



## Antibacterial activity of *Hibiscus rosa sinensis* against human pathogenic microorganisms by varying different parameters.

Sanika Patil, Shraddha Yadav, Amol Jadhav\* and Bandu Pawar

Yashvantrao Chavan Institute of Science (Autonomous), Satara, Maharashtra, India, 415001 Corresponding

Author- Amol Jadhav

Email id – [amoljsadhav12@gmail.com](mailto:amoljsadhav12@gmail.com)

### Abstract -

Members of the Malvaceae family, including the spectacular trumpet-shaped flower and the *Hibiscus Rosasinensis*, grows green leaves. In traditional folk medicine, its therapeutic benefits for a number of illnesses have been reported. The objective of this study is to determine the antibacterial effect of ethanol extract of different concentrations, pH, and temperature against six human pathogenic microorganisms. Using the well diffusion method and various concentrations, pH levels, and temperatures, the ethanol extract of *H. rosasinensis* obtained through maceration was tested in vitro against six human pathogenic microorganisms, including *Staphylococcus*, *Salmonella*, *Shigella*, *Pseudomonas*, *Bacillus*, and *E. coli*. In a recent study, we found that flower extract of *H. rosa sinensis* had a strong antibacterial effect against *E. coli*, *Salmonella*, and *Bacillus* as compared to other human pathogenic microorganisms. *H. rosa sinensis* shows more zone of inhibition; therefore we concluded that *H. rosa sinensis* shows antibacterial activity against many human pathogenic microorganisms.

**Keywords:** Antibacterial activity, *Hibiscus*, Pathogens.

Received 20.11.2022

Revised 30.11.2022

Accepted 25.12.2022

### INTRODUCTION:

There are various medicinal plants in our nature that have been used since ancient times for the treatment of various diseases. In the modern period, these medicinal plants also have a crucial role in human life. Synthetic drugs which are prepared cause various side effects on human life but indigenous medicines are exceptional cases. Pharmaceutical care for the treatment of various diseases has mostly relied on drugs derived from plants [1]. In our ayurveda, there are various medicinal plants have been mentioned for the treatment of several incurable diseases. Therapies have become less effective as a result of antibiotic side effects and increased bacterial infection [2]. Extract of medicinal plants contains disease curing components that are used for the treatment of diseases with little or without any side effects and they also boost the immune system. Ingredients present in medicinal plants are also used for the preparation of drugs either pharmacopeia, non-pharmacopeia, or synthetic drug [3]. In many parts of the country, medicinal plants such as Neem, Tulsi, Turmeric, Aloe vera are used to treat common illnesses. They are used as home remedies.

Some therapeutic herbs have the ability to disinfect, which kills bacteria that cause sickness. They also prevent the development of pathogenic microorganisms which are the source of contagious disease. Experts in herbal medicine advise using herbs that have calming effects on the body, such as calming herbs [4]. *H. rosa sinensis* one of the evergreen shrubs which has been used as medicinal plants. Globally, *Hibiscus* trees are preferred by people due to their medicinal properties. Flowers of *H. rosa sinensis* are found in multiple colors but mainly in red color and sore in taste. It is high in vitamin C. There are varieties of *Hibiscus* flower having numerous names such as Rosella, and Jamaican sorrel, it is also known as Lalmbhari and Jaswandh [5]. Mainly calyx of *H. rosasinensis* is green in color but some times the calyx of *H. rosasinensis* are red in color. They are used and act as food colorants and dyes. Fresh calyx is used preparation of herbal drinks, jams, jellies, chocolates, cakes, etc [6]. *H. rosa sinensis* used as a medicinal plant in Chinese herbology. Its flower petal have historically been used to make black shoe polish or a woman's black hair dye in China. Flowers, leaves, and seeds of *H. rosa sinensis* are consumed by people [7]. *H. rosa sinensis* is used for the treatment of skin such as for controlling acne and breaking down dead skin and increasing cell turnover. A traditional remedy for burning sensation and skin conditions included the use of hibiscus plant leaves. Hibiscus extract maybe able to keep weight under control because of its polyphenol and flavonoids which may prevent fats from accumulating and help to keep weight under control [8].

A number of biological actions including anticomplementary, anti-diarrhetic, and antiphlogistic activity

have been demonstrated by prior research on *H.rosa sinensis* [1]. In addition, it has been claimed that the plant's flower has anti-spermatogenic, androgenic, antitumor, and anticonvulsant qualities [9]. Its leaves and blooms have also been discovered to increase hair development and speed ulcer healing [10]. *H.rosasinensis* is used to treat flu and cough, bronchitis, stomach pain, dysentery, and diarrhea and also for the regulation of menstruation and stimulation of blood circulation [11]. *H.rosasinensis* can treat *H.pylori* by showing antibacterial activity [2]. This plant has been used to isolate a variety of chemical compounds including cyanidin, quercetin, hentriacontane, calcium oxalate, thymine, riboflavin, niacin, and ascorbic acid [12]. The previous study shows that the extract of leaves and flowers of *H.rosa sinensis* has antibacterial activity.

Human pathogenic microorganisms play an important role in causing harmful diseases. In this case, *H.rosasinensis* is used as traditional medicine [13]. In the recent study, we aimed to study the antibacterial activity of *H.rosa sinensis* against various human pathogenic microorganisms by varying different parameters.

## MATERIAL AND METHODS

### 1. Collection of Sample *H.rosa Sinensis*:

Fresh red color flowers of *H.rosa sinensis* were collected from Y.C.I.S Satara college campus.

### 2. Extraction of Sample:

The flowers which were collected are rinsed with distilled water. After that, it is further ground in a mortar and pestle. The paste which is formed is kept with 70% ethanol for enrichment in a dark room for 72 hours. After enrichment sample is observed and filtered with the help of Whatman filter paper. The filtered sample was collected in a petri plate and kept for evaporation in the oven at 50°C. After evaporation metabolites from the plate were collected.

### 3. Testing for Antimicrobial activity:

The well diffusion method was used to observe the antibacterial activity of the prepared ethanol extract. 100 µg/ml sample was prepared. Mueller Hinton agar was used to check the antibacterial activity. Six different human pathogenic microorganisms i.e. (*E.coli*, *Staphylococcus*, *Salmonella*, *Shigella*, *Pseudomonas*, *Bacillus*) were used. The bacterial suspension spread on the plate and with the help of the well diffusion method, samples were inoculated and incubated for 24 hours at 37°C. After 24 hours the zone of inhibition was observed and results were recorded by measuring the diameter of the clear zone of inhibition [14].

### 4. Antibacterial activity is checked by applying various parameters:

#### Concentration

The effect of different concentrations of *H.rosa sinensis* against human pathogenic microorganisms. Four different concentrations of hibiscus extract were prepared such as 25 µg/ml, 50 µg/ml, 75 µg/ml, and 100 µg/ml. By using the well diffusion method samples were inoculated on a Mueller Hinton agar plate and kept for incubation at 37°C for 24 hrs. Antibacterial activity by varying various concentrations against several bacteria was done by observing the zone of inhibition.

#### pH

The antibacterial activity of *H.rosa sinensis* was checked by varying various pH such as 5, 6, 7, 8, and 9 against different human pathogenic microorganisms. Samples were inoculated on a Mueller Hinton agar plate and incubated for 37°C at 24 hrs and results were noted.

#### Temperature

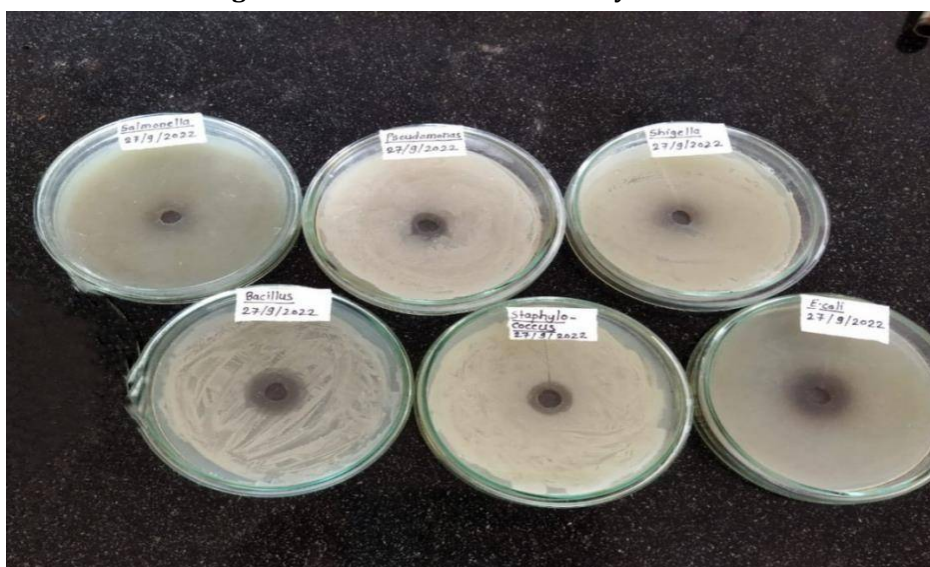
By varying different temperatures antibacterial activity of *H.rosa sinensis* against human pathogenic microorganisms were checked. Various temperature such as 25°C, 30°C, 35°C, and 40°C are used for incubation to observe the antibacterial activity.

## RESULT

### 1. Effect of Hibiscus abstract on different pathogens

In vitro antibacterial activity of *H.rosa sinensis*, ethanolic extract (100 µg/ml) against six different human pathogenic microorganisms (Table no.1). It shows that *Bacillus*, *E.coli*, *Salmonella* shows more zone of inhibition as compared to other human pathogenic microorganisms against the extract of *H.rosa sinensis* (Fig.1).

**Fig .1: Zone of inhibition shown by Hibiscus extract**



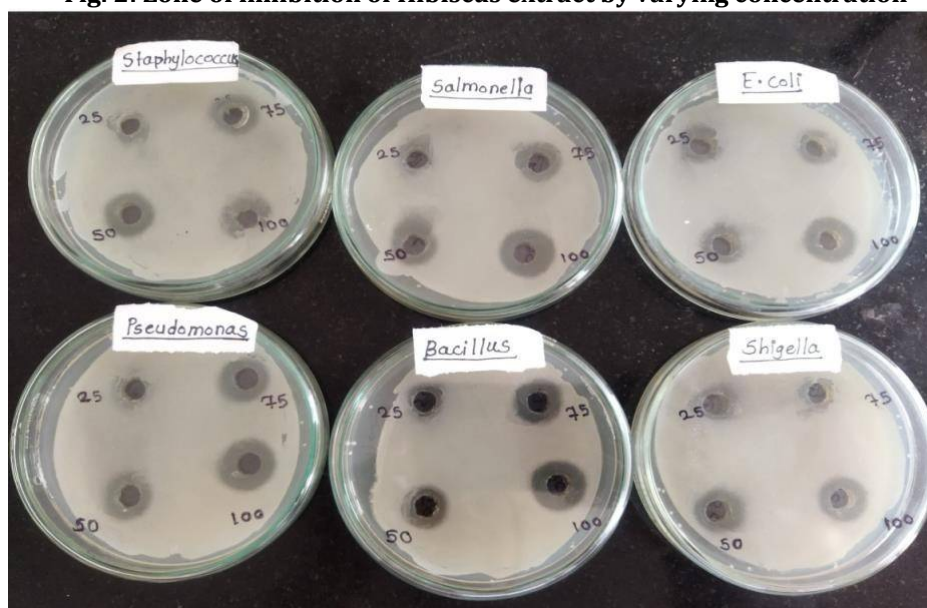
**Table 1: Diameter of the zone of inhibition shown hibiscus extract.**

Organisms	Diameter of zone
<i>Staphylococcus aureus</i>	16mm
<i>Salmonella typhirium</i>	15mm
<i>E.coli</i>	17mm
<i>Pseudomonas aeruginosa</i>	13mm
<i>Bacillus subtilis</i>	21mm
<i>Shigella dysenteriae</i>	No zone

## 2. Effect of different concentrations of Hibiscus extract on pathogens

A comparison of the different concentrations of Hibiscus extract against various human pathogenic microorganisms (Table no. 2) was studied. Hibiscus extract shows more antibacterial activity against *Bacillus*, *E.coli*, and *Salmonella* (Fig.2).

**Fig. 2: Zone of inhibition of Hibiscus extract by varying concentration**



**Table 2: Zone of inhibition of hibiscus extract by varying concentration.**

Organisms	Concentration of Samples			
	25µg	50µg	75µg	100µg
<i>Staphylococcus</i>	11mm	15mm	15mm	16mm
<i>Salmonella</i>	11mm	14mm	16mm	18mm
<i>E.coli</i>	12mm	15mm	13mm	20mm
<i>Pseudomonas</i>	10mm	13mm	15mm	17mm
<i>Bacillus</i>	11mm	13mm	15mm	18mm
<i>Shigella</i>	11mm	13mm	14mm	16mm

### 3. Effect of different pH of Hibiscus extract on pathogens

Extract of *H.rosa sinensis* shows antibacterial activity best at alkaline pH.As compared to other microorganisms *Bacillus*, *E.coli*, and *Salmonella* show a zone of inhibition at various pH such as 5,6,7,8,9 (Table no.3, Fig.3).



**Fig.3: Zone of inhibition shown by Hibiscus extract by varying different pH**

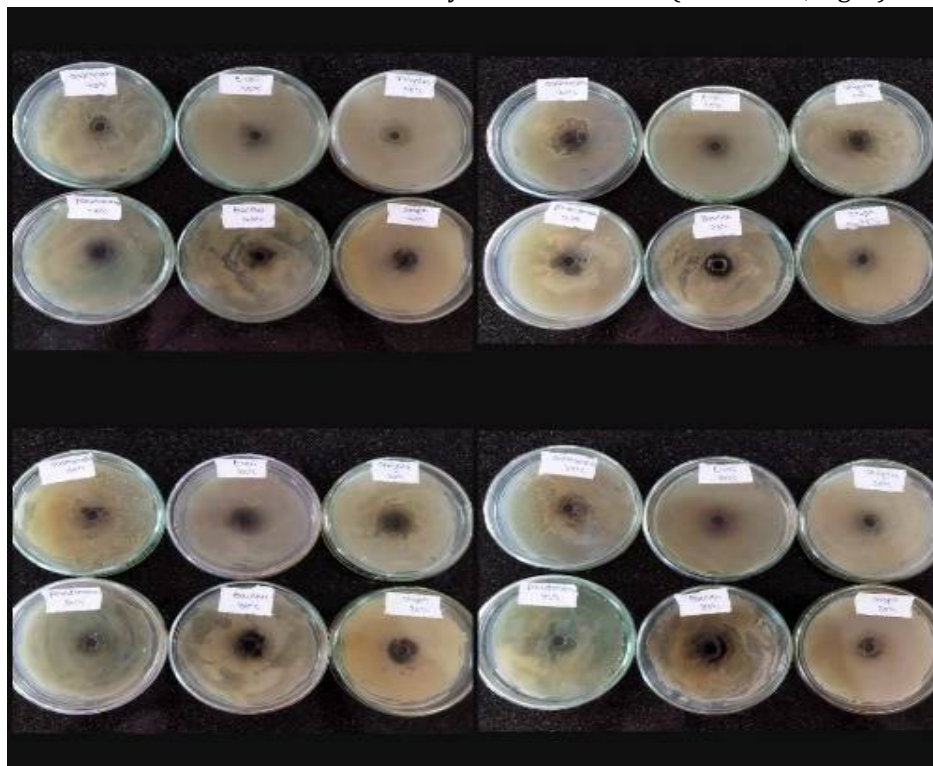
**Table 3: Zone of inhibition of hibiscus extract by varying pH**

Organisms	Different pH				
	5pH	6pH	7pH	8pH	9pH
<i>Staphylococcus</i>	10mm	11mm	13mm	15mm	10mm
<i>Salmonella</i>	No zone	12mm	12mm	10mm	11mm
<i>E.coli</i>	No zone	No zone	12mm	11mm	10mm
<i>Pseudomonas</i>	10mm	17mm	No zone	No zone	15mm
<i>Bacillus</i>	15mm	12mm	13mm	18mm	15mm
<i>Shigella</i>	No zone	No zone	11mm	11mm	No zone

### 4. Effect of different temperatures of Hibiscus extract on pathogens



Antibacterial activity of Hibiscus extracts by varying different temperatures (25°C, 30°C, 35°C, and 40°C). Hibiscus extract shows more antibacterial activity at 35°C and 40°C. (Table no. 4, Fig. 4)



**Fig. 4:Zone of inhibition shown by Hibiscus extract by varying different Temperatures**

**Table 4: Zone of inhibition of Hibiscus extract by varying Temperature**

Organisms	Different Temperatures			
	25°C	30°C	35°C	40°C
<i>Staphylococcus</i>	No zone	19mm	15mm	16mm
<i>Salmonella</i>	No zone	10mm	10mm	12mm
<i>E.coli</i>	No zone	No zone	12mm	10mm
<i>Pseudomonas</i>	12mm	No zone	15mm	11mm
<i>Bacillus</i>	18mm	16mm	15mm	10mm
<i>Shigella</i>	No zone	No zone	No zone	9mm

## CONCLUSION

The ethanolic extract of *H.rosa sinensis* has a significant effect on human pathogenic microorganisms. *Bacillus*, *E.coli*, and *Salmonella* were the pathogens with the highest activity. By varying Hibiscus extract at various concentrations, pH and temperature, the zone of inhibition also varies. Extract of *H.rosa sinensis* may be used as an effective antibacterial agent against different human pathogenic microorganisms. In this paper, we concluded that *Bacillus*, *E.coli*, and *Salmonella* show sensitive activity to Hibiscus.

## REFERENCES:

1. Arullappan, Sangeetha & Zakaria, Zubaidah & Basri, Dayang. (2009). Preliminary Screening of Antibacterial Activity Using Crude Extracts of *Hibiscus rosa sinensis*. *Tropical life sciences research*. 20.109-18.
2. Luong, Ngan & Tan, M.T. & Nguyen, Hoang & Thanh, D.T. & Linh, N.T.T. & Hoa, T.T.H. & My- Nuong, Nguyen & Tran, TrungHieu. (2021). Antibacterial activity of *Hibiscus rosa-sinensis* L. red flower against antibiotic-resistant strains of *Helicobacter pylori* and identification of the flower constituents. *Brazilian Journal of Medical and Biological Research*. 54. 10.1590/1414- 431x2020e10889.
3. Veeresham, Ciddi. (2012). Natural products derived from plants as a source of drugs. *Journal of advanced pharmaceutical technology & research*. 3. 200-1. 10.4103/2231-4040.104709.
4. Introduction and Importance of Medicinal Plants and Herbs | National Health Portal of India. (n.d.). Introduction and Importance of Medicinal Plants and Herbs | National Health Portal of India. Retrieved December 1, 2022, from <https://www.nhp.gov.in/introduction-and-importance-of->

5. ITIS - Report: Hibiscus rosa-sinensis. (n.d.). ITIS - Report: Hibiscus Rosa-sinensis. Retrieved December 1, 2022, from [https://www.itis.gov/servlet/SingleRpt?SingleRpt?search\\_topic=TSN&search\\_value=21616#null](https://www.itis.gov/servlet/SingleRpt?SingleRpt?search_topic=TSN&search_value=21616#null)
6. Al-Hashimi, Alaa. (2012). Antioxidant and antibacterial activities of Hibiscus sabdariffa L. extracts. 506-511. 10.5897/AJFS12.099.
7. Arulanantham R, Pathmanathan S, Ravimannan N, Niranjana K. Alternative culture media for bacterial growth using different formulation of protein sources. J Nat Prod Plant Resour. 2012;2(6):697-700.
8. Bodhare, D. A. (2022, June 3). Hibiscus: Uses, Benefits, Side Effects, And More! - PharmEasyBlog. PharmEasy Blog. Retrieved December 1, 2022, from <https://pharmeasy.in/blog/ayurveda-uses-benefits-side-effects-of-hibiscus/>
9. Sachdewa, Archana & Khemani, L. (2003). Effect of Hibiscus rosa sinensis Linn. ethanol flower extract on blood glucose and lipid profile in streptozotocin induced diabetes in rats. Journal of ethnopharmacology. 89. 61-6. 10.1016/S0378-8741(03)00230-7.
10. Ali, Özmen. (2010). Cytotoxicity of Hibiscus rosa-sinensis flower extract. Caryologia. 63. 157-161. 10.1080/00087114.2010.10589721.
11. Jadhav, V.M. & Thorat, R.M. & Sathe, V.J.. (2009). Traditional medicinal uses of Hibiscus rosa-sinensis. Journal of Pharmacy Research. 2.
12. Packiasamy, Dr. Ruban & Gajalakshmi, K. (2012). In vitro antibacterial activity of Hibiscus rosa-sinensis flower extract against human pathogens. Asian Pacific journal of tropical biomedicine. 2. 399-403. 10.1016/S2221-1691(12)60064-1.
13. Uddin, Borhan & Hossan, Tareq & Paul, Sudip & Ahmed, Tanjir & Nahar, Taslima & Ahmed, Sohel. (2010). Antibacterial activity of the ethanol extracts of Hibiscus rosa-sinensis leaves and flowers against clinical isolates of bacteria. Bangladesh Journal of Life Sciences; 2010. 22(2): 65-73. 22. 65-73.
14. Sirag, Nizar & Ahmed, Elhadi & Algaili, Algaili & Hassan, Hozeifa. (2013). Antibacterial Activity of Roselle (Hibiscus Sabdariffa L) Calyx Extract. 46. 1487 -1491.
15. Missoum, Asmaa. (2018). An update review on Hibiscus rosa sinensis phytochemistry and medicinal uses. Journal of Ayurvedic and Herbal Medicine. 4. 135-146. 10.31254/jahm.2018.4308.
16. Adhirajan, N & Kumar, T & Natesan, Shanmugasundaram & Babu, Mary. (2003). In vivo and in vitro evaluation of hair growth activity of Hibiscus Rosa -Sinensis Linn. Journal of ethnopharmacology. 88. 235-9. 10.1016/S0378-8741(03)00231-9.
17. S.A. Dahanukar, R.A. Kulkarni, N.N. Rege (2000). Pharmacology of medicinal plant and natural products. Indian Journal of Pharmacology; 32: S81-S118
18. Sobhy, Khadiga G. Abd Elaleem and Hagir G. Abd Elaleem, E. A. (n.d.). Potential Antibacterial Activity of Hibiscus rosasinensis Linn Flowers Extracts. Potential Antibacterial Activity of Hibiscus Rosasinensis Linn Flowers Extracts. Retrieved November 30, 2022, from <https://doi.org/10.20546/ijcmas.2017.604.132>
19. Karnwal, A. In vitro antibacterial activity of Hibiscus rosa sinensis, Chrysanthemum indicum, and Calendula officinalis flower extracts against Gram negative and Gram-positive food poisoning bacteria. ADV TRADIT MED (ADTM) 22, 607-619 (2022). <https://doi.org/10.1007/s13596-021-00562-x>.
20. Sikarwar, Mukesh & M.B. Patil. (2011). Antihyperlipidemic Effect of Ethanolic Extract of Hibiscus rosa sinensis Flowers in Hyperlipidemic Rats. RGUHS Journal of Pharmaceutical Sciences. 1. 117-122

#### CITATION OF THIS ARTICLE

S. Patil, S. Yadav, A. Jadhav and B. Pawar: Antibacterial activity of *Hibiscus rosa sinensis* against human pathogenic microorganisms by varying different parameters.. Bull. Env. Pharmacol. Life Sci., Spl Issue [1]: 2023:317-322.