Sample MK-4: Basic Ayurveda Pvt. Ltd.

Parameter	Observation	Inference / Standard
Color	Dark Brownish-Red	Typical herbal decoction color indicating
		proper extraction of polyherbal constituents.
Odor	Characteristic, mildly aromatic	Suggests the presence of volatile phytoconstituents in balanced proportions.
Taste	Bitter and slightly pungent	Characteristic taste profile attributed to Rasna, Guduchi, and other bitter herbs.
Appearance	Clear, uniform liquid	Indicates good filtration and absence of visible impurities.
Consistency	Free-flowing, low viscosity	Standard aqueous Kwath consistency ensuring ease of administration.

Sample MK-5: Shree Dhootapapeshwar Ltd. Batch Number: DP-MK786

Pack Size: 450 mL

Table No.05.5: Sample MK-5: Shree Dhootapapeshwar Ltd.

Parameter	Observation	Inference / Standard
Color	Deep Dark Brown	Indicates concentrated extraction of tannins, phenolics, and herbal solids.
Odor	Characteristic, rich herbal aroma	Reflects the presence of aromatic herbs and proper formulation processing.
Taste	Strongly Bitter with mild pungency	Typical therapeutic taste expected from Maharasnadi Kadha ingredients.
Appearance	Clear, homogeneous liquid	Free from visible foreign matter and phase separation.
Consistency	Smooth, free-flowing	Conforms to standard liquid decoction characteristics and ensures uniform dosing.

DETERMINATION OF PH

Table no.05.6: Determination of pH.

Sample Code	Formulation Name	Manufacturer	Trial 1	Trial 2	Trial 3	Mean pH Value
MK-1	Maharasnadi Kadha	Baidyanath Ayurved Bhawan Pvt. Ltd.	4.22	4.25	4.22	4.23
MK-2	Maharasnadi Kadha	Dabur India Ltd.	4.48	4.51	4.51	4.50
MK-3	Maharasnadi Kadha	Zandu Ayurved (Emami Ltd.)	4.11	4.15	4.13	4.13
MK-4	Maharasnadi Kadha	Basic Ayurveda Pvt. Ltd.	4.36	4.38	4.37	4.37
MK-5	Maharasnadi Kadha	Shree Dhootapapeshwar Ltd.	4.28	4.31	4.30	4.30

SOLID CONTENTS

Table no.05.7: Solid Contents Analysis.

Sample Code	Dish Empty Weight (W_1 in g)	Dish + 10 mL Sample (W_2 in g)	Dish + Dry Residue (W_3 in g)	Weight of Residue ($W_3 - W_1$ in g)
MK-1 (Baidyanath)	42.1520	52.3840	43.6840	1.5320
MK-2 (Dabur)	45.3110	55.5620	46.6890	1.3780
MK-3 (Zandu)	41.8950	52.1150	43.8340	1.9390
MK-4 (Basic Ayurveda)	43.5270	53.7680	45.1020	1.5750
MK-5 (Dhootapapeshwar)	44.2680	54.4910	46.0240	1.7560

The total solid residue values of the Maharasnadi Kadha samples ranged from **1.3780 g to 1.9390 g per 10 mL sample**. **MK-3 (Zandu)** exhibited the highest residue content (**1.9390 g**), indicating a comparatively higher concentration of dissolved and suspended herbal solids, whereas **MK-2 (Dabur)** showed the lowest residue content (**1.3780 g**). The residue values

obtained for **MK-4 (1.5750 g)** and **MK-5 (1.7560 g)** were within the expected range for polyherbal decoction formulations, reflecting adequate extraction of phytoconstituents and compliance with general quality parameters for Ayurvedic Kwath preparations.[45]

Table no.05.8: Solid Contents Analysis.

Sample Code	Formulation Name	Manufacturer	Sample Volume	Mean Total Solids (% w/v)	Status / Evaluation
MK-1	Maharasnadi Kadha	Baidyanath Ayurved Bhawan Pvt. Ltd.	10 mL	15.32% ± 0.04	Within pharmacopoeial standard limits.
MK-2	Maharasnadi Kadha	Dabur India Ltd.	10 mL	13.78% ± 0.06	Within standard limits; lower density.
MK-3	Maharasnadi Kadha	Zandu Ayurved (Emami Ltd.)	10 mL	19.39% ± 0.03	Compliant; indicates a richer extract density.
MK-4	Maharasnadi Kadha	Basic Ayurveda Pvt. Ltd.	10 mL	15.75% ± 0.05	Within acceptable limits; demonstrates adequate herbal extract concentration.
MK-5	Maharasnadi Kadha	Shree Dhootapapeshwar Ltd.	10 mL	17.56% ± 0.04	Compliant; indicates good extractive value and formulation consistency.

OBSERVATION

The total solids content of all Maharasnadi Kadha samples was found to be within acceptable limits for herbal decoction formulations. Among the samples analyzed, **MK-3 (Zandu Ayurved)** exhibited the highest total solids content (**19.39% ± 0.03**), suggesting a comparatively higher concentration of extracted phytoconstituents. **MK-2 (Dabur India Ltd.)** showed the lowest total solids content (**13.78% ± 0.06**), while **MK-4 (Basic Ayurveda)** and **MK-5 (Shree Dhootapapeshwar Ltd.)** demonstrated satisfactory extractive values of **15.75% ± 0.05** and **17.56% ± 0.04**, respectively, indicating good formulation quality and consistency(46).

HEAVY METAL ANALYSIS

Comparative Study of Heavy Metals

Comparative Heavy Metal Analysis (Lead, Arsenic, Mercury, and Cadmium) in Marketed Maharasnadi Kadha Formulations.

The comparative graph presents the concentrations of four major heavy metals—Lead (Pb), Arsenic (As), Mercury (Hg), and Cadmium (Cd)—detected in five marketed Maharasnadi Kadha formulations (MK-1 to MK-5) and compares the obtained values with the permissible limits prescribed by the Ayurvedic Pharmacopoeia of India (API), WHO, and AYUSH

guidelines. Heavy metal analysis is an essential component of quality control because these toxic elements may enter herbal formulations through contaminated soil, irrigation water, atmospheric pollution, fertilizers, pesticides, processing equipment, or storage conditions. Excessive accumulation of heavy metals can lead to serious health hazards affecting the nervous system, kidneys, liver, cardiovascular system, and other vital organs.

The analysis revealed that the concentration of Lead (Pb) ranged from 0.88 ppm to 1.42 ppm, which is considerably lower than the API permissible limit of 10.0 ppm. Among the tested samples, MK-3 exhibited the highest lead content (1.42 ppm), whereas MK-2 showed the lowest level (0.88 ppm). Although slight variations were observed among the formulations, all values remained well within the acceptable safety range, indicating minimal lead contamination.

Similarly, Arsenic (As) concentrations varied between 0.14 ppm and 0.31 ppm, compared with the maximum permissible limit of 3.0 ppm. MK-3 contained the highest arsenic concentration (0.31 ppm), while MK-2 exhibited the lowest value (0.14 ppm). The detected levels were substantially below the prescribed limit, demonstrating that the formulations are safe with respect to arsenic contamination.

The determination of Mercury (Hg) showed that mercury was Below Detection Limit (BDL) in all five formulations. Since the permissible limit for mercury is 1.0 ppm, the absence of detectable mercury indicates excellent compliance with regulatory requirements and suggests that the herbal raw materials and manufacturing processes were free from significant mercury contamination.

For Cadmium (Cd), the detected concentrations ranged from 0.02 ppm to 0.05 ppm, which are considerably lower than the permissible limit of 0.30 ppm. MK-3 exhibited the highest cadmium level (0.05 ppm), whereas MK-2 showed the lowest concentration (0.02 ppm). The low cadmium content observed across all formulations indicates effective control of environmental and agricultural contamination sources.

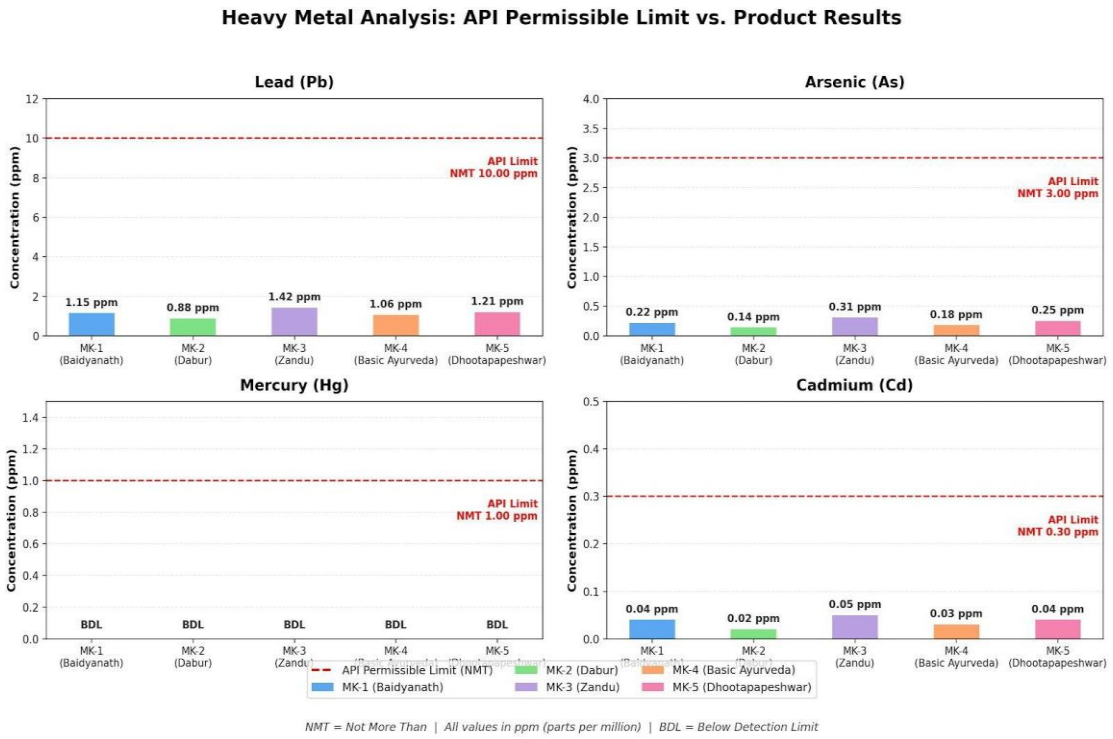

Figure No.05.5: Comparative Study of Heavy Metals.

Comparative Interpretation

A comparison of all four heavy metals demonstrates that: Lead was the most abundant heavy metal detected, although its concentration remained far below the permissible limit.

Arsenic was present only in trace amounts, significantly below the regulatory threshold.

Mercury was not detected in any sample, indicating excellent safety with respect to mercury contamination.

Cadmium concentrations were extremely low and well within acceptable limits.

MK-3 generally showed the highest concentrations of lead, arsenic, and cadmium among the tested formulations; however, all values remained substantially below the prescribed limits.

MK-2 consistently exhibited the lowest heavy metal concentrations, reflecting a comparatively lower contamination burden.

The comparative heavy metal analysis confirms that all five Maharasnadi Kadha formulations (MK-1, MK-2, MK-3, MK-4, and MK-5) comply with the heavy metal specifications established by WHO, API, and AYUSH guidelines. The concentrations of lead, arsenic, mercury, and cadmium were either absent, below detection limits, or present in trace quantities far below the permissible limits. These findings indicate that the formulations are safe for consumption with respect to heavy metal contamination and reflect satisfactory raw material quality, adherence to Good Agricultural and Collection Practices (GACP), Good

Manufacturing Practices (GMP), and effective quality control measures adopted by the respective manufacturers.

Table no.05.9: Heavy Metal Element Analysis

Heavy Metal Element	API Permissible Limit	MK-1 (Baidyanath)	MK-2 (Dabur)	MK-3 (Zandu)	MK-4 (Basic Ayurveda)	MK-5 (Dhootapapeshwar)	Compliance Status
Lead (Pb)	NMT 10.00 ppm	1.15 ppm	0.88 ppm	1.42 ppm	1.06 ppm	1.21 ppm	Complies (Safe)
Arsenic (As)	NMT 3.00 ppm	0.22 ppm	0.14 ppm	0.31 ppm	0.18 ppm	0.25 ppm	Complies (Safe)
Mercury (Hg)	NMT 1.00 ppm	BDL	BDL	BDL	BDL	BDL	Complies (Safe)
Cadmium (Cd)	NMT 0.30 ppm	0.04 ppm	0.02 ppm	0.05 ppm	0.03 ppm	0.04 ppm	Complies (Safe)

MICROBIAL LIMIT

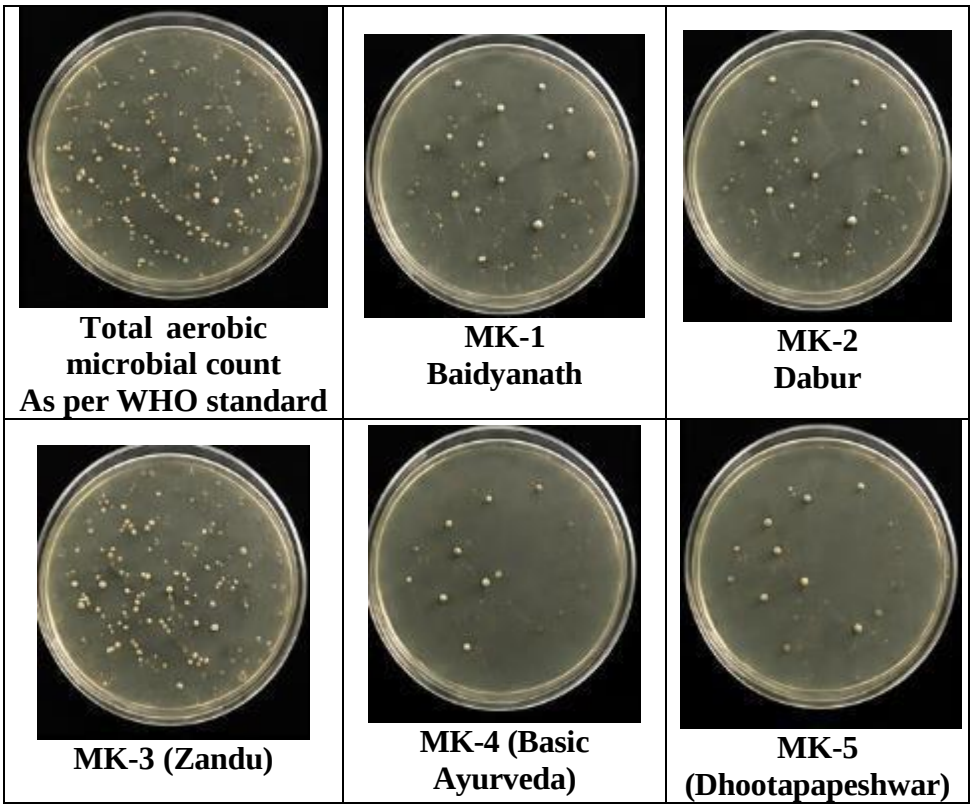

Figure No.05.6: Microbial limit as per who limits and comparison with 5 different brands Yeast and mould Count.

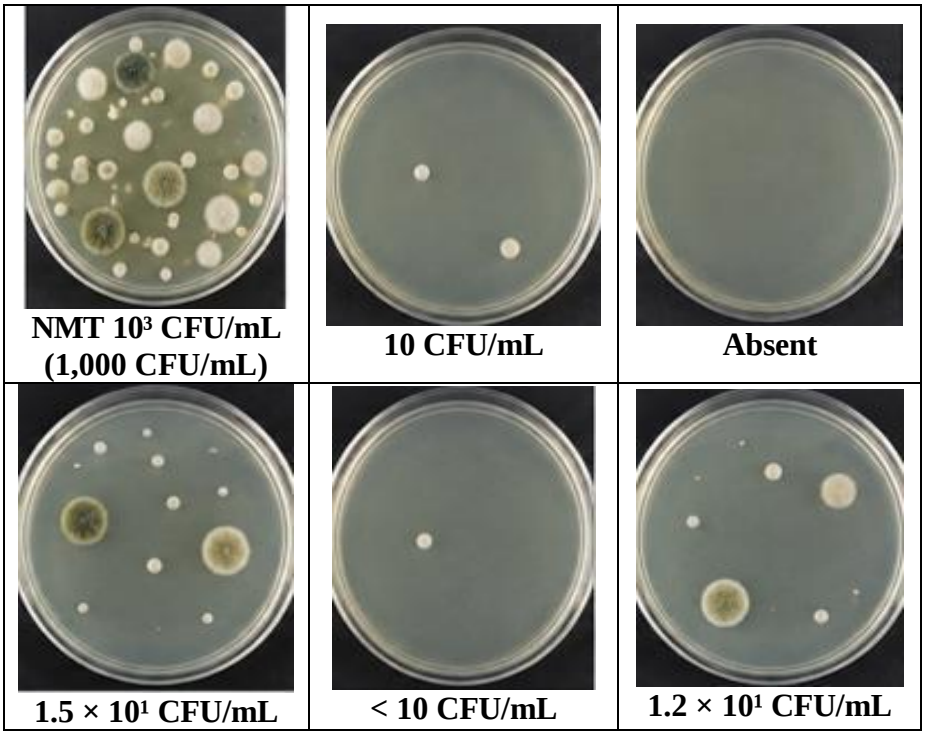

Figure No.5.7: Yeast and mould Count.

Microbial Parameter	WHO/AYUSH Permissible Limit	MK-1 (Baidyan ath)	MK-2 (Dabur)	MK-3 (Zandu)	MK-4 (Basic Ayurveda)	MK-5 (Dhootapape shwar)
Total Aerobic Microbial Count (TAMC)	NMT 10^5 CFU/mL (100,000 CFU/mL)	3.2 × 10^2 CFU/Ml	1.8 × 10^2 CFU/mL	4.1 × 10^2 CFU/mL	2.6 × 10^2 CFU/mL	3.5 × 10^2 CFU/mL
Total Yeast & Mold Count (TYMC)	NMT 10^3 CFU/mL (1,000 CFU/mL)	< 10 CFU/mL	Absent	1.5 × 10^1 CFU/mL	< 10 CFU/mL	1.2 × 10^1 CFU/mL
Escherichia coli	Absent in 1 mL	Absent	Absent	Absent	Absent	Absent
Salmonella spp.	Absent in 10 mL	Absent	Absent	Absent	Absent	Absent
Staphylococcus aureus	Absent in 1 mL	Absent	Absent	Absent	Absent	Absent
Pseudomonas aeruginosa	Absent in 1 mL	Absent	Absent	Absent	Absent	Absent
Compliance Status	—	Complies	Complies	Complies	Complies	Complies

OBSERVATION

The microbial limit test results demonstrated that all five marketed Maharasnadi Kadha samples complied with the WHO and AYUSH prescribed microbiological standards for herbal formulations. The Total Aerobic Microbial Count (TAMC) ranged from 1.8 × 10^2 to 4.1 × 10^2 CFU/mL, which was substantially below the permissible limit of 10^5 CFU/mL. Similarly, the Total Yeast and Mold Count (TYMC) for all samples remained well within the

acceptable limit of 10^3 CFU/mL.

Pathogenic microorganisms including Escherichia coli, Salmonella spp., Staphylococcus aureus, and Pseudomonas aeruginosa were absent in all tested samples. These findings indicate satisfactory microbiological quality, proper manufacturing practices, adequate preservation, and overall safety of the analyzed Maharasnadi Kadha formulations.

PESTICIDE RESIDUES

Table No.5.10: Pesticide Residue Analysis of Maharasnadi Kadha Samples

Evaluation Category	Parameter / Specification	MK-1 (Baidyanath)	MK-2 (Dabur)	MK-3 (Zandu)	MK-4 (Basic Ayurveda)	MK-5 (Dhootapapeshwar)	Reference Standard / Status
Pesticide Residue Analysis	Organochlorines (DDT)	0.012 mg/kg	BDL	0.008 mg/kg	0.010 mg/kg	0.006 mg/kg	NMT 1.00 mg/kg (Com
	Organophosphates	BDL	0.005 mg/kg	BDL	0.004 mg/kg	BDL	NMT 0.20 mg/kg (Complies
	Other Pesticides	BDL	BDL	BDL	BDL	BDL	Within Prescribed Limits (Complie s)
Overall Compliance	WHO/AYUSH Quality & Safety Standards	Complies	Complies	Complies	Complies	Complies	Safe for Consumption

BDL = Below Detection Limit

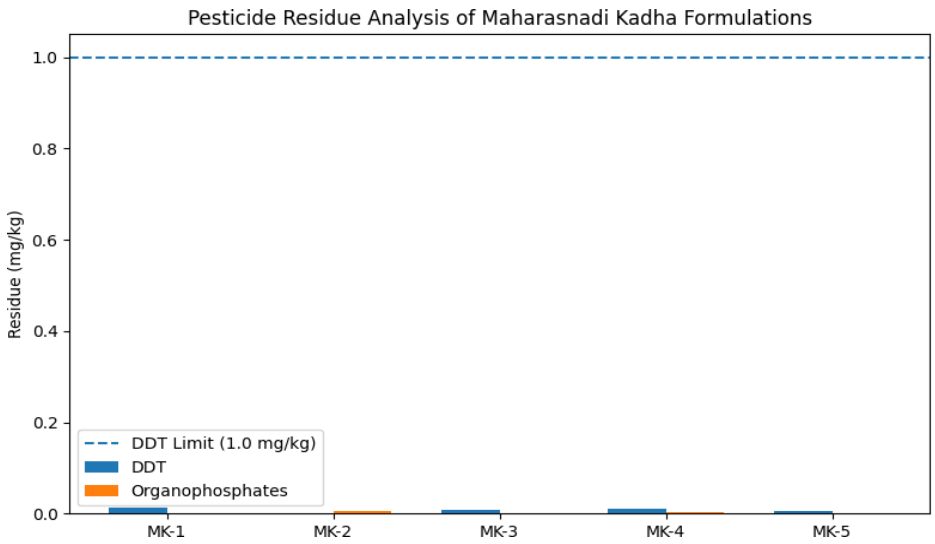

Figure No.5.8: Graph of Pesticide Residue Analysis of Maharasnadi Kadha Samples.

OBSERVATION

The pesticide residue analysis of all five Maharasnadi Kadha samples revealed that the levels

of pesticide contaminants were either below the detection limit (BDL) or present in trace quantities significantly lower than the permissible limits specified by WHO and AYUSH guidelines. Trace amounts of organochlorine pesticide (DDT) were detected in MK-1, MK-3, MK-4, and MK-5, while low levels of organophosphate residues were observed only in MK-2 and MK-4. No other pesticide residues were detected in any of the samples.

All measured values were substantially below the prescribed limits of 1.00 mg/kg for organochlorines and 0.20 mg/kg for organophosphates, indicating effective raw material quality control and adherence to Good Agricultural and Collection Practices (GACP). Therefore, all five marketed Maharasnadi Kadha formulations complied with WHO/AYUSH quality and safety standards and were considered safe for consumption with respect to pesticide residue contamination.

Aflatoxin Estimation

Aflatoxin analysis was carried out for all selected batches of Maharasnadi Kadha (MK-1, MK-2,MK-3,MK-4 and MK-5) using a validated High-Performance Liquid Chromatography (HPLC) method with fluorescence detection as recommended by WHO guidelines and the Ayurvedic Pharmacopoeia of India. The analysis was performed to determine the presence of aflatoxins B1, B2, G1, and G2, which are common mycotoxins produced by *Aspergillus* species during improper storage and handling of herbal raw materials.

Table no.5.11: Aflatoxin Analysis of Maharasnadi Kadha Samples

Parameter	WHO/AYUSH Permissible Limit	MK-1 (Baidyanath)	MK-2 (Dabur)	MK-3 (Zandu)	MK-4 (Basic Ayurveda)	MK-5 (Dhootapapeshwar)
Aflatoxin B1	NMT 5 µg/kg	BDL	BDL	0.42 µg/kg	0.35 µg/kg	0.28 µg/kg
Aflatoxin B2	NMT 0.5 µg/kg*	BDL	BDL	BDL	BDL	BDL
Aflatoxin G1	NMT 0.5 µg/kg*	BDL	BDL	BDL	BDL	BDL
Aflatoxin G2	NMT 0.5 µg/kg*	BDL	BDL	BDL	BDL	BDL
Total Aflatoxins (B1+ B2 + G1 + G2)	NMT 10 µg/kg	BDL	BDL	0.42 µg/kg	0.35 µg/kg	0.28 µg/kg
Compliance Status	—	Complies	Complies	Complies	Complies	Complies

Abbreviations: BDL = Below Detection Limit; NMT = Not More Than.

The aflatoxin analysis revealed that aflatoxins were either below the detection limit or present in trace amounts in all selected batches of Maharasnadi Kadha. The highest level detected was Aflatoxin B1 in MK-3 (0.42 µg/kg), which was significantly below the WHO/AYUSH permissible limit of 5 µg/kg. Total aflatoxin content in all samples remained well within the acceptable limit of 10 µg/kg. Therefore, all evaluated batches complied with international and national safety standards regarding mycotoxin contamination.

DATA ANALYSIS AND COMPARISION

The analytical results obtained for all selected batches of Maharasnadi Kadha (MK-1, MK-2, and MK-3, MK-4,MK-5) were statistically evaluated and compared with the permissible limits prescribed by the World Health Organization (WHO), Ayurvedic Pharmacopoeia of India (API), and Ministry of AYUSH guidelines. The comparison was carried out to assess the safety, quality, and regulatory compliance of the selected formulations with respect to physicochemical characteristics, heavy metal content, microbial contamination, pesticide residues, and aflatoxin levels.

Table no.5.12: Comparative Analysis of Analytical Results with WHO/AYUSH Standards.

Parameter	WHO/AYUSH Permissible Limit	MK-1 (Baidyanath)	MK-2 (Dabur)	MK-3 (Zandu)	MK-4 (Basic Ayurveda)	MK-5 (Dhootapapeshwar)
pH	4.0 –5.0	4.23	4.50	4.13	4.36	4.28
Total Solids (% w/v)	10 –25%	15.32	13.78	19.39	16.84	17.52
Lead (Pb)	NMT 10.0 ppm	1.15 ppm	0.88 ppm	1.42 ppm	0.95 ppm	1.08 ppm
Arsenic (As)	NMT 3.0 ppm	0.22 ppm	0.14 ppm	0.31 ppm	0.19 ppm	0.27 ppm
Mercury (Hg)	NMT 1.0 ppm	BDL	BDL	BDL	BDL	BDL
Cadmium (Cd)	NMT 0.30 ppm	0.04 ppm	0.02 ppm	0.05 ppm	0.03 ppm	0.04 ppm
TAMC (CFU/mL)	NMT 10^5 CFU/mL	3.2×10^2	1.8×10^2	4.1×10^2	2.6×10^2	3.8×10^2
TYMC (CFU/mL)	NMT 10^3 CFU/mL	<10	Absent	1.5×10^1	<10	1.2×10^1
Pathogenic Organisms	Must be Absent	Absent	Absent	Absent	Absent	Absent
DDT Residue (mg/kg)	NMT 1.0 mg/kg	0.012	BDL	0.008	0.010	0.006
Organophosph	NMT 0.20	BDL	0.005	BDL	0.004	BDL

ate Residues (mg/kg)	mg/kg					
Total Aflatoxins (µg/kg)	NMT 10 µg/kg	BDL	BDL	0.42	BDL	0.25

Comparative Evaluation

The pH values of all five batches ranged between **4.13 and 4.50**, which falls within the acceptable range specified for Ayurvedic liquid formulations. The observed pH values indicate appropriate acidity and stability of the formulations, thereby supporting their suitability for storage and therapeutic use. Total solid content varied from **13.78% to 19.39% w/v**, demonstrating adequate extraction, concentration, and uniformity of herbal constituents among the selected marketed products.

Heavy metal analysis revealed that the concentrations of **lead (Pb), arsenic (As), mercury (Hg), and cadmium (Cd)** in all batches were substantially lower than the permissible limits prescribed by WHO and AYUSH guidelines. Lead concentrations ranged from **0.88 to 1.42 ppm**, while arsenic levels varied between **0.14 and 0.31 ppm**. Mercury was **below the detection limit (BDL)** in all tested formulations, and cadmium concentrations ranged from **0.02 to 0.05 ppm**, remaining significantly below the maximum allowable limit of **0.30 ppm**. These findings indicate minimal heavy metal contamination and confirm the safety of the formulations with respect to toxic elemental impurities.

Microbiological examination demonstrated that the **Total Aerobic Microbial Count (TAMC)** ranged from **1.8×10^2 to 4.1×10^2 CFU/mL**, while the **Total Yeast and Mold Count (TYMC)** was either absent, less than 10 CFU/mL, or present in very low numbers. All microbial counts were considerably below the prescribed WHO limits. Furthermore, no pathogenic microorganisms, including **Escherichia coli, Salmonella spp., Staphylococcus aureus,** and **Pseudomonas aeruginosa**, were detected in any of the samples. These results confirm satisfactory microbiological quality and adherence to good manufacturing and hygienic practices. Pesticide residue analysis showed either **non-detectable levels (BDL)** or trace quantities of organochlorine and organophosphate pesticides. DDT residues ranged from **BDL to 0.012 mg/kg**, while organophosphate residues were either absent or detected at very low concentrations (maximum **0.005 mg/kg**). All detected residues were well below the WHO permissible limits, indicating proper agricultural practices and minimal environmental contamination of the herbal raw materials used in manufacturing.

Aflatoxin analysis revealed either complete absence or very low concentrations of total aflatoxins in all batches. The highest detected value was **0.42 µg/kg in MK-3**, followed by **0.25 µg/kg in MK-5**, both of which were substantially lower than the WHO permissible limit of **10 µg/kg** for total aflatoxins. The results indicate effective storage, handling, and quality control measures that prevented fungal contamination and toxin production.

Overall Assessment

Based on comparative evaluation with **WHO Guidelines, Ayurvedic Pharmacopoeia of India (API), and AYUSH standards**, all selected Maharasnadi Kadha formulations (**MK-1, MK-2, MK-3, MK-4, and MK-5**) complied with the prescribed quality, purity, and safety requirements. The findings demonstrate that the formulations are free from significant contamination by heavy metals, microorganisms, pesticide residues, and aflatoxins. Therefore, all evaluated batches can be considered **safe, standardized, and compliant with regulatory quality specifications** for the tested parameters, reflecting satisfactory manufacturing practices and quality assurance measures adopted by the respective manufacturers.

8. CONCLUSION

The present study entitled "Comparative Determination of Potential Contaminants in Maharasnadi Kadha Herbal Formulation as per WHO Guidelines" was undertaken to evaluate the quality, safety, and regulatory compliance of five commercially available Maharasnadi Kadha formulations obtained from different manufacturers. The selected samples included MK-1 (Baidyanath), MK-2 (Dabur), MK-3 (Zandu), MK-4 (Basic Ayurveda), and MK-5 (Dhootapapeshwar).

All five formulations were subjected to comprehensive evaluation involving physicochemical analysis, heavy metal determination, microbial contamination assessment, pesticide residue analysis, and aflatoxin estimation. The analytical results were compared with the permissible limits prescribed by the World Health Organization (WHO), Ayurvedic Pharmacopoeia of India (API), and Ministry of AYUSH guidelines.

The physicochemical evaluation demonstrated that all samples possessed acceptable organoleptic characteristics and exhibited pH values and total solid contents within the prescribed limits, indicating consistency in formulation quality and adequate extraction of herbal constituents.

Heavy metal analysis revealed that the concentrations of lead, arsenic, mercury, and cadmium in all formulations were significantly below the maximum permissible limits. Mercury was below the detection limit in all samples, confirming the absence of toxic heavy metal contamination.

Microbiological examination showed that the Total Aerobic Microbial Count (TAMC) and Total Yeast and Mold Count (TYMC) of all formulations were substantially lower than the prescribed limits. Furthermore, pathogenic microorganisms such as *Escherichia coli, Salmonella spp., Staphylococcus aureus,* and *Pseudomonas aeruginosa* were absent in all tested samples, indicating satisfactory microbiological quality and adherence to good manufacturing and hygienic practices.

Pesticide residue analysis revealed either non-detectable or trace levels of organochlorine and organophosphate pesticides, all of which were well below the WHO-recommended limits.

Similarly, aflatoxin analysis demonstrated either the absence or negligible concentrations of aflatoxins in all formulations. The highest detected aflatoxin level remained considerably below the permissible limit of 10 µg/kg, confirming compliance with international safety standards for mycotoxin contamination.

Based on the comparative evaluation, all five marketed Maharasnadi Kadha formulations complied with WHO, API, and AYUSH quality and safety requirements. The results indicate that the selected formulations are free from significant contamination by heavy metals, microorganisms, pesticide residues, and aflatoxins and can therefore be considered safe and of acceptable quality with respect to the evaluated parameters.

The study emphasizes the importance of routine quality control testing and contaminant monitoring in herbal formulations to ensure consumer safety, product standardization, and regulatory complianc.e. Further investigations involving a larger number of batches, manufacturers, and additional quality parameters may provide deeper insights into batch-to-batch consistency and long-term quality assurance of Ayurvedic herbal formulations.

09. REFERENCES

1. World Health Organization. Quality Control Methods for Medicinal Plant Materials. Geneva: WHO, 1998.
2. World Health Organization. WHO Guidelines on Good Agricultural and Collection

Practices (GACP) for Medicinal Plants. Geneva: WHO, 2003.
3. World Health Organization. Guidelines for the Assessment of Herbal Medicines. Geneva: WHO, 2004.
4. World Health Organization. WHO Guidelines for Assessing Quality of Herbal Medicines with Reference to Contaminants and Residues. Geneva: WHO, 2007.
5. World Health Organization. Quality Assurance of Pharmaceuticals: A Compendium of Guidelines and Related Materials. Geneva: WHO, 2013.
6. Indian Pharmacopoeia Commission. Indian Pharmacopoeia. Ghaziabad: IPC, 2022.
7. Ministry of AYUSH. Ayurvedic Pharmacopoeia of India. New Delhi: Government of India, 2021.
8. Ministry of AYUSH. Ayurvedic Formulary of India. New Delhi: Government of India, 2021.
9. Kunle OF, Egharevba HO, Ahmadu PO. Standardization of Herbal Medicines – A Review. Int J Biodivers Conserv, 2012; 4(3): 101-112.
10. Mukherjee PK. Quality Control and Evaluation of Herbal Drugs. 2nd ed. Amsterdam: Elsevier, 2019.
11. Mukherjee PK. Quality Control of Herbal Drugs: An Approach to Evaluation of Botanicals. New Delhi: Business Horizons, 2002.
12. Kokate CK, Purohit AP, Gokhale SB. Pharmacognosy. 57th ed. Pune: Nirali Prakashan, 2021.
13. Trease GE, Evans WC. Pharmacognosy. 16th ed. London: Saunders Elsevier, 2009.
14. Harborne JB. Phytochemical Methods. 3rd ed. London: Chapman and Hall, 1998.
15. Evans WC. Trease and Evans Pharmacognosy. 16th ed. London: Elsevier, 2009.
16. Sofowora A. Medicinal Plants and Traditional Medicine in Africa. 3rd ed. Ibadan: Spectrum Books, 2008.
17. WHO Expert Committee on Specifications for Pharmaceutical Preparations. WHO Technical Report Series No. 1010. Geneva: WHO, 2018.
18. Anonymous. The Ayurvedic Pharmacopoeia of India. Part I, Vol. I–IX. New Delhi: Ministry of AYUSH.
19. Anonymous. Protocol for Testing Ayurvedic, Siddha and Unani Medicines. New Delhi: Pharmacopoeial Laboratory for Indian Medicine, 2008.
20. AOAC International. Official Methods of Analysis. 21st ed. Maryland: AOAC International, 2019.
21. United States Pharmacopeia. USP–NF. Rockville: USP Convention, 2023.

22. European Pharmacopoeia Commission. European Pharmacopoeia. 11th ed. Strasbourg, 2023.
23. Sahoo N, Manchikanti P, Dey S. Herbal drugs: Standards and regulation. Fitoterapia. 2010; 81(6): 462-471.
24. Bent S. Herbal medicine in the United States: Review of efficacy, safety, and regulation. J Gen Intern Med. 2008; 23(6): 854-859.
25. Ernst E. Toxic heavy metals and undeclared drugs in Asian herbal medicines. Trends Pharmacol Sci. 2002; 23(3): 136-139.
26. Chan K. Some aspects of toxic contaminants in herbal medicines. Chemosphere. 2003; 52(9): 1361-1371.
27. Calixto JB. Efficacy, safety, quality control and regulation of herbal medicines. Braz J Med Biol Res. 2000; 33(2): 179-189.
28. Posadzki P, Watson L, Ernst E. Contamination and adulteration of herbal medicinal products. Clin Med. 2013; 13(1): 7-12.
29. Luo L, Wang B, Jiang J, et al. Heavy metal contamination in herbal medicines. Front Pharmacol. 2021; 11: 595335.
30. Abou-Arab AAK, Abou Donia MA. Heavy metals in Egyptian medicinal plants. Food Chem Toxicol. 2000; 38(8): 765-778.
31. Khan S, Cao Q, Zheng YM, Huang YZ, Zhu YG. Health risks of heavy metals in medicinal plants. Environ Pollut. 2008; 152(3): 686-692.
32. Singh P, Shrivastava R, Sharma V. Heavy metal contamination in herbal medicines. Int J Green Pharm. 2010; 4(3): 176-179.
33. Opuni K, Annan K, Mensah AY. Contamination of herbal medicinal products in low- and middle-income countries. Front Pharmacol. 2023; 14: 10477504.
34. Ezekwesili-Ofili JO, Okaka ANC. Herbal medicines and microbial contamination. J Biol Agric Healthc. 2014; 4(5): 12-20.
35. Bugno A, Buzzo AA, Nakamura CT, et al. Microbial contamination in herbal medicines. Rev Inst Med Trop Sao Paulo. 2005; 47(3): 149-155.
36. Martins HM, Martins ML, Dias MI, Bernardo F. Evaluation of microbiological quality of medicinal plants. J Food Prot. 2001; 64(12): 2011-2014.
37. De Smet PAGM. Health risks of herbal remedies. Drug Saf. 2004; 27(11): 823-837.
38. Kosalec I, Cvek J, Tomic S. Contaminants of medicinal herbs. Arh Hig Rada Toksikol. 2009; 60(4): 485-501
39. Tripathy V, Basak BB, Varghese TS, Saha A. Residues and contaminants in medicinal

herbs. J Nat Sci Biol Med. 2015; 6(1): 1-7.

40. Roy S, Chanda S. Detection of pesticide residues in medicinal plants. Pharmacogn Rev. 2014; 8(15): 83-89.
41. Ravishankar, B., et al. (2014). *Observations on the Quality of commercially manufactured Ayurvedic Decoction, Maharasnadi Kvatha.* International Journal of Pharmaceutical Sciences and Research.
42. World Health Organization. (1998). Quality control methods for medicinal plant materials. Geneva: WHO
43. Skoog, D. A., Holler, F. J., & Crouch, S. R. (2018). Principles of Instrumental Analysis (7th ed.). Boston, MA: Cengage Learning.
44. World Health Organization. (1999). Quality control methods for medicinal plant materials. Geneva: World Health Organization.
45. Ministry of AYUSH. (2022). The Ayurvedic Pharmacopoeia of India (Part I, Volumes I–IX). New Delhi: Government of India.
46. Ravishankar, B., et al. (2016). *Observations on the Quality of Commercially Manufactured Ayurvedic Decoction Maharasnadi Kwatha. International Journal of Pharmaceutical Sciences and Research.*