Original Article

# Response Of New Sesamol Analogue And Sesamum Indicum Seeds Extract Against Meningitis Triggering Pathogens

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## Abstract

A certain type of bacteria can cause an upper respiratory tract infection and then travel through the bloodstream to brain and can result in meningitis. Evidence over *S. aureus* and *E. coli* to trigger meningitis, antimicrobial potential of *Sesamum indicum*plant and sesamol intended present study to compare the antimicrobial response of new sesamol analogue (NSA) and *Sesamum indicum* seeds extract against meningitis triggering pathogens (MTP). Present study involved synthesis of NSA and preparation of sesame seeds extract (SSE). The NSA was characterized using ATR-IR, 1H-NMR and Mass spectrometric data. Both NSA and SSE were tested for antimicrobial potential against MTP, namely: *S. aureus* and *E. coli*. Among both, the NSA exhibited much higher antimicrobial potential when compared with SSE. Based on the resultant data present study concludes that NSA possess high inhibition potential against MTP and recommends that NSA should be further evaluated to support its clinical importance.

**Keywords**: Meningitis, sesamol, sesame, seeds, extract, and antibacterial

## INTRODUCTION

Bacterial meningitis (BM) is associated with high rates of unfavourable outcome and death. Atypical causative pathogens such as *S. aureus* and *E. coli* occur commonly and should be considered when starting empirical antimicrobial therapy for BM [1]. Human microbiota comprises 1:1 bacteria and human cells, little disturbance in such ratio may activate meningitis triggering pathogens (MTP)[2-5]. Evidence suggests role of *Staphylococcus aureus* (*S. aureus*) and *Escherichia coli* (*E. coli*) in BM [6]. Long-term administration of conventional antibiotics against various infections lead to multiple drug resistance and high mortality risk [7-17]. To combat such problem, one can use synthetic or phyto products. Literary evidence support plants products as an effective antibacterial agent [11-15], thereby can be used to combat MTP. Plants are known to possess numerous biological activities, so can be used in wide range of infections, disorders and diseases [18-40]. Several research highlighted numerous biological activities of plants [41-130]. A large number of studies revealed increase in biological potential of plant products together when combined with nanotechnology [131-162]. Literary facts suggest several synthetic moieties to possess high antimicrobial potential [163-176]. Numerous patents on plants and their products have been granted attributed to their higher biological activities [177-215]. Therefore, present study was intended to determine the antimicrobial potential of *Sesame* seeds extract (SSE) and new sesame analogue (NSA)derivative against meningitis triggering pathogens (MTP).

## MATERIAL AND METHODS

### Materials

The melting point of synthesized compound was determined using Thomas Hoover apparatus. IR spectra was recorded ATR-IR, Perkin Elmer, 1H-NMR on Bruker, DPX 300 and mass spectra on MASPEC (MSW/9629). Purity of synthesized compound was checked by TLC aluminium sheets – silica gel 60 F254 (0.2 mm). Plant material was collected from the

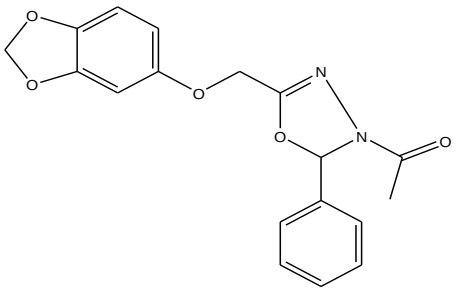
local market of Sungai Petani, Malaysia. Chemicals, and solvents were procured from the SD Fine, Sigma-Aldrich, and Merck Ltd.

### Preparation of Plant Extract

The SSE was prepared as per the standard procedure in the literature [15-25]. Briefly, sesame seeds free of decay were collected from the market of Sungai Petani, Kedah state, Malaysia and washed with tap water, followed by air drying, and macerated for 15 days using hydroalcoholic solvent (50:50). The mixture was filtered using double muslin cloth and a filter paper (Whatman No. 1) and the filtrate was dried to offer dark brown coloured SSE. The obtained SSE was stored at 4°C in refrigerator for further evaluation of its antimicrobial activity against MTP.

### Procedure for the synthesis of NSA

The synthesis of NSA was done as per the standard protocol with slight modifications [8, 172-175]. Briefly, a mixture of 2-(benzo[1,3]dioxol-5-yloxy)-N-benzylideneacetohydrazide(0.003 mol), was refluxed with acetic anhydride(10 mL)for 4 hours. After the reaction mixture attained room temperature, excess acetic anhydride was decomposed by adding water and the mixture was stirred for further 30 min. The separated product was filtered, washed with water, dried, and recrystallized from ethanol.



## RESPONSE OF SSE AND NSA AGAINST MTP

### *Preparation of bacterial culture*

Bacterial strains of *S.aureus* and *E.coli* were used for the antimicrobial experiment. The prepared stock culture of microorganism was maintained at 4°C. Subcultures were prepared by transferring loopful of microorganisms' colonies from stock cultures into the nutrient broth and incubated for 24 hours at 37˚C in the incubator. The broth turbidity indicated the microbial growth [11,12].

### *Well diffusion method*

The inhibitory potential of the prepared SSE and NSA against MTP was determined using well diffusion method-based zone of inhibition. The experimental protocol was followed as per the standard references with slight modifications [164-166]. Briefly, 20 µl of nutrient broth containing broth organism was poured into Muller Hinton agar plate, that was spread uniformly using L-shape rod. The wells were made on the agar medium with cork borer of 5 mm in diameter which was previously sterilized using autoclave at 121ºC for one hour. Each 50 µl of SSE and NSA were pipetted separately into the cup made on the agar plate. In the agar plate a few wells for SSE, NSA, standard and control. These plates contained the antibiotic streptomycin (standard) and tween 80 (control) solution for the purpose of comparison with the SSE and NSA. All the plates were incubated for 24 hours at 37ºC. The diameter of zone of inhibition around wells was measured in millimetres (mm) in triplicate and average values were calculated.

### Preliminary phytochemical screening of SSE

The SSE was subjected to preliminary phytochemical screening for the detection of various plant constituents. The prepared extract was screened for the presence of alkaloids, carbohydrates, flavonoids, glycosides, proteins, tannins, and phenols as per the procedure given in standard references [90,91].

## RESULTS

### Synthesis of SSE

Pale yellow crystals; Yield 65%; mp 172-173 ºC; ATR-IR: 3045, 2926, 1720, 1611, and 1259 $cm^{-1}$;$^{1}$H-NMR δ (ppm): 2.04 (3H, s, $CH_3$), 4.08 (2H, s, O-$CH_2$), 5.96 (2H, s, O-$CH_2$-O), 6.62 (1H, s, O-CH-N), 6.12-6.54 (3H, m, Ar-H), and 7.2 (5H, m, Ar-H); MS: m/z: 340 ($M^+$).

### Response of SSE and NSA against MTP

In present study, the prepared SSE and evaluated for their inhibitory potential against MTP such as *S. aureus* and *E. coli* using agar well diffusion for measurement of zone of inhibition. The prepared SSE and NSA were evaluated for their antimicrobial potential against bacterial strains of *S. aureus* and *E. coli*. The results so obtained are given in table 1.

**Table 1: Zone of inhibition of SSE and NSA**

| Compound | Microorganism | Zone of inhibition | | | |
|---|---|---|---|---|---|
| | | Reading 1 | Reading 2 | Reading 3 | Average Value |
| SSE | E. coli | 14 | 14 | 14 | 14 |
| | S. aureus | 12 | 12 | 12 | 12 |
| NSA | E. coli | 23 | 23 | 23 | 23 |
| | S. aureus | 21 | 13 | 21 | 19 |
| Streptomycin | E. coli | 24 | 24 | 24 | 24 |
| | S. aureus | 25 | 25 | 25 | 25 |
| Tween 80 | E. coli | - | - | - | - |
| | S. aureus | - | - | - | - |

### Preliminary phytochemical screening of SSE

The SSE was subjected to qualitative testing as per the procedure given in standard references [18,20]. The group of compounds identified in SSE are given in table 2.

**Table 2: Phytoconstituents of the SSE**

| S. No. | Tests | Phytoconstituents |
|---|---|---|
| 1 | Alkaloids | + |
| 2 | Flavonoids | + |
| 3 | Glycosides | + |
| 4 | Proteins | - |
| 5 | Tannins and Phenolic compounds | + |
| 6 | Sterols | + |

Where, (+) positive represent presence, and (-) negative represent absence

## DISCUSSION

The preliminary phytochemical screening of prepared SSE revealed presence of alkaloids, flavonoids, glycosides, sterols, tannins, and phenolic compounds. The IR,1H-NMR, and mass spectral data of NSA was found to be in agreement with its structure. The characteristic 1H-NMR signal at 6.62 for O-CH-N, appearance of IR band at 1259cm$^{-1}$ and m/z value at 340 supported the successful synthesis of NSA. These spectral values were also further confirmed based on the literary facts [185]. Evidence reports *S. aureus* and *E. coli*, to trigger meningitis. The growing incidences of microbial resistance towards conventional antibiotics raises the demand for evaluation of new antimicrobials [7-10]. Reports suggests use of *Sesamum indicum* plant in the treatment of various diseases and to possess strong antimicrobial potential. As per the literature available over different parts of *Sesamum indicum* plant and yet much more has to be explored for this plant. Hence, investigators of present study planned to evaluate the in-vitro inhibition potential of *Sesamum indicum* seeds extract against MTP (*S. aureus* and *E. coli*) using well diffusion method. The SSE was prepared using hydroalcoholic extract 50%. The prepared SSE was investigated for anti-microbial activity (using well diffusion method) and phytochemical screening. The SSE showed good inhibitory effect overgrowth of *S. aureus* and *E. coli*. On the other hand, the NSA was prepared by amination of ester derivative of eugenol, and when tested against MTP (*S. aureus* and *E. coli*) exhibited high inhibitory potential study revealed that synthetic derivative (NSA) possesses high potential when compared with SSE. The results of the present study were also supported by many other studies [187-246]. However, further preclinical, and clinical studies are required to further support the antimicrobial potential of NSA.

## CONCLUSION

The results of the present study over inhibitory potential of NSA and SSE against MTP, it is here by concluded that synthetic derivative NSA possess high antimicrobial potential against MTP especially *S. aureus* and *E. coli*. Present study recommends that highly potent NSA should be further evaluated based on the preclinical and clinical data.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## CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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