



## Research Article

# EFFECT OF POTASSIUM IODIDE IN VARYING CONCENTRATIONS ON THE NORMAL GROWTH CURVE OF *SPOROTHRIXSCHENCKII* IN MYCELIALPHASE: AN INVITRO STUDY

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Received: November 27, 2018; Revised: December 26, 2018; Accepted: December 27, 2018; Published: December 30, 2018

**Abstract- Introduction and Objective:** Potassium iodide (KI) has been used as an effective drug for the treatment of sporotrichosis since centuries. Still it is one of the best drug in the treatment of infections caused by *Sporothrix schenckii*, but its exact mechanism of action is unknown. Hence, it was aimed to study the effect of increasing concentration of KI on the normal growth curve of *S. schenckii* in mycelial phase. **Material and Method:** A MTCC 1359 strains of *S. schenckii* was used to study the effect of KI on its growth. A filamentous stage of this thermally dimorphic fungus was inoculated in broth culture medium i.e., yeast nitrogen base (YNB) medium and incubated at 25 °C for 1 week. The broth culture was adjusted to turbidity of 90% transmittance at 530 nm in a colorimeter to obtain a master inoculum. A varying concentration of KI i.e., 0.05 gm%, 0.1 gm%, 0.2 gm%, 0.4 gm%, 0.8 gm%, 1.6 gm%, 3.2 gm%, 6.4 gm%, 12.8 gm% and blank (0.00gm%) was added in ten tubes containing 5ml of YNB. All the tubes were inoculated with 0.1 ml of master inoculum and then incubated at 25°C. Optical densities (ODs) of all the tubes were measured by taking readings of ODs at 530 nm of wavelength in a colorimeter for 34 days. **Result:** A significant change in ODs of all the phases of normal growth curve of mycelial form of *S. schenckii* was seen in increasing concentration of KI. The effect was more prominent in higher concentration of KI that too in exponential and decline phase of the normal growth curve than the lag and logarithmic phase. **Conclusion:** The study reveals an inhibitory effect of KI in its increasing concentration on normal growth curve of mycelial form of *S. schenckii*, but higher concentration had always shown the pronounced effect on all the phases of the normal growth curve. Hence, this study suggests that KI has an inhibitory activity on the growth of *S. schenckii*.

**Keywords-** Potassium iodide, *Sporothrix schenckii*, Mycelial phase, Varying concentration, Colorimeter

**Citation:** Shah R.K., et al., (2018) Effect of Potassium Iodide in Varying Concentrations on the Normal Growth Curve of *Sporothrixschenckii* in Mycelialphase: an in vitro Study. International Journal of Microbiology Research, ISSN: 0975-5276 & E-ISSN: 0975-9174, Volume 10, Issue 12, pp.-1422-1424.

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## Introduction

Sporotrichosis is a subacute or chronic pyogranulomatous mycosis of subcutaneous tissue and most often shows the clinical pictures of lymphangitis, which may have lymph node enlargement [1]. *Sporothrix schenckii* is a thermally dimorphic fungus which is prevalent throughout the globe, but endemic in Latin America, South Africa, India and Japan [2] The fungus generally lives their saprophytic life on decaying organic matter, soil, and sphagnum moss. The fungus is transmitted through traumatic inoculation of the organism to subcutaneous tissue of exposed area of the body mainly lower extremities, feet and hands. Disseminated forms of sporotrichosis like pulmonary, osseous and CNS are rare but these infections are mostly seen in immuno-compromised host via inhalation of conidial spores of *S. schenckii* and then their dissemination through blood to the different organs of the body. There have been the reports that the Infection can also be transmitted through the scratches of infected cats or digging animals like armadillos [3]. Treatment of sporotrichosis depends essentially on the clinical form of the disease, the host's immunological conditions and the different species of the genus *Sporothrix*. Potassium iodide (KI) is used in the treatment of sporotrichosis and is most common drug giving satisfactory results since 19th century but its exact mechanism of action remains unknown [4]. There have been different claims on the action of KI that it resolves granuloma with its enhanced proteolytic activity and increased phagocytosis [5,6]. There was a dearth of study on the action of KI on the growth of *S. schenckii*. Therefore, it was planned to design a study to see action of KI in its increasing concentration on the normal growth curve of *S. schenckii* in its filamentous form.

## Material and Methods

The study was designed and conducted at the Microbiology Dept. of a Medical College and Hospital in central India. A filamentous form of *S. schenckii* MTCC 1359 (a standard strain) was used in this study. A master inoculum was prepared by sub culturing filamentous form of *S. schenckii* in a 100 ml capacity screw capped bottle containing 50 ml of yeast nitrogen medium (YNB). The bottle was incubated at 25°C. On seventh day, turbidity was standardized by taking OD at 530 nm of wave length in a colorimeter by adjusting transmittance to 90% [7]. The master inoculum so prepared was further used in this study. A normal growth curve of mycelial phase of *S. schenckii* was estimated with the method similar to the method used by Shah et al to estimate the normal growth curve of *S. schenckii* [8]. In this test, the increasing concentration of KI i.e., 0.05 gm%, 0.1 gm%, 0.2 gm%, 0.4 gm%, 0.8 gm%, 1.6 gm%, 3.2 gm%, 6.4 gm%, 12.8 gm% and blank (0.00gm%) was added in ten tubes containing 5ml of YNB for separately. All the tubes were inoculated with 0.1 ml of master inoculum and then incubated at 25°C. Optical densities (ODs) of all the tubes were measured at 530 nm of wavelength daily in a colorimeter for 34 days with the methods similar to Bareja et al [9].

## Results

The normal growth curve of filamentous form of *S. schenckii* was estimated by taking daily readings of ODs for 34 days to plot a graph. On observation of the graph of normal growth curve, It had shown a lag phase from 0 to 2<sup>nd</sup> day with ODs measuring 0.01, a logarithmic phase from 3<sup>rd</sup> to 18<sup>th</sup> day with ODs measuring in range between 0.03 to 0.26, a stationary phase from 18<sup>th</sup> to 21<sup>st</sup> day with OD

Table-1 Readings of ODs in increasing concentration of KI on different days of the broth culture of mycelia phase of *S. schenckii*.

Days	0	2	4	6	8	10	12	14	16	18	20	22	24	26	28	30	32	34
Cons of KI	Optical Densities																	
0	0.01	0.01	0.04	0.09	0.12	0.18	0.21	0.22	0.24	0.26	0.26	0.25	0.23	0.22	0.21	0.19	0.18	0.17
0.05	0.01	0.02	0.04	0.1	0.14	0.19	0.16	0.23	0.25	0.25	0.23	0.23	0.21	0.2	0.2	0.18	0.17	0.17
0.1	0.01	0.02	0.04	0.1	0.13	0.15	0.18	0.25	0.26	0.25	0.23	0.2	0.2	0.18	0.19	0.17	0.16	0.16
0.2	0.01	0.03	0.05	0.09	0.12	0.15	0.26	0.27	0.26	0.25	0.25	0.2	0.18	0.18	0.18	0.16	0.16	0.16
0.4	0.01	0.04	0.05	0.12	0.17	0.2	0.24	0.23	0.23	0.23	0.22	0.22	0.2	0.17	0.17	0.16	0.15	0.14
0.8	0.01	0.03	0.04	0.1	0.16	0.18	0.17	0.23	0.23	0.23	0.23	0.21	0.2	0.17	0.17	0.14	0.14	0.14
1.6	0.01	0.02	0.03	0.07	0.14	0.16	0.11	0.18	0.19	0.18	0.2	0.2	0.16	0.14	0.14	0.13	0.12	0.12
3.2	0.01	0.01	0.02	0.04	0.08	0.1	0.12	0.18	0.18	0.18	0.18	0.19	0.17	0.12	0.11	0.11	0.11	0.11
6.4	0.01	0.01	0.02	0.01	0.05	0.08	0.01	0.03	0.04	0.03	0.03	0.04	0.03	0.03	0.03	0.03	0.03	0.03
12.8.	0.01	0.01	0.01	0.01	0.02	0.02	0.02	0.02	0.02	0.01	0.01	0.01	0	0	0	0	0	0

measuring 0.26 and a decline phase from 22<sup>nd</sup> to 34<sup>th</sup> day with ODs measuring in range between 0.25 to 0.17. The daily readings of ODs of all the ten tubes containing increasing concentration of KI ranging from 0.05 gm% to 12.8gm%, had shown the change in curves of different concentration of KI on different days in comparison to the ODs readings taken on different days of normal growth curve of *S. schenckii* in filamentous form.. All the phases of the growth curves of different concentration of KI had shown the effect, but the effect was more pronounced in stationary and decline phase with higher concentration of KI in comparison to the normal growth curve (blank).

Fig-1 Filamentous form of *S. schenckii*Fig-2 Measurement of ODs readings in broth culture of filamentous form of *S. schenckii* in increasing concentration of KI in a colorimeter.

### Discussion

Potassium iodide (KI) had been a drug of choice in the treatment of almost all form of sporotrichosis since early 19<sup>th</sup> century and most commonly it was used in the treatment of fixed and lymphocutaneous sporotrichosis [10]. In 1903, the use of KI in the treatment of sporotrichosis was suggested by Saboraud because of unavailability of antifungal drugs at that time. Iodide was routinely used for treatment of several infectious and non-infectious diseases during 19<sup>th</sup> century

[6]. But the exact mechanism of action of KI is not yet completely understood. Though, there was the several claims that KI had immuno-modulatory effect and was very much effective in destruction of granuloma with enhanced immune response i.e., neutrophil chemotaxis and phagocytosis of *Sporothrix* cells [11]. Thus, the present study was designed to estimate normal growth curve of the filamentous form of *S. schenckii* and the result of normal growth curve (i.e., blank) so obtained so was almost similar to the study by Shah *et al* [8]. Further, the effect of KI on the normal growth curve *S. schenckii* in different concentration of KI was also studied. The results had shown the action of KI in its varying concentration ranging from 0.05 gm % to 12.8 gm% in their all the growth phases in comparison to normal growth curve of *S. schenckii* in filamentous form but the effect was more prominent in higher concentration of KI that too in stationary and decline phase as shown in [Fig-1].

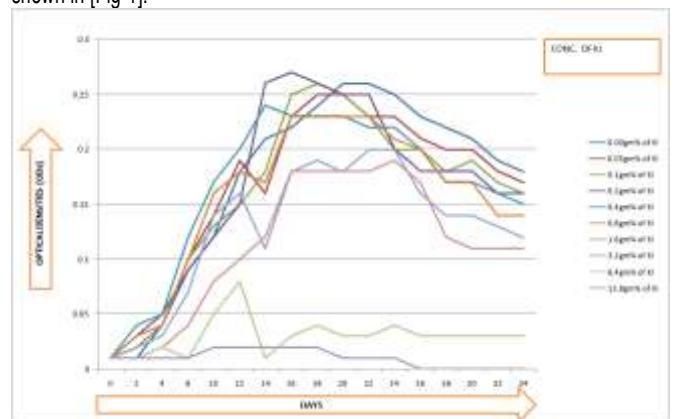


Fig-3 Growth curves showing effect of KI in its increasing concentration on different days.

On observation, the resultant graphs of broth culture in increasing concentration of KI ranging from 0.05 gm% to 12.8gm% had shown the significant in comparison to normal growth curve (a curve obtained in blank) from 2<sup>nd</sup> to 34<sup>th</sup> day. Where, there was first inclination in graphs of logarithmic phase of the growth curves in lower concentration of KI than higher concentration on 2<sup>nd</sup> day. This was followed with declination in all the concentration of potassium iodide except 0.4 gm% concentration of KI on 6<sup>th</sup> day and 0.05 gm% to 0.2gm% concentration of KI from 8<sup>th</sup> to 16<sup>th</sup> day of the logarithmic growth phase of the growth curve of filamentous form of *S. schenckii*. The action of KI was effective in all the growth phases of growth curve in comparison to the normal growth curve but it was more significant in higher concentration (i.e., 6.4-12.8gm %) of KI mainly in stationary and decline phase of growth curve which was similar to the study by Bareja *et al* [9].

### Conclusion

KI had been a drug of choice for fixed and lymphocutaneous sporotrichosis since the ancient time. The present study was designed to study the effect of KI on the normal growth curve of *S. schenckii* in filamentous form. The results of the study so obtained, had shown the effect of KI in different concentration ranging from 0.05gm% to 12.8gm% on all the growth phases in comparison to the normal growth curve of filamentous form of *S. schenckii* (i.e., blank). It had shown the effect of KI significantly on exponential phase (logarithmic phase) all the growth curves in

different concentration of KI, but the effect was more prominent on stationary and decline phase of the growth curves in different concentration of KI on filamentous form of *S. schenckii*. The higher concentration of KI had always shown the highest effect on all the phases of the growth curve in comparison to normal growth curve.

**Application of research:** Study the effective concentration of KI on the *in vitro* growth of filamentous form of *S. schenckii* in broth culture.

**Research Category:** Medical Microbiology

**Abbreviations:** KI: Potassium Iodide, YNB: Yeast nitrogen base medium, ODs: Optical densities, gm%: Gram percentage

**Acknowledgement / Funding:** Authors are thankful to RKDF Medical College Hospital and Research Centre, SRK University, Bhopal, 462026, Madhya Pradesh, India

**\*Research Guide or Chairperson of research:** Dr Hemant B Gadekar

University: SRK University, Bhopal, 462026, Madhya Pradesh

Research project name or number: Research station trials

**Author Contributions:** All authors equally contributed

**Author statement:** All authors read, reviewed, agreed and approved the final manuscript

**Conflict of Interest:** None declared

**Ethical approval:** This article does not contain any studies with human participants or animals performed by any of the authors.

**Ethical Committee Approval Number:** NIL

**Sample Collection:** A standard strain of MTCC *S.schenckii* was taken for this study

## References

- [1] Chander J. (2002) *Textbook of Medical Mycology*. Mehta publisher, second edition, 128-137.
- [2] Lopez-Romero E., Reyes-Montes M.R., Pérez-Torres A., Ruiz-Baca E., Villagómez-Castro J.C., Mora-Montes H.M., Flores-Carreón A. & Toriello C. (2011) *Future Microbiology*, 6,1, 85-102.
- [3] Kauffman C.A., Bustamante B., Champan S.W., Pappas P.G. (2007) *IDSA guidelines for management of sporotrichosis*, 45,1255-1265.
- [4] Orofino-Costa R., de Macedo P.M., Rodrigues A.M., Baernardes-Engemann A.R. (2017) *Anais brasileiros dermatologia*, 92, 5, 606-620.
- [5] Barrows M.B.L., Paes R.A., Schubach A.O. (2011) *Clinical Microbiology Reviews*, 24, 4, 633-654.
- [6] Rippon J. (1988) *Medical mycology-the pathogenic fungi and the pathogenic actinomycetes*, In J. Rippon (ed.), 3<sup>rd</sup> ed. W. B. Saunders Company, Philadelphia, PA, 325-352.
- [7] Shadomy S., Espinel-Ingroff A., Paxton L. (1978) *Laboratory Methods in Medical Mycology*, 4<sup>th</sup> edition, U.S. Department of health education and welfare, Public health services: Centre for disease control, Atlanta, Georgia, 173.
- [8] Shah R.K., Gadekar H.B., Jain V., Kumari S., Tripathi A., Gupta S. (2018) *International Journal of Current Microbiology and Applied Sciences*, 7,11, 1078-1084.
- [9] Bareja R., Grover P.S., Mehra S.K. (2015) *Scholars Journals of Applied Medical Sciences*, 2015,3,7D, 2676-2679.
- [10] Sterling J.B., Heymann W.R. (2000) *Journal of the American Academy of Dermatology*, 43, 4, 691-697.
- [11] Xue S.L., Li L. (2009) *Mycopathologia*, 167, 6, 355-356.