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IN VITRO EVALUATION OF THE ANTIBACTERIAL ACTIVITY OF *ARKADI GANA* AGAINST ENTEROPATHOGENIC BACTERIA ASSOCIATED WITH FOOD POISONING

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ABSTRACT

Background: Food poisoning caused by enteropathogenic bacteria is a major global health burden, and rising antimicrobial resistance (AMR) among these pathogens has renewed interest in plant-based alternatives. Ayurveda describes a group of drugs, *Arkadi Gana* (*Sushruta Samhita, Sutra Sthana* 38/16–17), traditionally credited with *Krimighna* (antimicrobial) and *Vishaghna* (antitoxic) properties. **Objective:** To evaluate, in vitro, the bactericidal/bacteriostatic activity of aqueous extracts of four constituent drugs of *Arkadi Gana* — *Arka* (*Calotropis procera*, leaf), *Apamarga* (*Achyranthes aspera*, seed), *Karanja* (*Pongamia pinnata*, seed) and *Bharangi* (*Clerodendrum serratum*, root) — individually and in combination, against bacterial strains implicated in food poisoning. **Methods:** Aqueous extracts were prepared by cold maceration and concentrated using a rotary evaporator. Physicochemical (pH, loss on drying, ash values, extractive values) and phytochemical standardisation was carried out as

per *Ayurvedic Pharmacopoeia of India* protocols. Antibacterial activity was screened by the agar well diffusion method at 5 mg/mL and 10 mg/mL against Bacillus subtilis MTCC121, Staphylococcus aureus MTCC96, Salmonella typhimurium MTCC733 and Escherichia coli MTCC43, using tetracycline as positive control and distilled water as negative control. **Results:** At 5 mg/mL, none of the extracts produced a measurable zone of inhibition (ZOI) against any strain. At 10 mg/mL, the *Karanja* extract produced a 3 mm ZOI and the combined *Arkadi Gana* extract a 1 mm ZOI, both selectively against S. typhimurium; no activity was observed against S. aureus, B. subtilis or E. coli. Tetracycline produced robust inhibition across all four strains (4–8 mm), validating the assay. **Conclusion:** Aqueous extracts of *Karanja* show selective, concentration-dependent antibacterial activity against Salmonella typhimurium, partially supporting the classical *Krimighna* attribution of *Arkadi Gana*. Overall antibacterial activity of the aqueous extracts was limited and solvent-dependent, indicating that alternative extraction media and higher concentrations should be explored in future work.

KEYWORDS: *Arkadi Gana*; Ayurveda; food poisoning; antibacterial activity; Salmonella typhimurium; *Krimighna*; antimicrobial resistance; agar well diffusion.

1. INTRODUCTION

Food poisoning, defined as acute gastroenteritis following ingestion of food or water contaminated with pathogenic microorganisms or their toxins, remains a significant global public health problem. The worldwide burden of infectious diarrhoea is estimated at 3–5 billion cases and nearly 1.8 million deaths annually, predominantly among young children.[1] Bacterial pathogens such as Salmonella spp., Shigella spp., Staphylococcus aureus, Bacillus cereus, Clostridium perfringens and pathogenic Escherichia coli are among the most frequently implicated agents.[2]

Ayurveda, the science of life, describes the awareness of disease-causing factors, clinical presentation and therapeutics as an eternal and sacred *Trisutra* (three fundamental principles), as codified by *Acharya Charaka*:

[illegible] |
[illegible] ||

(Charaka Samhita, Sutra Sthana 1/24)

The knowledge of causative factors (*Hetu*), clinical signs (*Linga*) and therapeutics

(*Aushadha*) constitutes this *Trisutra*, which forms the philosophical bedrock for the diagnosis and management of every disease described in classical Ayurveda, including food-borne toxic conditions.

The problem is compounded by the escalating global crisis of antimicrobial resistance (AMR). The World Health Organization attributed approximately 1.27 million deaths in 2019 directly to drug-resistant bacterial infections, with low- and middle-income countries, including India, bearing the greatest burden; unaddressed, AMR is projected to cause up to 10 million deaths annually by 2050.[3] Indian surveillance data (ICMR) have documented rising resistance among Escherichia coli, Klebsiella pneumoniae and Salmonella Typhi, alongside declining efficacy of key antibiotic classes.[4] This has intensified the search for effective, low-toxicity antimicrobial alternatives, including plant-derived agents.

Ayurveda, the traditional Indian system of medicine, describes the concept of *Krimi* (organisms, ranging from macroscopic parasites to microscopic pathogens) as a causative factor in disease, and *Annavisha* (food-derived toxicity arising from improperly digested or putrefied food) as a cause of diarrhoeal illness resembling modern food poisoning.[5] *Acharya Sushruta* defined the specialised branch dealing with poisons and their management, *Agada Tantra*, as follows:

[illegible]
[illegible] च [illegible]

(Sushruta Samhita, Sutra Sthana 1/6)

This defines *Agada Tantra* as the branch concerned with identifying poisoning caused by snakes, insects and other creatures, and with the pacification of diverse forms of poisoning, providing the classical toxicological framework within which food poisoning is discussed. *Acharya Atreya*, in the *Grahani Dosha Chikitsa* chapter of *Charaka Samhita*, further describes how improperly digested food acquires a fermented, toxic character:

[illegible] ‖ [illegible] च [illegible] |
[illegible]

(Charaka Samhita, Chikitsa Sthana 15/44–49)

Undigested food (*Apachyamana Anna*), if it persists in the *Amashaya*, develops a fermented (*Shukta*) quality and is transformed into a potent food-poison (*Ghora Annavisha*), which then vitiates the *Doshas* and produces disease — a description that closely parallels the modern

understanding of bacterial contamination and toxin elaboration in improperly stored or handled food. *Acharya Gananath Sen* was the first to use the term *Jeevanu* (literally, 'living atom' or microbe) and to describe *Annavisha*-associated diarrhoea in terms closely paralleling bacterial food poisoning and its toxin-mediated pathogenesis.[6] Classical Ayurvedic management of *Krimi*-related disease follows a threefold strategy described by *Acharya Charaka* — *Apakarshana* (mechanical/therapeutic removal of the organism), *Prakriti Vighata* (creation of an environment unfavourable to microbial growth using drugs of *Katu-Tikta rasa*, *Ushna virya* and *Katu vipaka*) and *Nidana Parivarjana* (avoidance of causative factors).[7]

Among the drug groups (*Gana*) described by *Acharya Sushruta* in *Sutra Sthana* (Chapter 38), *Arkadi Gana* comprises 10 drugs traditionally attributed with *Kapha-Medohara*, *Vishapaha* and *Krimikushtha-Prashamana* properties, and is specifically indicated for *Vrana Shodhana* (wound cleansing).[8] *Acharya Sushruta* describes the therapeutic actions of this Gana thus:

[illegible]

(Sushruta Samhita, Sutra Sthana 38/17)

This *Gana*, headed by Arka, is said to alleviate *Kapha*, *Medas* (fat) and *Visha* (poison/toxins), to pacify *Krimi* (microorganisms/worms) and *Kushtha* (skin disorders), and to be particularly effective in *Vrana Shodhana* (wound cleansing) — a direct classical statement of the group's *Krimighna* (antimicrobial) potential that provided the specific rationale for the present investigation. Four constituent drugs of this Gana — *Arka* (*Calotropis procera* Linn., leaf), *Apamarga* (*Achyranthes aspera* Linn., seed), *Karanja* (*Pongamia pinnata* Linn., seed) and *Bharangi* (*Clerodendrum serratum* Linn., root) — are independently documented across the *Brihatrayi* and the *Nighantu* literature for *Krimighna* (anthelmintic/antimicrobial) and *Vishaghna* (antitoxic) actions.[9] Despite these long-standing textual claims, the antimicrobial efficacy of these four drugs, singly and in combination, against clinically relevant enteropathogenic bacteria has not been adequately validated through modern experimental methods.

The present study was therefore undertaken to scientifically evaluate the in vitro antibacterial activity of aqueous extracts of *Arka*, *Apamarga*, *Karanja* and *Bharangi*, individually and in a combined (*Arkadi Gana*) preparation, against four bacterial strains representative of common food-poisoning pathogens — Bacillus subtilis MTCC121, Staphylococcus aureus MTCC96, Salmonella typhimurium MTCC733 and Escherichia coli MTCC43 — with the specific aims

of: (i) assessing the bactericidal effect of each drug and of the combined formulation; (ii) assessing the bacteriostatic effect of each drug and of the combined formulation; and (iii) generating experimental evidence to substantiate or refute the classical Ayurvedic attribution of *Krimighna*/*Vishaghna* activity to Arkadi Gana in the specific context of bacterial food poisoning.

2. MATERIALS AND METHODS

2.1 Study design and setting

This was an experimental, in vitro laboratory study conducted as part of a postgraduate dissertation in the P.G. Department of *Agada Tantra*, Rajiv Gandhi Govt. Post Graduate Ayurvedic College & Hospital, Paprola, Distt. Kangra, Himachal Pradesh (affiliated to Atal Medical and Research University, Himachal Pradesh). Collection, authentication and pharmacognostic evaluation of raw drugs were carried out at the Department of *Ras Shastra* of the same institute, and extract preparation, physicochemical standardisation and the microbiological assay were performed at the Fermentation and Phyto-Farming Technology Division, CSIR – Institute of Himalayan Bioresource Technology (IHBT), Palampur, Himachal Pradesh, India.

2.2 Plant material

Four ingredients of *Arkadi Gana* (*Sushruta Samhita*, *Sutra Sthana* 38/16) were selected for study, as summarised in Table 1.

Table 1: Ingredients of Arkadi Gana selected for the study.

Drug (Sanskrit name)	Botanical name	Family	Part used
Arka	*Calotropis procera* (Ait.) R.Br.	Apocynaceae (Asclepiadaceae)	Leaf
Apamarga	*Achyranthes aspera* Linn.	Amaranthaceae	Seed
Karanja	*Pongamia pinnata* (L.) Pierre	Leguminosae (Fabaceae)	Seed
Bharangi	*Clerodendrum serratum* Linn.	Verbenaceae/Lamiaceae	Root

2.3 Preparation of aqueous extract

Each drug was cleaned, shade-dried and coarsely powdered separately. For standardisation, 4 g of each air-dried coarse powder was macerated with 100 mL distilled water, shaken intermittently over 6 hours and allowed to stand for 18 hours (total maceration 24 hours), then filtered. For the antimicrobial study, 5 g of each air-dried powder was macerated with

100 mL distilled water (solvent-to-drug ratio maintained at 3–4 times the drug weight), filtered sequentially through muslin cloth and Whatman filter paper, and the filtrate concentrated under reduced pressure using a rotary evaporator at 70–80 °C. The extraction cycle was repeated three times per batch to maximise recovery of active constituents. A combined extract of all four drugs (equal proportions) was prepared using an identical protocol. Extractive value was calculated as: Extractive value (%) = [(W2 − W1)/W] × 100, where W = initial drug weight, W1 = weight of empty flask, and W2 = weight of flask with dried extract.

2.4 Physicochemical and phytochemical standardisation

Physicochemical parameters — pH, loss on drying (moisture content), total ash, acid-insoluble ash, water-soluble ash, and water/alcohol/hydro-alcohol soluble extractive values — were determined as per standard protocols of the *Ayurvedic Pharmacopoeia of India*. Preliminary phytochemical screening for alkaloids (Dragendorff's test), carbohydrates (Molisch's test), proteins/amino acids (Ninhydrin test), flavonoids (H_2 SO_4 and alkaline reagent tests), phenolic compounds (iodine test), tannins (Braymer's test), terpenoids (Salkowski test), quinones (sulphuric acid test), cardiac glycosides (Keller-Killiani test) and volatile oils (fluorescence test) was carried out on each extract.

2.5 Bacterial strains and culture conditions

Four characterised bacterial strains, procured from the Department of Fermentation and Phyto-Farming Technology, CSIR-IHBT, Palampur, were used (Table 2). These strains were selected as representative pathogens implicated in bacterial food poisoning and febrile illness, and for their documented multidrug-resistance patterns per WHO GLASS surveillance data.[10]

Table 2: Bacterial strains used in the antimicrobial study.

Bacterial strain	MTCC number	Gram status
Bacillus subtilis	MTCC121	Gram-positive
Staphylococcus aureus	MTCC96	Gram-positive
Salmonella typhimurium	MTCC733	Gram-negative
Escherichia coli	MTCC43	Gram-negative

Stock cultures were revived in nutrient broth (20 mL per flask, autoclaved at 121 °C for 20 minutes), inoculated aseptically in a laminar flow chamber and incubated overnight at room temperature on a shaker. Pure cultures were obtained by sub-culturing onto Muller-Hinton

agar (MHA) plates, incubated in an inverted position at 37 °C for 24 hours.

2.6 Antibacterial assay — agar well diffusion method

Antibacterial activity was assessed by the agar well diffusion method.[11] A 100 µL bacterial suspension was spread on MHA plates using a sterile cotton swab to form a confluent lawn and allowed to stand for 10–15 minutes. Wells of 6 mm diameter were bored using a sterile gel puncture. Fifty microlitres of each test extract (10% w/v aqueous solution, corresponding to 5 mg/mL and 10 mg/mL working concentrations) were introduced into the respective wells. Distilled water served as the negative control and tetracycline (concentration selected as per Clinical and Laboratory Standards Institute [CLSI] guidelines) as the positive control. Plates were incubated at 37 °C for 18–20 hours. The experiment was performed in triplicate for each extract–strain combination.

Table 3: Sample coding used in the antimicrobial evaluation.

Sample	Extract	Code
1	*Apamarga* (aqueous)	E1
2	*Bharangi* (aqueous)	E2
3	*Arka* (aqueous)	E3
4	*Karanja* (aqueous)	E4
5	Combined *Arkadi Gana* (aqueous)	E5
6	Distilled water	C (negative control)

Sample codes used throughout the study are given in Table 3.

Sample	Extract	Code
7	Tetracycline	TE (positive control)

2.7 Measurement and interpretation of results

Following incubation, the diameter of the zone of complete inhibition (ZOI, in mm, including the well diameter) was measured with a ruler on the underside of the plate. Faint growth (~80% inhibition) within the zone was disregarded, and measurement was taken to the margin of heavy growth. The mean of triplicate readings was recorded. The Activity Index (AI) of each test sample was calculated as: AI = ZOI of test sample / ZOI of standard (tetracycline).

3. RESULTS

3.1 Organoleptic and physicochemical characteristics

Macroscopic evaluation confirmed the identity of the raw drugs (Table 4). Physicochemical parameters (Table 5) were within the ranges reported in the *Ayurvedic Pharmacopoeia of*

India for these drugs, confirming acceptable quality of the raw material used.

Table 4: Organoleptic characteristics of raw drugs of Arkadi Gana.

Drug	Colour	Odour	Taste	Touch
Arka	Greyish green	Characteristic	Bitter & pungent	Rough
Apamarga	Yellowish brown	Characteristic	Slightly bitter & pungent	Rough & hairy
Karanja	Brown	Strong, unpleasant, pungent	Bitter & acrid	Smooth
Bharangi	Dark brown	Slightly earthy	Bitter, pungent	Rough

Table 5: Physicochemical parameters of raw drugs of Arkadi Gana.

Drug	pH	Loss on drying (%)	Total ash (%)	Acid-insoluble ash (%)	Water-soluble ash (%)
Arka	5.94	9.37	9.1	1.6	2.18
Apamarga	6.52	11.65	9.75	2.29	2.49
Karanja	6.34	12.23	7.98	0.69	3.19
Bharangi	6.73	7.78	2.74	2.25	0.48

3.2 Phytochemical screening

Preliminary phytochemical screening (Table 6) revealed that alkaloids were most prominent in *Bharangi* (+++), carbohydrates in *Karanja* (+++), and flavonoids and terpenoids were notably present in *Apamarga* and *Karanja* extracts (++), while Arka extract was largely negative for flavonoids, tannins, terpenoids and quinones. Tannins were detected only in Karanja.

Table 6: Phytochemical screening of aqueous extracts of Arkadi Gana ingredients. (+++ strongly present, ++ moderately present, + weakly present, − absent).

Phytoconstituent	*Arka*	*Apamarga*	*Karanja*	*Bharangi*
Alkaloids	++	+	+	+++
Carbohydrates	+	−	+++	+
Proteins/amino acids	+	++	+	−
Flavonoids	−	++	++	+
Phenolic compounds	+	++	+	−
Tannins	−	−	+	−
Terpenoids	−	++	+	−
Quinones	−	+	+	−
Volatile oils	−	+	++	+

3.3 Antibacterial activity

At a concentration of 5 mg/mL, none of the five test extracts (E1–E5) produced a measurable zone of inhibition against any of the four bacterial strains tested (Table 7). Tetracycline

produced ZOIs of 3 mm, 6 mm, 8 mm and 5 mm against B. subtilis, S. aureus, S. typhimurium and E. coli respectively, confirming assay validity.

Table 7: Zone of inhibition (mm) at 5 mg/mL extract concentration. (— = no zone of inhibition).

Sample	B. subtilis (G+)	S. aureus (G+)	S. typhimurium (G−)	E. coli (G−)
E1 (*Apamarga*)	—	—	—	—
E2 (*Bharangi*)	—	—	—	—
E3 (*Arka*)	—	—	—	—
E4 (*Karanja*)	—	—	—	—
E5 (Combined)	—	—	—	—
C (Water)	—	—	—	—
TE (Tetracycline)	3	6	8	5

At 10 mg/mL (Table 8), the *Karanja* extract (E4) produced a 3 mm ZOI against S. typhimurium, and the combined *Arkadi Gana* extract (E5) produced a 1 mm ZOI against the same organism. None of the extracts showed activity against B. subtilis, S. aureus or E. coli at this concentration. Tetracycline again showed activity across all strains (4, 7, 8 and 4 mm respectively against B. subtilis, S. aureus, S. typhimurium and E. coli).

Table 8: Zone of inhibition (mm) at 10 mg/mL extract concentration. (— = no zone of inhibition).

Sample	B. subtilis (G+)	S. aureus (G+)	S. typhimurium (G−)	E. coli (G−)
E1 (*Apamarga*)	—	—	—	—
E2 (*Bharangi*)	—	—	—	—
E3 (*Arka*)	—	—	—	—
Sample	**B. subtilis (G+)**	**S. aureus (G+)**	**S. typhimurium (G−)**	**E. coli (G−)**
E4 (*Karanja*)	—	—	3	—
E5 (Combined)	—	—	1	—
C (Water)	—	—	—	—
TE (Tetracycline)	4	7	8	4

Activity Index (AI = ZOI of test sample / ZOI of standard) at 10 mg/mL was 0.375 for *Karanja* (E4) and 0.125 for the combined extract (E5) against S. typhimurium, indicating modest but reproducible activity relative to tetracycline.

3.4 Summary of antibacterial findings

Across both tested concentrations: (i) none of the individual extracts of *Arka*, *Apamarga* or *Bharangi* produced any measurable ZOI against any of the four strains; (ii) *Karanja* aqueous extract was the only individual extract to show activity, and this was concentration-dependent

(absent at 5 mg/mL, present at 10 mg/mL) and organism-selective (active only against the Gram-negative S. typhimurium); (iii) the combined Arkadi Gana extract showed a smaller ZOI than *Karanja* alone against S. typhimurium, suggesting no additive or synergistic enhancement of activity under the aqueous extraction conditions used; and (iv) none of the test extracts showed any activity against the Gram-negative E. coli or either Gram-positive strain tested, at either concentration.

4. DISCUSSION

This study aimed to experimentally validate the classical Ayurvedic attribution of *Krimighna* and *Vishaghna* properties to *Arkadi Gana* in the specific context of bacterial food poisoning. The *Rasa Panchaka* (pharmacodynamic attributes) of the four study drugs — predominantly *Katu-Tikta Rasa*, *Laghu-Ruksha-Tikshna guna*, *Ushna Virya* and *Katu Vipaka* — are classically associated with *Kapha-Vatahara* and *Krimighna karma*, and *Acharya Charaka* specifically describes drugs of this pharmacodynamic profile as effective in *Prakriti Vighata chikitsa*, wherein an environment unfavourable to microbial proliferation is created. The selective activity observed for Karanja against Salmonella typhimurium is consistent with this classical framework and also aligns with prior reports of antimicrobial, anti-giardial and anti-rotaviral activity of Pongamia pinnata leaf and seed extracts, and of the ability of this plant's extracts to reduce cholera toxin production and inhibit bacterial invasion of epithelial cells — indicating a degree of selective anti-diarrhoeal potential that is broadly consistent with the present findings.[12]

However, the overall magnitude and spectrum of antibacterial activity observed in this study were modest. No zone of inhibition was recorded for any extract at 5 mg/mL, and only *Karanja* and the combined extract showed activity, and that too only against S. typhimurium, at 10 mg/mL. None of the extracts inhibited E. coli, S. aureus or B. subtilis at either concentration tested. This restricted spectrum may be explained by several factors. First, aqueous extraction, while classically described (*Jala Siddha kalpana*) and practically convenient, may not efficiently solubilise the lipophilic or high-molecular-weight antimicrobial constituents (e.g., cardenolides in *Arka*, triterpenoid saponins in *Apamarga*, furanoflavonoids in *Karanja*) that have shown activity in prior studies using alcoholic or hydro-alcoholic solvents.[13] Second, the structurally complex, lipopolysaccharide-rich outer membrane of Gram-negative bacteria such as E. coli, and the thick peptidoglycan layer combined with efficient efflux mechanisms in Gram-positive S. aureus and B. subtilis, are

known to restrict penetration of many plant-derived phytoconstituents, which may explain the lack of activity against these organisms despite activity against S. typhimurium.[14] Third, the relatively low concentrations tested (5–10 mg/mL) are modest compared with concentrations used in several published phytochemical antibacterial screens, and higher concentrations may reveal activity not detected here.

The phytochemical screening results provide some mechanistic context: *Karanja* extract showed the strongest reaction for carbohydrates and detectable tannins, phenolics and terpenoids, phytoconstituent classes that have each independently been associated with antibacterial activity through mechanisms including membrane disruption, enzyme inhibition and metal ion chelation.[15] *Bharangi* extract, despite showing the strongest alkaloid reaction, did not translate this into measurable antibacterial activity in the present assay, underscoring that qualitative phytochemical presence does not necessarily predict functional antimicrobial potency at the concentrations and solvent system tested.

These findings should be interpreted in light of the Ayurvedic conceptual framework reviewed in this study. As early as the *Atharvaveda*, two categories of causative organism were recognised — visible (*Drishya*) and invisible (*Adrishya*):

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(Atharvaveda 2/31/2)

This early classification of pathogenic (*Durnama*) and non-pathogenic (*Sunama*) *Krimi*, together with the *Samhita*-period articulation of contagion (*Aupasargika Vyadhi*) and epidemic causation (*Janapadodhwansa*), anticipates several elements of the modern germ theory of disease, and specifically links improperly digested or putrefied food (*Annavisha*) to diarrhoeal illness resembling bacterial food poisoning.[5,6] The selective activity of *Karanja* against a Gram-negative enteropathogen provides a degree of scientific corroboration for these classical descriptions, even though the present aqueous-extract data do not support broad-spectrum antibacterial efficacy for the Gana as a whole.

The strong, consistent inhibition produced by tetracycline across all four strains, contrasted with no activity for the negative control (distilled water), confirms the internal validity of the experimental design and supports confidence that the absence of activity for most test extracts reflects a genuine biological finding rather than a methodological artefact.

5. CONCLUSION

Within the limits of this in vitro study, the aqueous extract of *Karanja* (Pongamia pinnata seed) demonstrated selective, concentration-dependent antibacterial activity against Salmonella typhimurium, partially corroborating the classical Ayurvedic *Krimighna* attribution of *Arkadi Gana*. Aqueous extracts of *Arka*, *Apamarga* and *Bharangi*, individually, and the combined *Arkadi Gana* preparation, showed minimal to no measurable antibacterial activity against the panel of Gram-positive and Gram-negative organisms tested under the conditions of this study. These findings suggest that the antibacterial potential of *Arkadi Gana* is real but narrow in spectrum and highly dependent on the specific drug, solvent system and concentration used, and that further optimisation of extraction methodology is required before the traditional indication of this *Gana* for food-poisoning-associated bacterial infections can be considered fully validated.

REFERENCES

1. Foodborne Infections and Intoxications. 5th ed. Morris JG Jr, Vugia DJ, editors. Amsterdam: Academic Press (Elsevier), 2021; 3–17.
2. Bad Bug Book: Foodborne Pathogenic Microorganisms and Natural Toxins Handbook. 2nd ed. Silver Spring (MD): U.S. Food and Drug Administration, 2012.
3. World Health Organization. Antimicrobial resistance: global report on surveillance. Geneva: WHO, 2022.
4. Indian Council of Medical Research. Annual Report, Antimicrobial Resistance Surveillance and Research Network (AMRSN). New Delhi: ICMR, 2022.
5. Agnivesha, Charaka, Dridhabala. Charaka Samhita, Chikitsa Sthana 15/44–49, Vimana Sthana 7/9–14. Shastri K, Chaturvedi G, editors (Hindi commentary). Varanasi: Chaukhambha Bharati Academy, 2019.
6. Sen G. Siddhanta Nidana, Antrika Jwara Adhyaya and Atisara Adhikara. Varanasi: Chaukhambha Sanskrit Series.
7. Agnivesha, Charaka, Dridhabala. Charaka Samhita, Vimana Sthana 7/14. Shastri K, Chaturvedi G, editors. Varanasi: Chaukhambha Bharati Academy, 2019.
8. Sushruta. Sushruta Samhita, Sutra Sthana 38/16–17. Shastri AD, editor (Ayurveda-tattva-sandipika Hindi commentary). Varanasi: Chaukhambha Sanskrit Sansthan.
9. Sharma PV. Dravyaguna Vijnana. Vol. 2. Varanasi: Chaukhambha Bharati Academy, 2009; 543–544.

10. World Health Organization. Global Antimicrobial Resistance and Use Surveillance System (GLASS) Report 2022–2024. Geneva: WHO, 2024.
11. Perez C, Pauli M, Bazerque P. An antibiotic assay by the agar-well diffusion method. Acta Biol Med Exp., 1990; 15: 113–115.
12. Brijesh S, Daswani PG, Tetali P. Studies on Pongamia pinnata (L.) Pierre leaves: understanding the mechanism(s) of action in infectious diarrhea. J Zhejiang Univ Sci B., 2006; 7(8): 665–674.
13. Government of India, Ministry of AYUSH. The Ayurvedic Pharmacopoeia of India. Part I, Vol I. 1st ed. New Delhi: Dept. of ISM & H, 1999.
14. Nikaido H. Molecular basis of bacterial outer membrane permeability revisited. Microbiol Mol Biol Rev., 2003; 67(4): 593–656.
15. Cowan MM. Plant products as antimicrobial agents. Clin Microbiol Rev., 1999; 12(4): 564–582.