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Research


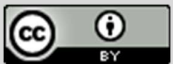
Exploring The Antimicrobial Activity Of Kabasura Kudineer Chooranam And Nilavembu Kudineer Chooranam

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	Abstract
Published on: 23 Jan 2024	The present study examines the efficacy of Kabasura kudineer chooranam and Nilavembu kudineer chooranam, a siddha formulation. Kabasura kudineer is a well-known Siddha medicine comprising a total of 15 herbal ingredients. This powdered chooranam is primarily intended for enhancing respiratory health, strengthening the lungs, and addressing conditions such as fever, cold, and cough. Nilavembu Kudineer chooranam is an renowned for enhancing immunity levels. Crafted from a blend of 9 distinct herbs, this formulation serves as an effective defense against fever, exerting anti-inflammatory, analgesic, and anti-viral properties. The primary component, Nilavembu, known as Kalmegh in Sanskrit, possesses remarkable capabilities in treating liver disorders, viral fevers, jaundice, dengue, chikungunya, and respiratory issues. Nilavembu Kudineer exhibits antimicrobial, analgesic (pain-relieving), antipyretic (fever-reducing), and antiviral properties, making it effective in addressing viral and malaria infections.
Published by: DrSriram Publications	Keywords: Kabasura kudineer, Nilavembu kudineer, chooranam, antimicrobial activity, Siddha medicine.
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INTRODUCTION

SIDDHA SYSTEM OF MEDICINE

Siddha medicine, an ancient medical system extensively practiced in South India, has a history that spans many centuries⁽¹⁾. The Siddha system of medicine stands as one of India's remarkable cultural inheritances⁽²⁾. The Siddha system of medicine, a distinctive science and unique healing art, emerged and prospered in South India, continuing to be practiced through both traditional and hereditary methods⁽³⁾. The central principle of Siddha medicine revolves around achieving harmony with nature and the universe, which is pivotal in the treatment of various illnesses⁽⁴⁾.

KABASURA KUDINEER CHOORANAM

Kabasura kudineer is a well-known Siddha medicine comprising a total of 15 herbal ingredients. This powdered chooranam is primarily intended for enhancing respiratory health, strengthening the lungs, and addressing conditions such as fever, cold, and cough. It is typically consumed in the form of a decoction after soaking it in water, often referred to as Kadha⁽⁵⁾. Kapacurak Kutinir (KK) is a polyherbal Siddha formulation documented in the manuscript Citta Vaittiyattirattu, employed for the treatment of phlegmatic fever and fever accompanied by flu-like symptoms⁽³⁾. "Kaba" signifies the kapha dosha, representing fever arising from an excess accumulation of kapha (mucus or phlegm). "Asura" refers to herbs that alleviate these symptoms, "Kudineer" indicates a decoction, and "Chooranam" denotes a powdered form. This preparation has been noted for its antibacterial, anti-inflammatory, and antipyretic properties. This product is readily accessible and culturally embraced⁽⁶⁾. KSK chooranam is a traditional Siddha medicine formulation used for common respiratory issues, known for its anti-inflammatory, analgesic, antiviral, antioxidant, and antibacterial properties. It helps enhance lung health and respiratory function⁽⁷⁾.

INGREDIENTS OF KABASURA KUDINEER CHOORANAM**Table 1: Ingredients of Kabasura Kudineer Chooranam**

S.No	Botanical name of ingredients	Part used
1	<i>Zingiber officinale</i> Roscoe	Dried Rhizome
2	<i>Andrographis paniculate</i> Burm.f.Nees	Whole plant
3	<i>Syzygium aromaticum</i> (L.) Merril. And Perry	Flower bud
4	<i>Cyperus rotundus</i> L.	Rhizome
5	<i>Sida acuta</i> Burm. f.	Root
6	<i>Rothea serrata</i> (L.) Steane and Mabb.	Root
7	<i>Piper longum</i> L.	Fruit
8	<i>Justicia adathoda</i> L.	Leaves
9	<i>Plectranthus amboinicus</i> (Lour.) Spreng.	Leaf
10	<i>Terminalia chebula</i> (Gaertn.) Retz.	Fruit rind
11	<i>Tinospora cordifolia</i> (Willd.) Miers ex Hook.f and Thoms	Stem
12	<i>Saussurea costus</i> (Falc.) Lipsh	Tuber
13	<i>Tragia involucrata</i> L.	Root
14	<i>Anacyclus pyrethrum</i> (L.) Lag.	Root
15	<i>Hygrophila auriculata</i> (K. Schum.) Heine	Root

NILAVEMBU KUDINEER CHOORANAM

Nilavembu Kudineer chooranam is renowned for enhancing immunity levels. Crafted from a blend of 9 distinct herbs, this formulation serves as an effective defense against fever, exerting anti-inflammatory, analgesic, and anti-viral properties. The primary component, Nilavembu, known as Kalmegh in Sanskrit, possesses remarkable capabilities in treating liver disorders, viral fevers, jaundice, dengue, chikungunya, and respiratory issues. Nilavembu Kudineer exhibits antimicrobial, analgesic (pain-relieving), antipyretic (fever-reducing), and antiviral properties, making it effective in addressing viral and malaria infections. It contributes to bolstering the body's immunity, enabling it to resist and combat recurring fevers. This well-balanced formulation acts on Vata, Pitta, and Kapha, promoting comprehensive health benefits⁽⁸⁾. It is a polyherbal blend, utilizes *Andrographis paniculata* as the key component to manage various fevers and associated body aches. Widely referred to as the Neem of the ground, Bile of earth, and King of bitters, this herb is indigenous to India and Sri Lanka⁽⁹⁾.

INGREDIENTS OF NILAVEMBU KUDINEER CHOORANAM**Table 2: Ingredients of Nilavembu Kudineer Chooranam**

S.No	Botanical name of ingredients	Part used
1	<i>Andrographis paniculate</i> Burm.f.Nees	Whole plant
2	<i>Chrysopogon zizanoides</i> (L.) Roberty	Root
3	<i>Santalum album</i> L.	Bark
4	<i>Zingiber officinale</i> Roscoe	Rhizome
5	<i>Piper nigrum</i> L.	Fruit

6	<i>Cyperus rotundus</i> L.	Root tuber
7	<i>Mollugo cerviana</i> (L.) Ser.	Whole plant
8	<i>Plectranthus vittiveroides</i> (Jacob) Singh and Sharma	Root
9	<i>Trichosanthes cucumerina</i> L.	Whole plant

MATERIALS AND METHODS

MATERIALS USED FOR THE STUDY

Table 3: List of Equipments and Apparatus

Equipments	Apparatus
Isomantle	Soxhlet extractor
Mortar and Pestle	Porous cellulose thimble
Stands and Clamps	Round bottom flask
Petriplates	Test tubes
Spirit lamp	Beakers
Autoclave	Conical flasks
	Siphon

Table 4: List of Drugs and Test samples

Drugs	Test samples
Oxytetracycline	Kabasura kudineer chooranam
Amphotericin B	Nilavembu kudineer chooranam

Table 5: List of Chemicals and Reagents

Chemicals	Reagents	Others
Ethanol	Bradford reagent	Whatman No.1 filter paper
Hydroalcohol	Mayer's reagent	Glass wool
Sodium bicarbonate	Benedict reagent	Distilled water
Hydrochloric acid		McFarland standard
Chloroform		Graph Pad Prism 6.0 software
Sulphuric acid		Test tubes
Ammonia		Conical flask
Ferric chloride		Dextrose
Sodium hydroxide		Agar
Absolute alcohol		
Magnesium sulfate		
CuSO ₄		
Antimycotic solution		

Table 6: List of Bacterial strains, Fungal strains and Medium used

Bacterial strains	Fungal strains	Medium used
<i>Pseudomonas aeruginosa</i> - 424	<i>Aspergillus niger</i>	Nutrient Agar medium
<i>Aeromonas hydrophila</i>	<i>Candida albicans</i>	Nutrient broth
<i>Proteus mirabilis</i>	<i>Cryptococcus</i>	Potato dextrose agar medium
<i>Staphylococcus aureus</i> -902	neoformans	
<i>Bacillus subtilis</i> -441	<i>Aspergillus fumigatus</i>	
<i>Enterococcus faecalis</i> -439		
<i>Streptococcus pyogenes</i> -1928		
<i>Klebsiella pneumoniae</i>		

FORMULATION PROFILE

NAME

Kabasura kudineer chooranam

Nilavembu kudineer chooranam

DESCRIPTION

Extraction is prepared by using hydroalcohol.

INDICATION

Antibacterial activity

Antifungal activity

METHODOLOGY

COLLECTION OF FORMULATION

Kabasura kudineer chooranam and Nilavembu kudineer chooranam was procured from Siddha Sasthiriya Medicine, Erode.

EXTRACTION OF THE FORMULATION

Soxhlet Extraction

Soxhlet extraction is a very useful tool for preparative purposes in which the analyte is concentrated from the matrix as a whole or separated from particular interfering substances. Sample preparation of environmental samples has been developed for decades using a wide variety of techniques. Solvent extraction of solid samples, which is commonly known as solid-liquid extraction (also referred to as leaching or Lixiviation in a more correct use of the physicochemical terminology), is one of the oldest methods for solid sample pretreatment. Conventional Soxhlet extraction remains as one of the most relevant techniques in the environmental extraction field.

EXTRACTION OF KABASURA KUDINEER CHOORANAM

Materials Required

Ethanol was purchased from Merck, USA. Whatman No.1 filter paper was purchased from Millipore, USA.

Procedure

1. Test sample (KKC) can be fresh or dried. It needs to be crushed, using a Mortar and Pestle to provide a greater surface area.
2. The test sample should be sufficient to fill the Porous cellulose thimble (in our experiments we use an average of 14 g of thyme in a 25- x 80-mm Thimble).
3. All equipment should be assembled. Build a rig using Stands and Clamps to support the extraction apparatus.
4. Following this, the Hydroalcohol is added to a Round bottom flask, which is attached to a Soxhlet extractor and condenser on an Isomantle.
5. The crushed Test sample (KKC) is loaded into the Thimble, which is placed inside the Soxhlet extractor.
6. The side arm is lagged with Glass wool.
7. The solvent is heated using the Isomantle and will begin to evaporate, moving through the apparatus to the condenser.
8. The condensate then drips into the reservoir containing the Thimble.
9. Once the level of solvent reaches the Siphon it pours back into the flask and the cycle begins again.
10. The process should run for a total of 4 hours.
11. Once the extraction set up, it can be left to run without direct supervision.
12. It is advised to not leave the equipment completely alone due to the mix of running water and an electrical appliance, so a technician should be made aware.
13. The equipment can be turned off.

EXTRACTION OF NILAVEMBU KUDINEER CHOORANAM

Materials Required

Ethanol was purchased from Merck, USA. Whatman No.1 filter paper was purchased from Millipore, USA.

Procedure

1. Test sample (NKC) can be fresh or dried. It needs to be crushed, using a Mortar and Pestle to provide a greater surface area.
2. The test sample should be sufficient to fill the Porous Cellulose Thimble (in our experiments we use an average of 14 g of thyme in a 25- x 80-mm Thimble).
3. All equipment should be assembled. Build a rig using Stands and Clamps to support the extraction apparatus.

4. Following this, the Hydroalcohol is added to a Round bottom flask, which is attached to a Soxhlet extractor and condenser on an Isomantle.
5. The crushed Test sample (NKC) is loaded into the Thimble, which is placed inside the Soxhlet extractor.
6. The side arm is lagged with Glass wool.
7. The solvent is heated using the Isomantle and will begin to evaporate, moving through the apparatus to the condenser.
8. The condensate then drips into the reservoir containing the Thimble.
9. Once the level of solvent reaches the Siphon it pours back into the flask and the cycle begins again.
10. The process should run for a total of 4 hours.
11. Once the extraction set up, it can be left to run without direct supervision.
12. It is advised to not leave the equipment completely alone due to the mix of running water and an electrical appliance, so a technician should be made aware.
13. The equipment can be turned off.

ANTIBACTERIAL ACTIVITY OF KABARUSA KUDINEER CHOORANAM, NILAVEMBU KUDINEERN CHOORANAM AND COMBINATION OF BOTH⁽¹⁰⁾⁽¹¹⁾

PRINCIPLE

The Antimicrobials present in the given sample were allowed to diffuse out into the medium and interact in a plate freshly seeded with the test organisms. The resulting zones of inhibition will be uniformly circular as there will be a confluent lawn of growth. The diameter of zone of inhibition can be measured in millimeters.

MATERIALS REQUIRED

(*Pseudomonas aeruginosa*- 424, *Aeromonas hydrophila*, *Proteus mirabilis*, *Staphylococcus aureus*-902, *Bacillus subtilis*-441, *Enterococcus faecalis*-439, *Streptococcus pyogenes*-1928 and *Klebsiella pneumoniae*) was purchased from MTCC, Chandihar, India. Nutrient Agar medium, Nutrient broth, Oxytetracycline solution was purchased from Himedia, India. Test samples, Petri-plates, Test tubes, Beakers, Conical flasks were from Borosil, India. Spirit lamp, Double distilled water.

AGAR- WELL DIFFUSION METHOD

Nutrient Agar Medium

The medium was prepared by dissolving 2.8 g of the commercially available Nutrient Agar Medium (HiMedia) in 100ml of distilled water. The dissolved medium was Autoclaved at 15 lbs pressure at 121°C for 15 minutes. The autoclaved medium was mixed well and poured onto 100mm petriplates (25-30ml/plate) while still molten.

Nutrient Broth

Nutrient Broth was prepared by dissolving 2.8 g of commercially available nutrient medium (HiMedia) in 100ml distilled water and boiled to dissolve the medium completely. The medium was dispensed as desired and sterilized by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

PROCEDURE

Petri plates containing 20 ml nutrient agar medium were seeded with 24 hr culture of bacterial strains were adjusted to 0.5 OD value according to McFarland standard, (*Pseudomonas aeruginosa*- 424, *Aeromonas hydrophila*, *Proteus mirabilis*, *Staphylococcus aureus*-902, *Bacillus subtilis*-441, *Enterococcus faecalis*-439, *Streptococcus pyogenes*-1928 and *Klebsiella pneumoniae*) wells were cut and concentration of sample KKC+NKC (500, 250, 100 and 50 µg/ml) was added. The plates were then incubated at 37°C for 24 hours. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the wells. Oxytetracycline was used as a positive control. The values were calculated using Graph Pad Prism 6.0 software (USA).

ANTIFUNGAL ACTIVITY OF KABARUSA KUDINEER CHOORANAM, NILAVEMBU KUDINEERN CHOORANAM AND COMBINATION OF BOTH⁽¹²⁾⁽¹³⁾

PRINCIPLE

The anti-fungal agent present in the given sample was allowed to diffuse out into the medium and interact in a plate freshly seeded with the test organisms. The resulting zones of inhibition will be uniformly circular as there will be a confluent lawn of growth. The diameter of zone of inhibition can be measured in millimeters.

MATERIALS REQUIRED

Potato dextrose agar medium, Amphotericin B, antimycotic solution, test samples, test tubes, beakers, conical flask, spirit lamp, double distilled water and petri-plates.

DISC DIFFUSION METHOD**Potato Dextrose Agar Medium**

The potato dextrose agar medium was prepared by dissolving 20 gm of potato infusion, 2 gm of dextrose and 1.5 gm of agar in 100ml of distilled water. The dissolved medium was autoclaved at 15 lbs pressure at 121°C for 15 minutes. The autoclaved medium was mixed well and poured onto 100mm Petri plates (25-30 ml/plate) while still molten.

PROCEDURE

Petri plates containing 20ml Potato dextrose agar medium was seeded with 72 hr culture of fungal strains (*Aspergillus niger*, *Candida albicans*, *Cryptococcus neoformans* and *Aspergillus fumigatus*) different concentration of sample KKC+NKC (500, 250, 100 and 50 µg/ml) was added. The plates were then incubated at 28°C for 72 hours. The Antifungal activity was assayed by measuring the diameter of the inhibition zone formed around the wells. Amphotericin B was used as a positive control. The values were calculated using Graph pad prism 6.0 software (USA).

RESULTS**PRELIMINARY PHYTOCHEMICAL ANALYSIS****PRELIMINARY PHYTOCHEMICAL ANALYSIS OF KABASURA KUDINEER CHOORANAM****Table 7: Results of Qualitative Phytochemical Screening Methods of KKC**

S.No.	Name of the Sample	Phytochemical compound	Result
1.	KKC	Resins	+
2.		Carboxylic acid	-
3.		Tannins	-
4.		Steroids	-
5.		Flavonoid	+
6.		Carbohydrates	+
7.		Glycosides	-
8.		Saponification	+
9.		Protein	+
10.		Phenol	-
11.		Biuret	-
12.		Soponin	-
13.		Gum	+
14.		Flavanoglycosides	-
15.		Alkaloids	-

PRELIMINARY PHYTOCHEMICAL ANALYSIS OF NILAVEMBU KUDINEER CHOORANAM**Table 8: Results of Qualitative Phytochemical Screening Methods of NKC**

S.No.	Name of the Sample	Phytochemical compound	Result
1.	NKC	Resins	+
2.		Carboxylic acid	-
3.		Tannins	+
4.		Steroids	-
5.		Flavonoid	+
6.		Carbohydrates	+
7.		Glycosides	-
8.		Saponification	+
9.		Protein	+
10.		Phenol	-
11.		Biuret	-
12.		Saponin	+
13.		Gum	+

14.	Flavonoglycosides	-
15.	Alkaloids	-

ANTIBACTERIAL ACTIVITY
ANTIBACTERIAL ACTIVITY OF KABASURA KUDINEER CHOORANAM

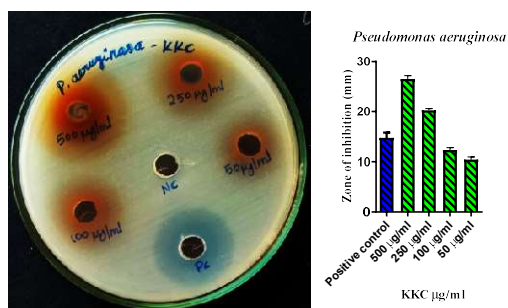


Fig 1: Effect of sample KKC against *Pseudomonas aeruginosa*.

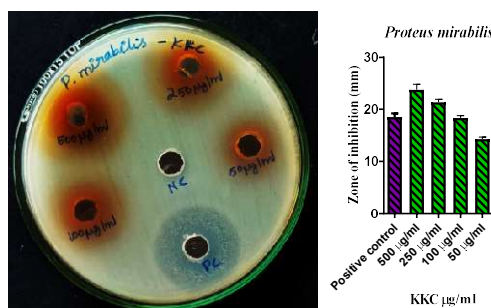


Fig 2: Effect of sample KKC against *Proteus mirabilis*.

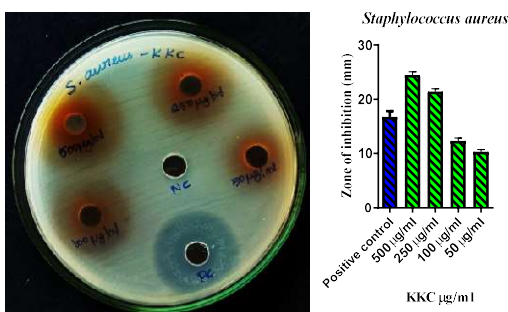


Fig 3: Effect of sample KKC against *Staphylococcus aureus*.

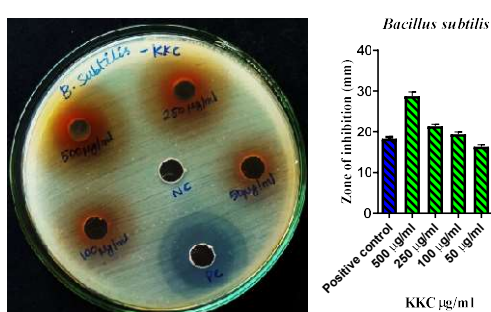


Fig 4: Effect of sample KKC against *Bacillus subtilis*.

Table 9: Means ± SD of zone of inhibition obtained by sample KKC against *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Staphylococcus aureus* and *Bacillus subtilis*.

S.No	Name of the test organism	Name of the test sample	Zone of inhibition (mm) Mean±SD				
			500 µg/ml	250 µg/ml	100 µg/ml	50 µg/ml	PC
1.	<i>Pseudomonas aeruginosa</i>	KKC	26.5±0.707	20.25±0.35	12.35±0.49	10.4±0.56	14.75±1.06
2.	<i>Proteus mirabilis</i>		23.75±1.06	21.4±0.56	18.35±0.49	14.3±0.42	18.5±0.70
3.	<i>Staphylococcus aureus</i>		24.45±0.63	21.4±0.56	12.35±0.49	10.3±0.42	16.75±1.06
4.	<i>Bacillus subtilis</i>		28.75±1.06	21.35±0.49	19.4±0.56	16.35±0.49	18.3±0.42

ANTIBACTERIAL ACTIVITY OF NILAVEMBU KUDINEER CHOORANAM

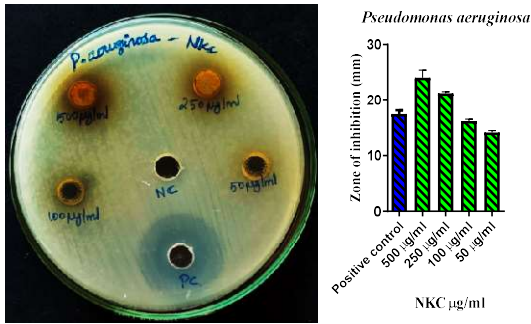


Fig 5: Effect of sample NKC against *Pseudomonas aeruginosa*.

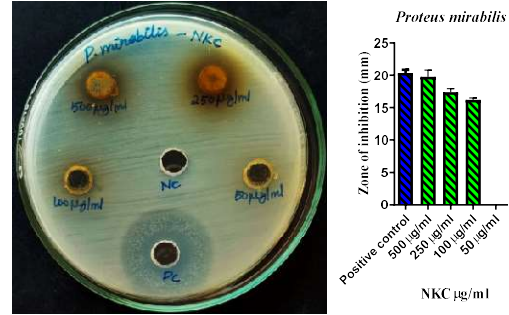


Fig 6: Effect of sample NKC against *Proteus mirabilis*.

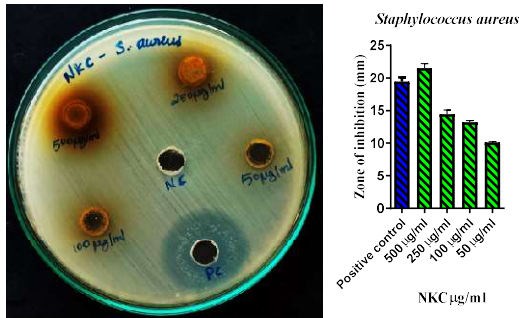


Fig 7: Effect of sample NKC against *Staphylococcus aureus*.

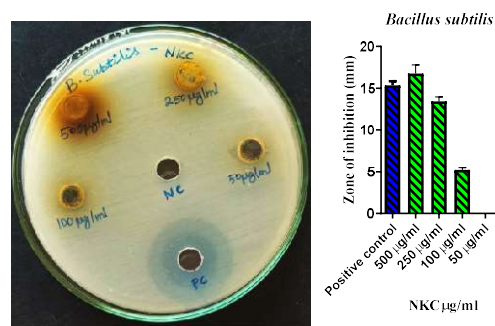


Fig 8: Effect of sample NKC against *Bacillus subtilis*.

Table 10: Means \pm SD of zone of inhibition obtained by sample NKC against *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Staphylococcus aureus* and *Bacillus subtilis*.

S. No	Name of the test organism	Name of the test sample	Zone of inhibition (mm) Mean \pm SD				
			500 µg/ml	250 µg/ml	100 µg/ml	50 µg/ml	PC
1.	<i>Pseudomonas aeruginosa</i>	NKC	24 \pm 1.41	21.2 \pm 0.28	16.25 \pm 0.35	14.2 \pm 0.28	17.5 \pm 0.707
2.	<i>Proteus mirabilis</i>		19.75 \pm 1.06	17.4 \pm 0.56	16.2 \pm 0.28	0	20.35 \pm 0.49
3.	<i>Staphylococcus aureus</i>		21.5 \pm 0.707	14.45 \pm 0.63	13.2 \pm 0.28	10.1 \pm 0.14	19.45 \pm 0.63
4.	<i>Bacillus subtilis</i>		16.75 \pm 1.06	13.4 \pm 0.56	5.2 \pm 0.28	0	15.35 \pm 0.49

ANTIBACTERIAL ACTIVITY OF KABASURA KUDINEER CHOORANAM AND NILAVEMBU KUDINEER CHOORANAM

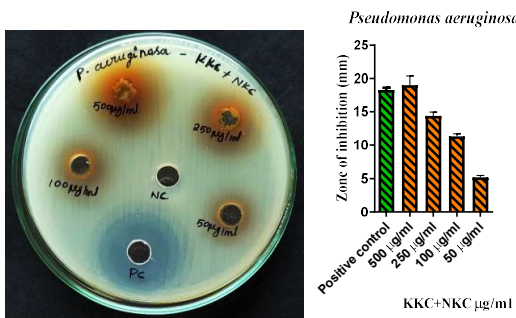


Fig 9: Effect of sample KKC+NKC against *Pseudomonas aeruginosa*.

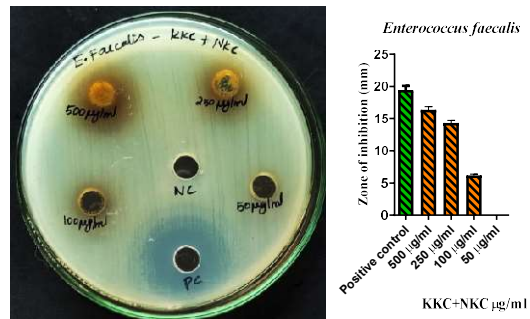


Fig 10: Effect of sample KKC+NKC against *Enterococcus faecalis*.

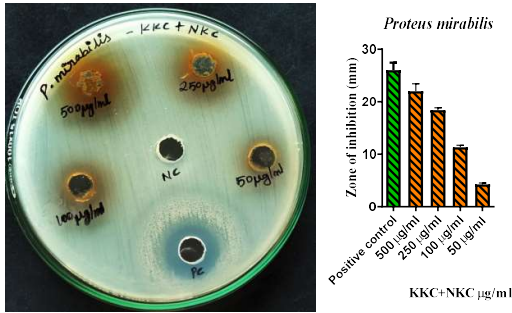


Fig 11: Effect of sample KKC+NKC against *Proteus mirabilis*.

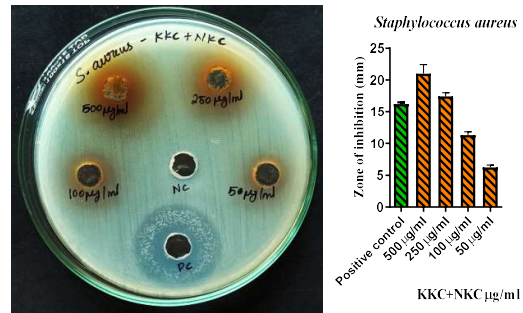


Fig 12: Effect of sample KKC+NKC against *Staphylococcus aureus*.

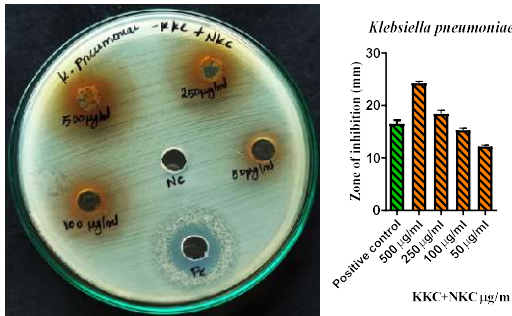


Fig 13: Effect of sample KKC+NKC against *Klebsiella pneumoniae*.

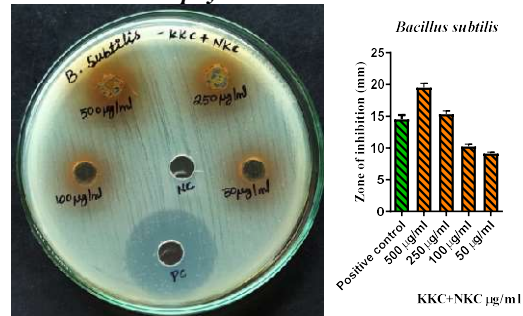


Fig 14: Effect of sample KKC+NKC against *Bacillus subtilis*.

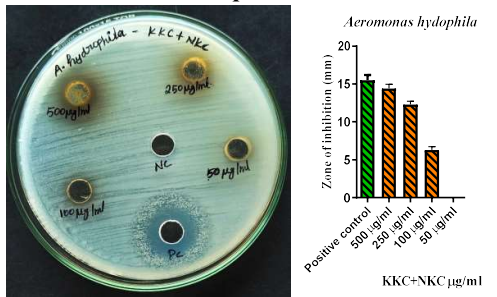


Fig 15: Effect of sample KKC+NKC against *Aeromonas hydrophila*.

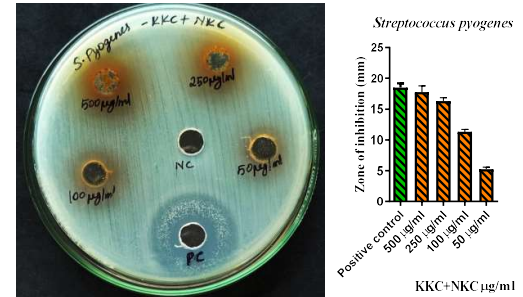


Fig 16: Effect of sample KKC+NKC against *Streptococcus pyogenes*.

Table 11: Means \pm SD of zone of inhibition obtained by sample KKC+NKC against *Pseudomonas aeruginosa*, *Aeromonas hydrophila*, *Proteus mirabilis*, *Staphylococcus aureus*, *Bacillus subtilis*, *Enterococcus faecalis*, *Streptococcus pyogenes* and *Klebsiella pneumoniae*.

S.No	Name of the test organism	Name of the test sample	Zone of inhibition (mm) Mean \pm SD				
			500 µg/ml	250 µg/ml	100 µg/ml	50 µg/ml	PC
1.	<i>Pseudomonas aeruginosa</i>	KKC+NKC	19 \pm 1.41	14.4 \pm 0.56	11.3 \pm 0.42	5.2 \pm 0.28	18.25 \pm 0.35
2.	<i>Proteus mirabilis</i>		22 \pm 1.41	18.35 \pm 0.49	11.3 \pm 0.42	4.2 \pm 0.28	26 \pm 1.41
3.	<i>Streptococcus pyogenes</i>		17.75 \pm 1.06	16.35 \pm 0.49	11.3 \pm 0.42	5.25 \pm 0.35	18 \pm 0.707
4.	<i>Bacillus subtilis</i>		19.5 \pm 0.70	15.35 \pm 0.49	10.25 \pm 0.35	9.15 \pm 0.21	14.5 \pm 0.707
5.	<i>Klebsiella pneumoniae</i>		24.25 \pm 0.35	18.45 \pm 0.63	15.3 \pm 0.42	12.2 \pm 0.28	16.5 \pm 0.707
6.	<i>Staphylococcus aureus</i>		21 \pm 1.41	17.4 \pm 0.56	11.35 \pm 0.49	6.25 \pm 35	16.2 \pm 0.28
7.	<i>Enterococcus faecalis</i>		16.35 \pm 0.49	14.3 \pm 0.42	6.15 \pm 0.21	0	19.45 \pm 0.63

8.	<i>Aeromonas hydrophila</i>	14.4±0.56	12.3±0.42	6.3±0.42	0	15.5±0.707
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SD – Standard Deviation, *Significance - $p < 0.05$

ANTIFUNGAL ACTIVITY

ANTIFUNGAL ACTIVITY OF KABASURA KUDINEER CHOORANAM

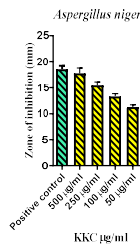
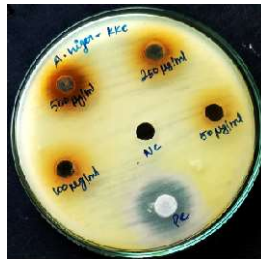


Fig 17: Effect of sample KKC against *Aspergillus niger*.

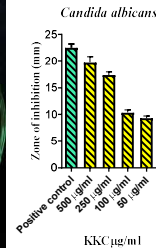
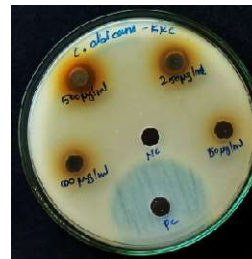


Fig 18: Effect of sample KKC against *Candida albicans*.

Table 12: SD± Means of zone of inhibition obtained by sample KKC and NKC against *Aspergillus niger*, *Candida albicans*, *Cryptococcus neoformans* and *Aspergillus fumigatus*.

S.NO	Name of the test organism	Name of the test sample	Zone of inhibition (mm) SD ± Mean				
			500 µg/ml	250 µg/ml	100 µg/ml	50 µg/ml	PC
1.	<i>Aspergillus niger</i>	KKC	17.75±1.06	15.45±0.63	13.35±0.49	11.3±0.42	18.5±0.70
2.	<i>Candida albicans</i>		19.75±1.06	17.4±0.56	10.35±0.49	9.3±0.42	22.5±0.707

ANTIFUNGAL ACTIVITY OF NILAVEMBU KUDINEER CHOORANAM

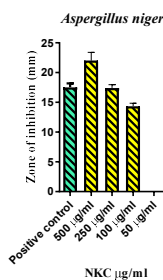
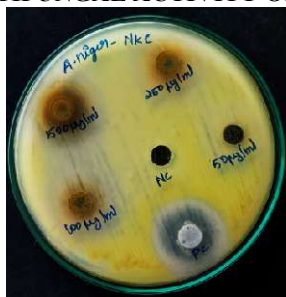


Fig 19: Effect of sample NKC against *Aspergillus niger*.

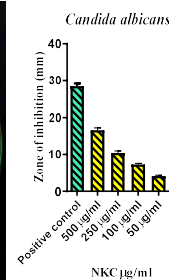


Fig 20: Effect of sample NKC against *Candida albicans*.

Table 13: SD± Means of zone of inhibition obtained by sample KKC and NKC against *Aspergillus niger*, *Candida albicans*, *Cryptococcus neoformans* and *Aspergillus fumigatus*.

S.NO	Name of the test organism	Name of the test sample	Zone of inhibition (mm) SD ± Mean				
			500 µg/ml	250 µg/ml	100 µg/ml	50 µg/ml	PC
3.	<i>Aspergillus niger</i>	NKC	22±1.41	17.4±0.56	14.35±0.49	0	17.8±0.707
4.	<i>Candida albicans</i>		16.5±0.70	10.4±0.56	7.25±0.35	4.15±0.21	28.5±0.7071

ANTIFUNGAL ACTIVITY OF KABASURA KUDINEER CHOORANAM AND NILAVEMBU KUDINEER CHOORANAM

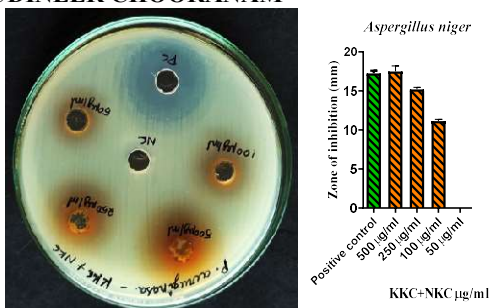


Fig 21: Effect of sample KKC+NKC against *Aspergillus niger*

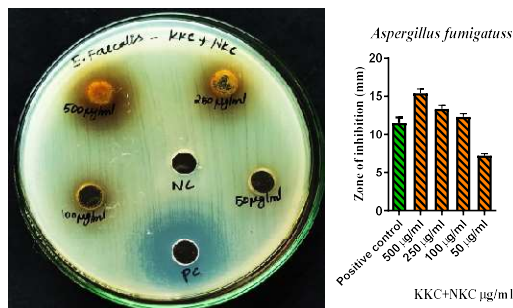


Fig 22: Effect of sample KKC+NKC against *Aspergillus fumigatus*.

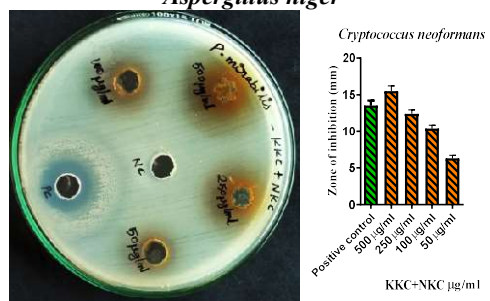


Fig 23: Effect of sample KKC+NKC against *Cryptococcus neoformans*.

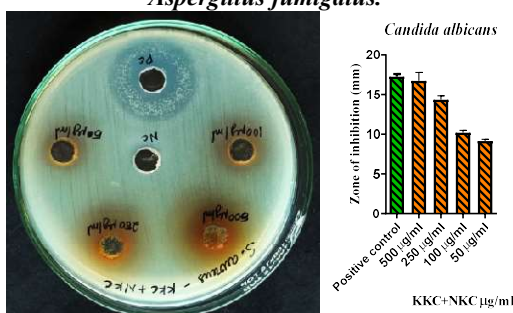


Fig 24: Effect of sample KKC+NKC against *Candida albicans*.

Table 14: SD± Means of zone of inhibition obtained by sample KKC+NKC against *Aspergillus niger*, *Candida albicans*, *Cryptococcus neoformans* and *Aspergillus fumigatus*.

S.No	Name of the test organism	Name of the test sample	Zone of inhibition (mm)SD ± Mean				
			500 µg/ml	250 µg/ml	100 µg/ml	50 µg/ml	PC
1.	<i>Aspergillus fumigatus</i>	KKC+NKC	15.4±0.56	13.35±0.49	12.3±0.42	7.2±0.28	11.5±0.707
2.	<i>Aspergillus niger</i>		17.5±0.70	15.2±0.28	11.15±0.21	0	17.25±0.35
3.	<i>Candida albicans</i>		16.75±1.06	14.35±0.49	10.2±0.28	9.15±0.21	17.25±0.35
4.	<i>Cryptococcus neoformans</i>		15.5±0.35	12.4±0.56	10.35±0.49	6.3±0.42	13.5±0.707

SD – Standard Deviation, *Significance - $p < 0.05$

DISCUSSION

In many developing countries, the rising cost of medicine and concerns regarding side effects have prompted researchers to explore polyherbal formulations. These formulations aim to harness the therapeutic potential of multiple herbs to address various health issues. As a result, traditional medical systems and herbal/herbo-mineral preparations have gained popularity due to their perceived effectiveness and often fewer reported side effects compared to some conventional medications.

Siddha medicine is indeed one of the oldest traditional systems of medicine in India. Kabasura kudineer choornam is a formulation commonly used in Siddha medicine, particularly for managing and preventing conditions like swine flu. Nilavembu kudineer choornam is indeed a traditional herbal formulation used in traditional Indian medicine. It contains nine ingredients, with Nilavembu (*Andrographis paniculata*) being one of them. This herb is known for its immunomodulating properties and has been recommended traditionally for managing dengue, chikungunya, and more recently, some people have explored its potential for alleviating

symptoms of COVID-19. However, it's important to note that while traditional remedies may have certain beneficial properties, their effectiveness in treating specific diseases like COVID-19 needs further scientific research and validation. Always consult healthcare professionals before using herbal remedies for serious conditions. It's believed that the phytochemical constituents present in this chooranam contribute to various health benefits, including anti-inflammatory, antipyretic, analgesic, antiviral, antibacterial, antifungal, antioxidant, hepato-protective, antidiabetic, anti-asthmatic, anti-tussive, immunomodulatory, antidiarrheal, and additional antioxidant activities⁽¹⁾⁽²⁾⁽³⁾.

Kabasura kudineer chooranam and Nilavembu kudineer chooranam was extracted by using Soxhlet extraction.

The results of the phytochemical screening of Kabasura kudineer chooranam showed the presence of carbohydrate, flavonoid, saponification, protein, gum, resin. Then the results of the phytochemical screening of Nilavembu kudineer chooranam showed the presence of resins, tannins, flavonoid, carbohydrate, saponification, protein, saponin, gum.

The Anti-bacterial activity of the extract was measured by observing bacterial free zones formed around the discs. The Anti-bacterial study was carried out for Kabasura kudineer chooranam and Nilavembu kudineer chooranam against different strain of bacteria (four Gram +ve such as *Streptococcus pyogenes*, *Bacillus subtilis*, *Staphylococcus aureus*, *Enterococcus faecalis* and four Gram -ve such as *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Klebsiella pneumoniae*, *Aeromonas hydrophila*) are known to cause infection in human and plants by agar-well diffusion method. Oxytetracycline was used as a positive control. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the wells. The zone of inhibition was observed for both Gram +ve and Gram -ve bacteria. The maximum zone of inhibition (21mm) was observed for *Staphylococcus aureus* for the Gram +ve organisms and Gram-ve organisms the maximum zone of inhibition (24.25mm) was found for *Klebsiella pneumoniae*. Thus Kabasura kudineer chooranam and Nilavembu kudineer chooranam was observed to have significant antibacterial activity. Kabasura kudineer chooranam and Nilavembu kudineer chooranam were found to have anti-bacterial activity with MIC of 500 µg/ml for both gram positive and gram negative organisms.

The Anti-fungal activity of the extract was measured by observing fungal free zones formed around the discs. The anti-fungal study was carried out for Kabasura kudineer chooranam and Nilavembu kudineer chooranam against different strain of fungi (*Aspergillus niger*, *Candida albicans*, *Cryptococcus neoformans* and *Aspergillus fumigatus*), that are known to cause infection in human and plants by disc diffusion method. Amphotericin B was used as a positive control. The antifungal activity was assayed by measuring the diameter of the inhibition zone formed around the wells. The maximum zone of inhibition (17.5mm) was found for *Aspergillus niger*. Kabasura kudineer chooranam and Nilavembu kudineer chooranam were found to have anti-fungal activity with MIC of 500 µg/ml.

SUMMARY

The present study supports that hydroalcoholic extract of KABASURA KUDINEER CHOORANAM AND NILAVEMBU KUDINEER CHOORANAM have significant antibacterial and antifungal activity. The investigation into the antibacterial and antifungal activity of Kabasura Kudineer Chooranam and Nilavembu Kudineer Chooranam has provided valuable insights into the potential therapeutic benefits of these traditional herbal formulations. This research aimed to explore and understand the extent of their antimicrobial properties, shedding light on their efficacy in combating bacterial and fungal infections.

The antimicrobial study of Kabasura Kudineer Chooranam and Nilavembu Kudineer Chooranam, as well as their combination, provides a comprehensive perspective on the potential therapeutic applications of these traditional herbal formulations. Kabasura Kudineer Chooranam, a polyherbal Siddha formulation, and Nilavembu Kudineer Chooranam, derived from the Nilavembu plant, have long been revered in traditional medicine for their purported health benefits, particularly in combating infections.

Kabasura Kudineer Chooranam, comprising a blend of herbs such as Kabasa (*Glycyrrhiza glabra*), Vilamichai (*Adhatoda vasica*), and Sirukanpeelai (*Andrographis paniculata*), has been historically utilized for its antimicrobial and immune-modulatory properties. Nilavembu Kudineer Chooranam, primarily composed of Nilavembu (*Andrographis paniculata*), is renowned for its antipyretic and anti-inflammatory attributes, making it a popular choice in treating fevers and infectious conditions.

The individual antimicrobial studies of Kabasura Kudineer Chooranam and Nilavembu Kudineer Chooranam unveil their respective abilities to inhibit the growth of various microorganisms. Kabasura Kudineer Chooranam demonstrates efficacy against bacteria and viruses, while Nilavembu Kudineer Chooranam exhibits antimicrobial activity with a focus on febrile illnesses. The nuanced differences in their antimicrobial spectra suggest a complementary approach in addressing diverse microbial challenges.

The combination of Kabasura Kudineer Chooranam and Nilavembu Kudineer Chooranam introduces a novel dimension to this study. The synergistic effects observed in the combined formulation hint at a potential enhancement of antimicrobial activity. The diverse array of bioactive compounds from the different herbs in each

formulation may act collaboratively, potentially broadening the spectrum of effectiveness against a wider range of pathogens. This synergy underscores the holistic nature of traditional herbal medicine, where multiple components work in concert to yield therapeutic benefits.

However, while these findings are promising, a more in-depth exploration is necessary to elucidate the precise mechanisms and interactions responsible for the observed antimicrobial effects. Identifying and understanding the specific bioactive compounds will contribute to the validation and optimization of these traditional formulations for modern therapeutic use. Furthermore, comprehensive studies involving in vivo experiments and clinical trials are essential to ascertain the safety, efficacy, and dosage considerations for practical application.

In the context of the global challenge posed by antimicrobial resistance, the antimicrobial study of Kabasura Kudineer Chooranam and Nilavembu Kudineer Chooranam, individually and in combination, suggests a potential avenue for alternative and complementary strategies in infectious disease management. Integrating the rich knowledge of traditional medicine with contemporary scientific methodologies could offer innovative solutions to address evolving healthcare needs.

CONCLUSION

In conclusion, the antimicrobial study of Kabasura Kudineer Chooranam and Nilavembu Kudineer Chooranam, along with their combination, not only highlights the potential therapeutic benefits of these traditional formulations but also advocates for a holistic and integrated approach to healthcare. As we navigate the complexities of infectious diseases and antibiotic resistance, traditional herbal remedies may contribute to a more diversified and resilient healthcare framework, marrying the wisdom of the past with the advancements of the present.

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