



Antibacterial Efficacy of Essential Oils from Four Spices against *Salmonella typhimurium*: Mathematical Modelling and Application in Enhancing Salad Cream Safety

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Abstract

The study was conducted to investigate the inhibitory effects of certain natural substances (finger root, clove, lemongrass, cardamom, and the combination of lemongrass with cardamom) against *Salmonella typhimurium*, a type of bacteria known to cause foodborne illnesses. The result showed that finger root, clove, lemongrass, cardamom, and the combination of lemongrass with cardamom exhibited strong inhibitory effects against *S. typhimurium*. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were evaluated. MIC values ranged from 0.049 to 0.781 µl/ml, and MBC values ranged from 0.049 to 6.250 µl/ml. Furthermore, the study aimed to develop mathematical models that accurately describe *S. typhimurium* survival in the presence of these essential oils. By understanding how the *S. typhimurium* respond to the oils over time, it was found that the mathematical models accurately described bacterial survival, with the modified Gompertz model fitting for finger root essential oil and the Weibull and modified Gompertz models suitable for the other oils. Additionally, the study sought to evaluate the practical viability of incorporating these essential oils into salad cream formulations, primarily aiming to assess their potential in reducing *S. typhimurium* counts and ensuring compliance with established quality standards. Specifically, the inclusion of finger root, clove, lemongrass, cardamom, and the combination of lemongrass with cardamom in salad cream formulations, maintained at a controlled temperature of 4 °C, yielded



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positive outcomes, meeting the required quality standards. Importantly, the presence of *S. typhimurium* was rendered undetectable, and an overall reduction in microbial count was observed when compared to cream formulations lacking these essential oils. This study effectively underscores the potential of the examined essential oils as natural antimicrobial agents suitable for incorporation in food products.

Introduction

The rise in consumer demand for convenient, ready-to-eat, and health-conscious food options has led to an increase in the consumption of fresh foods like salads, seafood, meat, and horticultural products.¹ Salads have gained substantial popularity among health-conscious individuals in contemporary times. These wholesome food preparations are deemed essential, accompanied by salad dressings, which play a pivotal role. Typically, salad dressings consist of a combination of eggs and oil, along with the potential inclusion of additional components such as vinegar, sugar, salt, mustard, pepper, or various spices.² Eggs, renowned for their exceptional nutritional value and economical nature, stand as one of nature's most formidable offerings. However, it is crucial to exercise caution when consuming salad cream derived from raw eggs, as it presents a substantial risk of contamination by *Salmonella spp.*³ Furthermore, the absence of heat during salad cream production further exacerbates the susceptibility of consumers to *Salmonella spp.* infection. Manifesting in symptoms such as diarrhea, stomach ache, nausea, vomiting, and even mortality, the ramifications of *Salmonella spp.* infection are dire.^{4,5,6} In light of escalating concerns regarding the safety of chemical preservatives employed to regulate microbial proliferation in food, there exists a burgeoning demand for natural alternatives to these agents. Consequently, the exploration of plant essential oils as a safer substitute for such additives has emerged as an area of intense focus.^{7,8}

The antimicrobial potential of essential oils derived from various spices and herbs, including finger root, clove, lemongrass, and cardamom, has been extensively demonstrated. However, the composition of essential oils in a species is influenced by numerous factors, including genotype-cultivar variations,⁹ the geographical region of cultivation,¹⁰ the plant parts selected for oil extraction,¹¹ and the specific extraction methods employed.¹² These

essential oils exhibit remarkable efficacy against a wide range of pathogenic microorganisms.^{13,14} According to Burt,¹⁵ essential oils can be defined as aromatic oily liquids obtained from plant materials. Steam distillation is the common means to derive essential oil for commercial production. The mentioned finger root, clove, lemongrass and cardamom, examples among others, have been incorporated in human dishes for many centuries, and have been claimed for a long time to show a benefit for health improvement in the human body, some even being used in medical treatment for patients.¹⁶ Focusing on the antibacterial properties of those essential oils, finger root essential oil has been found to inhibit pathogens such as *Listeria monocytogenes*, *Salmonella spp.*,¹⁷ *Citrobacter freundii*, *Escherichia coli*, *Klebsiella pneumonia* and *Serratia marcescens*.¹⁸ Clove essential oil has been discovered to possess antimicrobial properties that can restrain the growth of *Yersinia enterocolitica*, *E. coli*, *Pseudomonas aeruginosa*, *S. choleraesuis*, *Staphylococcus aureus*, *Bacillus cereus*, *L. monocytogenes* and *Enterococcus faecalis*.¹⁹ Lemongrass essential oil can inhibit *Bacillus cereus*, *B. subtilis*, *E. coli*, *S. aureus* and *K. pneumonia*.²⁰ Moreover, cardamom essential oil inhibits *B. subtilis*, *B. cereus*, *L. monocytogenes*, *S. aureus* and *S. typhimurium*.²¹ Through hydrophobic interaction, essential oils can penetrate the bacterial cellular membranes and mitochondria, which can disrupt their structure, increase permeability, and eventually lead to cell leakage and destruction.¹⁵

Mathematical models play a crucial role in elucidating and predicting the growth and survival patterns of microorganisms within specific experimental conditions. However, it is important to acknowledge that the data derived from mathematical modelling merely represents predictions. Consequently, it is essential to validate these predicted outcomes by comparing them with empirical experimental data. The utilization of mathematical models to

explicate and forecast microorganism growth and survival involves merging experimental data with mathematical frameworks.²² These models enable the anticipation of the future abundance or persistence of microorganisms, by employing mathematical equations that capture their respective growth or decline processes. Two fundamental types of equations are commonly employed for this purpose: linear equations and non-linear equations. The linear mathematical model, which encompasses the first-order kinetic model,^{23,24} serves as a popular tool for explaining and predicting microbial growth and reduction. Both Weibull^{25,26,27} and modified Gompertz^{28,29,30,31} models are indeed widely employed mathematical models that are used to describe the temporal changes in microbial growth. These models consider various factors and parameters to accurately represent the growth patterns of microorganisms over time. The first-order kinetic model, being linear in nature, finds common application in forecasting microbial growth and reduction as it pertains to the studied temperature conditions. The Weibull model, on the other hand, is frequently employed to predict microbial survival and the decline phase, while non-linear models enable the explanation and prediction of microbial reduction in processes involving heat and those that do not involve heat.

Henceforth, the present study aimed to assess the efficacy of commercially available essential oils derived from four distinct spices, namely finger root, clove, lemongrass, and cardamom, both individually and in combination, as potential antibacterial agents against *S. typhimurium*. The evaluation involved determining the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) of the essential oils obtained from these spices. In addition, mathematical models, specifically the first-order kinetic, Weibull, and modified Gompertz models, were developed to forecast the survival of *S. typhimurium* following treatment with the essential oils, with consideration given to a storage temperature of 4 °C. These models were designed to enhance understanding and facilitate the management of the impact of essential oils on microbial growth and survival throughout the storage duration. Furthermore, this investigation explored the potential utilization of these essential oils in salad cream.

Materials and methods

Preparation of bacterial cultures for inhibitory tests
The *Salmonella Typhimurium* strain (TISTR 1469) was preserved on tryptic soy agar (TSA) slants at a temperature of 5 °C. To prepare the active slants, a loopful of *S. typhimurium* was transferred into a tube containing 10 ml of tryptic soy broth (TSB), and subsequently incubated at 37 °C for a period of 18-24 hours. The turbidity of *S. typhimurium* suspensions was adjusted using sterile saline solution (0.85% NaCl) to match that of a McFarland Standard No. 0.5 (10⁸ CFU/ml), which had been modified following the methodology described by de Nova³² and the Clinical and Laboratory Standards Institute.³³

Preparation of Essential Oils from Spices For Inhibitory Tests

Four essential oils of spices were used in this study: finger root (*Boesenbergia pandurata* (Roxb.) Schltr.) and lemongrass (*Cymbopogon citratus* (DC. ex Nees) Stapf) from AP Operation Co., Ltd. (Chonburi, Thailand), and clove (*Syzygium aromaticum* (L.) Merr. & Perry) and cardamom (*Elettaria cardamum* Maton) from Lapis Tropical Spa Products (Bangkok, Thailand). The finger root samples utilized in this study are sourced as fresh rhizomes, carefully hand-harvested directly from local farmers. Within the operations of AP Operation Co., Ltd., a specialized quality control department is tasked with the diligent evaluation of both the size and quality of these finger root rhizomes. Subsequently, an extensive cleaning process is undertaken to prepare them for the extraction of essential oil using a precise steam distillation method. The resulting essential oil is then meticulously packaged in 100 ml containers. Similarly, in the case of lemongrass, this company obtains fresh leaves from farmers through meticulous hand-harvesting. A dedicated unit within the company is responsible for the selection of lemongrass leaves based on criteria such as size, colour, and overall quality. These carefully chosen lemongrass leaves then undergo a thorough cleaning process in preparation for essential oil extraction, utilizing the same meticulous steam distillation method. The resulting lemongrass essential oil is also elegantly packaged, this time in 10 ml containers.

Lapis Tropical Spa Products, located in Bangkok, Thailand, operates as an intermediary for the

procurement of essential oils, specifically clove and cardamom varieties. The clove essential oil samples are derived from freshly harvested flower buds, obtained from a specialized Indonesian enterprise engaged in essential oil extraction through the steam distillation method. The clove essential oil samples are carefully packaged in conveniently-sized 10 ml containers. In contrast, the cardamom essential oil samples are sourced from the desiccated seeds of cardamom plants cultivated and processed in India, also utilizing the steam distillation technique. These cardamom oil samples are presented in 10 ml containers.

The essential oils obtained from finger root, lemongrass, cloves, and cardamom possess a shelf life of two years. These oils can be suitably stored either refrigeration at 4 °C or storage at room temperature, maintained at 25 °C. All four essential oils were stored in amber glass bottles to protect them from UV light and reduce the potential degradation of the oils. The research study was

conducted during the period from 2020 to 2022, during which all experiments were meticulously initiated and successfully concluded prior to the expiration of the two-year shelf life for each of the four types of essential oils. However, further research will focus on laboratory-based essential oil extraction to determine the optimal conditions, with a consideration of important factors such as extraction methods, temperature, solvent-to-plant ratio, and the type of solvent.

The preparation of essential oils from spices for inhibitory tests involved 15 treatments, as detailed in Table 1. In the case of combined essential oils, this study will collect essential oil samples to be blended together based on the combination ratios specified in Table 1.

Subsequently, a volume of 50 µl from the blended essential oil was combined with 950 µl of 10% v/v DMSO, resulting in a final concentration of 50 µl/ml.

Table 1: Treatments, essential oils and their combination

Treatments	Essential oil samples	Combination ratio (w/w)	Essential oil sample volume combined with DMSO (µl)
1	Finger root oil	1	50
2	Clove oil	1	50
3	Lemongrass oil	1	50
4	Cardamom oil	1	50
5	Finger root oil + clove oil	1:1	50
6	Finger root oil + lemongrass oil	1:1	50
7	Finger root oil + cardamom oil	1:1	50
8	Clove oil + lemongrass oil	1:1	50
9	Clove oil + cardamom oil	1:1	50
10	Lemongrass oil + cardamom oil	1:1	50
11	Finger root oil + clove oil + lemongrass oil	1:1:1	50
12	Finger root oil + clove oil + cardamom oil	1:1:1	50
13	Clove oil + lemongrass oil + cardamom oil	1:1:1	50
14	Finger root oil + lemongrass oil + cardamom oil	1:1:1	50
15	Finger root oil + clove oil + lemongrass oil + cardamom oil	1:1:1:1	50

Screening of Essential Oils using Paper Disk Diffusion Technique

Using Lorian's method,³⁴ the entire surface of Mueller Hinton agar (MHA) was swabbed with a sterile

cotton swab that had been dipped in an inoculum suspension of *S. typhimurium*. After that, 6 mm diameter sterile paper discs infused with essential oils from 15 different treatments were placed on

the agar medium by slightly pressing them. The plates were incubated at 37 °C for 24 hours, and the diameters of the inhibition zones around the discs were measured using a digital caliper. To ensure accuracy, a negative control containing 10% v/v DMSO and a positive control containing 30 µg of tetracycline and 30 µg of chloramphenicol were added as well. The plates were left to rest at room temperature of 25 °C for 15 minutes before incubating to allow for excess pre-diffusion of essential oils.

Determination of Minimum Inhibitory Concentration

The determination of the minimum inhibitory concentration (MIC) of each essential oil sample against *S. typhimurium* was conducted following the outlined procedure (modification of the Clinical and Laboratory Standards Institute).³³ Initially, 0.5 ml of Mueller Hinton broth (MHB) was prepared in tubes, which were subsequently lightly sealed and sterilized in an incubator. Once sterilized, the tubes were allowed to cool, after which the essential oil was added at a concentration of 50 µl/ml, with a volume of 0.5 ml. The essential oils derived from spices were then subjected to a two-fold serial dilution method, resulting in the following dilution sequence of essential oil concentrations: 25, 12.50, 6.250, 3.125, 1.5625, 0.781, 0.390, 0.195, 0.098, 0.049 and 0.025 µl/ml, respectively. Following the preparation of *S. typhimurium*, 0.5 ml of the bacterial culture was added to each respective test tube. A positive control tube was included, containing *S. typhimurium* without any essential oil, while negative controls were prepared without the presence of bacteria. All test tubes were then incubated at a temperature of 37 °C for a duration of 24 hours. Subsequently, the turbidity of the culture media was observed to determine the minimum concentration of essential oils required to inhibit bacterial growth, in comparison to the control test tubes. The MIC refers to the lowest concentration of essential oil necessary to entirely inhibit the growth of microorganisms. This concentration is determined through the observation of the absence of turbidity, indicating the absence of visible growth. The MIC value is expressed in microliters per milliliter (µl/ml) as a unit of essential oil concentration.

Determination of Minimum Bactericidal Concentration at 37 °C and 4 °C

The determination of the minimal bactericidal concentration (MBC) was performed following a modified approach recommended by the Clinical and Laboratory Standards Institute.³³ The test tube samples showing no turbidity, as per the method described in the "Determination of minimum inhibitory concentration" section, were streaked onto TSA plates. The MBC of each essential oil was assessed at both 37 °C and 4 °C, representing the lowest concentration required to completely kill the microorganisms after a 24-hour incubation period. In this study, the MBC values are expressed in microliters per milliliter (µl/ml), serving as a unit for quantifying the concentration of essential oil.

Enumeration of Surviving *Salmonella typhimurium* Populations

Four single essential oils and combinations that showed the strongest inhibitory effect against *S. typhimurium* were selected and then prepared in MHB. MHB tubes containing essential oils at 1-, 4-, 16- and 64-fold the MBC were inoculated with a suspension of *S. typhimurium* including one positive control (*S. typhimurium* + MHB) to confirm that *S. typhimurium* could grow in MHB and one negative control (MHB) to ensure that the MHB was not contaminated. All samples in the tubes were incubated at 4 °C and stored under a controlled refrigerated temperature of 4 °C. The samples were analysed every 0.5 hr interval for 6 hr.

Mathematical Models of Survival Curves

To describe the survival of *S. typhimurium*, mathematical models of *S. typhimurium* survival curves were obtained by creating graphs of the interaction of surviving *S. typhimurium* populations and time. This study selected three inactivation models, namely the first-order kinetic, Weibull, and modified Gompertz models.

First-Order Kinetic Model

The first-order kinetic model is a linear model that can be used to predict microbial growth or reduction over time. It provides insights into the changes in the number of microorganisms at the experimental temperature.

$$N(t) = N_0 e^{-kt} \quad \dots(1)$$

where the variable $N(t)$ represents the quantity of microorganisms present at a given time, t (log CFU/ml), the variable N_0 represents the initial quantity of microorganisms at the start of the experiment or at time zero (log CFU/ml), k is the survival rate of microorganisms (1/hr) and t is time (hr).

Weibull Model

The Weibull model is a non-linear model used to predict survival or reduction of microorganisms.

$$\log(N) = \log(N_0) - (bt)^n \quad \dots(2)$$

where the variable N represents the quantity of microorganisms present at a given time, t (log CFU/ml), the variable, the variable N_0 represents the initial quantity of microorganisms at the start of the experiment or at time zero (log CFU/ml), b is the survival rate of microorganisms (1/hr), n is the shape of the survival curve and t is time (hr).³⁵

Modified Gompertz model

$$N = Me^{(\ln(N_0/M) e^{-At})} \quad \dots(3)$$

where the variable N represents the quantity of microorganisms present at a given time, t (log CFU/ml), the variable, the variable N_0 represents the initial quantity of microorganisms at the start of the experiment or at time zero (log CFU/ml), M is the number of microorganisms that enter the constant state (log CFU/ml), A is the survival rate of microorganisms (1/hr) and t is time (hr).²⁸

Quality of the Model

The precision of the models was evaluated based on the adjusted coefficient of determination (R^2) and root mean square error (RMSE), which were calculated as follows.

$$RMSE = \sqrt{\frac{\sum_{i=1}^n (\text{fitted value}_i - \text{experimental value}_i)^2}{n}} \quad \dots(4)$$

where n is the number of trials.

Preparation of Salad Cream

Salad cream samples were prepared using a Philips VHR 2061 homogenizer in Indonesia. The

ingredients used in the preparation included vinegar, sugar, sweetened condensed milk, ground pepper, salt, mustard, and eggs, constituting 14%, 19%, 8%, 1%, 1%, 1%, 47%, and 9% (w/w) of the salad cream, respectively. The ingredients were blended and homogenized for a duration of 2 minutes. Subsequently, the mixture was slowly incorporated into the vegetable oil phase while continuously stirring until a consistent and uniform texture was obtained.

Salmonella typhimurium Analysis of Salad Cream Samples

The analysis of *S. typhimurium* was modified based on the study conducted by Andrews.³⁶ Each sample of salad cream, weighing 25 g, was meticulously measured and placed in a sterile plastic stomacher bag containing 225 ml of sterile lactose broth. The mixture was then aseptically blended for 2 min. The pH of the mixture was assessed using test paper, and if necessary, adjusted to 6.8 ± 0.2 with sterile 1 N NaOH or 1 N HCl. The homogenized sample within the stomacher bag was incubated for 24 hours at 37 °C. After incubation, 1 ml of the mixture was transferred to 10 ml of Tetrathionate (TT) broth, vortexed, and then incubated at 37 °C for an additional 24 hours. A 3 mm loopful of the incubated TT broth was streaked onto xylose lysine desoxycholate (XLD) agar. Following incubation of the XLD agar at 37 °C for 24 hours, colonies suspected to be *Salmonella* were selected based on their characteristic appearance. *Salmonella* colonies typically exhibited pink or red coloration, with or without a black center on XLD agar. In cases where the colonies did not display these characteristics, colonies with a yellow colour and with or without a black center on XLD agar were selected. At least two colonies meeting these criteria were streaked onto tryptic soy agar (TSA) and incubated at 37 °C for 24 hours. Subsequently, biochemical tests were conducted to confirm the presence of *Salmonella*. These tests involved the utilization of sugars in triple sugar iron (TSI) agar, the production of the enzyme lysine decarboxylase in lysine iron agar (LIA), and the assessment of sulfur, indole, and motility in sulfide indole motility (SIM) medium. Following the biochemical tests, the cultures were further incubated at 37 °C for 24 hours before confirming the presence of *Salmonella* based on the biochemical results.

Microbiological Analysis of Salad Cream Samples

The microbiological analysis of salad cream samples was performed, utilizing a modified methodology based on the approach proposed by Maturin and Peeler.³⁷ After aseptically weighing 25 g of each salad cream sample into a sterile plastic stomacher, the sample was homogenized in 225 ml sterile peptone water (PW), beaten and blended for 1 min.

To prepare decimal dilutions of 10^{-2} , 10^{-3} , 10^{-4} , and subsequent dilutions, 1 ml of the previous dilution was transferred to 9 ml dilution tubes using a sterile automatic pipette. Each dilution (1 ml) was pipetted into plate count agar (PCA). Using a sterile glass spreader, the diluted sample was evenly spread over the entire plate's agar surface. The plates were then incubated for 24 hr at 37 °C.

Table 2: Samples of salad cream prepared for testing the survival of *Salmonella typhimurium*

Treatments	Type of salad cream
1	Salad cream without essential oils from spices and without <i>S. typhimurium</i>
2	Salad cream with finger root essential oil and with added <i>S. typhimurium</i>
3	Salad cream with clove essential oil and with added <i>S. typhimurium</i>
4	Salad cream with lemongrass essential oil and with added <i>S. typhimurium</i>
5	Salad cream with cardamom essential oil and with added <i>S. typhimurium</i>
6	Salad cream with combined essential oils giving the highest activity and with added <i>S. typhimurium</i>
7	Salad cream without essential oils from spices and with added <i>S. typhimurium</i>

Table 3: Salad cream samples prepared for testing the survival of microorganisms

Treatments	Salad cream samples
1	Salad cream with finger root essential oil
2	Salad cream with clove essential oil
3	Salad cream with lemongrass essential oil
4	Salad cream with cardamom essential oil
5	Salad cream with combined essential oils giving the highest activity
6	Salad cream without essential oils from spices

Analysis of *Salmonella typhimurium* In Salad Cream Incorporating Essential Oils from Spices

Seven salad cream samples (650 g each) were prepared as shown in Table 2. The concentrations of essential oil which exhibited the best antimicrobial activity to kill *S. typhimurium* at 4 °C and during the storage period (at temperature of 4 °C) were used for further study. Subsequently, samples were subjected to microbiological analysis to determine the presence of *S. typhimurium* populations. The analysis was conducted at the onset of storage, followed by daily assessments up to day 7.

Analysis of Microorganisms in Salad Cream Incorporating Essential Oils from Spices

Six salad cream samples (700 g each) which were prepared as shown in Table 3 – salad cream from

a local market and supermarket, salad cream from Mae Hia market (shop 1), salad cream from Mae Hia market (shop 2) and salad cream from the Royal Project – were used to determine the number of microorganisms. Moreover, all salad cream samples were packed, stored at a temperature of 4 °C and analysed on days 0, 7 and 15 for the survival of *S. typhimurium* and every 3 days from 0 to 15 days for total microorganisms and pH value.

Sensory Evaluation of Salad Cream Incorporating Essential Oils from Spices

A sensory laboratory was utilized to conduct the sensory analysis, wherein a trained sensory panel comprising of 30 assessors (18 women and 12 men aged between 20 and 32 years, all non-smokers) affiliated with the Faculty of Agro-Industry at

Chiang Mai University. This panel assessed nine coded samples of salad cream, rating them on a 9-point hedonic scale based on attributes such as appearance, colour, aroma, and texture.

Table 4: Antibacterial efficacy of spice-derived essential oils against *Salmonella typhimurium* via paper disk diffusion

Essential oil samples (50 µl/ml)	Diameter of inhibition zone (mm)
Finger root oil	10.83 ± 0.63 ^a
Clove oil	22.00 ± 0.43 ⁱ
Lemongrass oil	17.33 ± 0.61 ^g
Cardamom oil	14.78 ± 0.45 ^{de}
Finger root oil + clove oil	17.33 ± 0.66 ^g
Finger root oil + lemongrass oil	14.11 ± 0.93 ^c
Finger root oil + cardamom oil	13.55 ± 0.50 ^b
Clove oil + lemongrass oil	19.39 ± 0.78 ^h
Clove oil + cardamom oil	17.44 ± 0.46 ^g
Lemongrass oil + cardamom oil	15.00 ± 0.71 ^e
Finger root oil + clove oil + lemongrass oil	14.89 ± 0.78 ^e
Finger root oil + clove oil + cardamom oil	15.33 ± 0.43 ^e
Clove oil + lemongrass oil + cardamom oil	16.56 ± 0.46 ^f
Finger root oil + lemongrass oil + cardamom oil	14.33 ± 0.43 ^{cd}
Finger root oil + clove oil + lemongrass oil + cardamom oil	14.94 ± 0.30 ^e
Tetracycline	29.78 ± 0.44 ^j
Chloramphenicol	33.89 ± 0.33 ^k

Note: Tests used 6 mm diameter discs. Values are the average ± standard deviation. Statistically significant differences within the same column are denoted by different lowercase letters in superscript, with a significance level set at $p \leq 0.05$.

Expression of Results and Statistical Analysis

Each experiment was conducted at least three times to ensure accuracy and reproducibility of results. The data obtained were subjected to statistical analysis using one-way ANOVA, and mean values were compared using Duncan's mean comparison test. The statistical analyses were performed using SPSS 17.0 for Windows, and a significance level of 5% was set for the analyses.

Results and Discussion

Test of Ability of Essential Oils from Spices to Inhibit *Salmonella typhimurium*

The results for the ability of essential oils from spices to inhibit *S. typhimurium* at a concentration of 50 µl/ml using the paper disk diffusion technique on MHA indicated that essential oils from all four types of spices and their combinations exhibited good inhibitory ability against *S. typhimurium*. According to the findings presented in Table 4, the essential oil derived from clove exhibited the highest potency

in inhibiting *S. typhimurium* compared to those from lemongrass, cardamom, and finger root, in descending order. The average diameter of the clear zone was 22.00 ± 0.50, 17.36 ± 0.69, 14.80 ± 0.45 and 10.80 ± 0.76 mm, respectively. As shown in Table 4, the combined clove and lemongrass essential oils showed the greatest inhibitory ability (19.39 mm). Furthermore, when essential oil from clove was combined with others with poor ability, a synergistic effect could be observed; for example, clove oil combined with finger root oil, clove oil combined with cardamom oil and clove oil combined with lemongrass oil.

Antimicrobial Activity of Essential Oils from Spices against *Salmonella typhimurium* At 37 °C: Determining Minimum Inhibitory Concentration and Minimal Bactericidal Concentration

The results for the MIC and MBC of spice essential oils against *S. typhimurium* using a two-fold dilution technique are shown in Table 5. The best result

was obtained for lemongrass essential oil, the second best for cardamom, clove and finger root oils. The MIC were 0.049, 0.390, 0.390 and 0.781 $\mu\text{l/ml}$ and MBC were 0.049, 0.781, 0.781 and 0.781 $\mu\text{l/ml}$, respectively. For combined essential oils, lemongrass oil combined with cardamom oil showed the greatest activity, with an MIC of 0.195 $\mu\text{l/ml}$ and MBC of 0.195 $\mu\text{l/ml}$.

The MBC is either greater than or equal to the MIC. This is because the MBC specifies the minimum concentration of the essential oil needed to completely eradicate the microorganism, while the MIC represents a preliminary value used to

determine the essential oil concentration required to inhibit bacterial growth. The present result, shown in Table 5, showed that the MBC obtained is greater than or equal to the MIC, which is in agreement with the results of Snoussi,²¹ who studied the effect of cardamom essential oil on bacteria; their results revealed that the MBC value was greater than the MIC value. Also, Raybaudi-Massilia³⁸ studied the antimicrobial activity of essential oils (lemongrass, cinnamon, geraniol, palmarosa, clove, benzaldehyde) against *S. Enteritidis*, *E. coli* and *Listeria innocua* using the agar broth dilution technique; they also revealed that the MBC was greater than the MIC.

Table 5: Antimicrobial activity of essential oils from spices: minimum inhibitory concentration and maximum bactericidal concentration

Essential oil samples	MIC ($\mu\text{l/ml}$)	MBC ($\mu\text{l/ml}$)
Finger root oil	0.781 \pm 0.00 ^d	0.781 \pm 0.00 ^d
Clove oil	0.390 \pm 0.00 ^c	0.781 \pm 0.00 ^d
Lemongrass oil	0.049 \pm 0.00 ^a	0.049 \pm 0.00 ^a
Cardamom oil	0.390 \pm 0.00 ^c	0.781 \pm 0.00 ^d
Finger root oil + clove oil	0.781 \pm 0.00 ^d	0.781 \pm 0.00 ^d
Finger root oil + lemongrass oil	0.195 \pm 0.00 ^b	0.390 \pm 0.00 ^c
Finger root oil + cardamom oil	0.781 \pm 0.00 ^d	0.781 \pm 0.00 ^d
Clove oil + lemongrass oil	0.195 \pm 0.00 ^b	0.390 \pm 0.00 ^c
Clove oil + cardamom oil	0.390 \pm 0.00 ^c	0.781 \pm 0.00 ^d
Lemongrass oil + cardamom oil	0.195 \pm 0.00 ^b	0.195 \pm 0.00 ^b
Finger root oil + clove oil + lemongrass oil	0.390 \pm 0.00 ^c	0.781 \pm 0.00 ^d
Finger root oil + clove oil + cardamom oil	0.390 \pm 0.00 ^c	0.781 \pm 0.00 ^d
Clove oil + lemongrass oil + cardamom oil	0.390 \pm 0.00 ^c	0.390 \pm 0.00 ^c
Finger root oil + lemongrass oil + cardamom oil	0.390 \pm 0.00 ^c	0.390 \pm 0.00 ^c
Finger root oil + clove oil + lemongrass oil + cardamom oil	0.390 \pm 0.00 ^c	0.781 \pm 0.00 ^d

Note: Values are the average \pm standard deviation. Statistically significant differences within the same column are denoted by different lowercase letters in superscript, with a significance level set at $p \leq 0.05$.

Essential oils from spices can inhibit *S. typhimurium* because of the compounds they contain: clove essential oil has eugenol as its main constituent (70–80%)³⁹, finger root essential oil has 1,8-cineole and geraniol as its main constituents,⁴⁰ lemongrass essential oil has α -citral and β -citral as its main constituents⁴¹ and cardamom essential oil had α -terpinyl acetate and 1,8-cineole as its main

effective constituents.²¹ These compounds break the bacterial cell wall, leading to cell death.¹⁵

Minimal bactericidal concentration of essential oils from spices against *Salmonella typhimurium* at 4 °C As shown in Table 6, the essential oil for which the lowest concentration was required to kill *S. typhimurium* was that obtained from lemongrass,

with an MBC of 0.781 µl/ml. The clove, finger root and lemongrass oils combined with cardamom oil had the same MBC of 3.125 µl/ml. The essential oil with the highest concentration required to kill *S. typhimurium* was that from cardamom, with the highest MBC of 6.250 µl/ml.

Table 6: Minimum bactericidal concentration of essential oils from spices at 4 °C

Essential oil samples	MBC (µl/ml)
Finger root oil	3.125 ± 0.00 ^b
Clove oil	3.125 ± 0.00 ^b
Lemongrass oil	0.781 ± 0.00 ^a
Cardamom oil	6.250 ± 0.00 ^c
Lemongrass oil + cardamom oil	3.125 ± 0.00 ^b

Note: Values are the average ± standard deviation. Statistically significant differences within the same column are denoted by different lowercase letters in superscript, with a significance level set at $p \leq 0.05$.

The MBC of essential oils from spices was found to be greater at 4°C compared to 37 °C, which is consistent with the experiments of Govaris⁴² who evaluated the inhibitory activity of oregano essential oil at concentrations of 0.6% v/v and 0.9% v/v against *S. Enteritidis* in minced sheep meat during storage for 12 days at 4 °C and 10 °C. The results showed that oregano essential oil was more effective at inhibiting the bacteria in minced sheep meat at 10°C compared to 4 °C, with concentrations of 6% v/v and 0.9% v/v proving to be the most effective. In addition to investigating the inhibitory activity of essential oils, researchers have also explored the inhibitory effects of the active compounds found in these oils. Belda-Galbis⁴³ studied the ability of citral in essential oil from lemongrass to inhibit *E. coli* at 15, 30 and 37 °C and *Listeria innocua* at 8, 15, 30 and 37 °C. The results showed that the MIC of citral against *E. coli* at 30 and 37 °C was 0.325 µl/ml, but at 15 °C it decreased to 3.000 µl/ml. As for the inhibitory activity against *L. innocua*, at 37 and 30 °C, the MIC was 0.300 µl/ml. At 15 °C, the MIC was 0.500 µl/ml and at 8 °C it was 1.000 µl/ml. The authors concluded that temperature has an effect on the ability of essential oils to inhibit and kill bacteria. Also, it affects the concentration of active substance in essential oils that are used to inhibit and kill bacteria because when the temperature drops, the microbial cells are stable and increased in strength, so that the essential oils or active substances in them penetrate the cell membrane in

smaller quantities. Therefore, the concentration of essential oil and active substances in essential oils should be increased. In other words, the MIC and MBC of essential oil need to be increased as well.⁴⁴

Survival of *Salmonella typhimurium* at a Storage Temperature of 4 °C

Highly perishable food is usually stored at 4 °C. The antibacterial activity test in this experiment thus was of interest at this temperature. Five essential oils and their combination giving the highest activity (or lowest MBC) – finger root oil, clove oil, lemongrass oil, cardamom oil and lemongrass oil combined with cardamom oil – were selected for testing against *S. typhimurium* at 4 °C. The result showed that the concentration of finger root and clove essential oils required to kill *S. typhimurium* at 4 °C was four times the MBC at 37 °C (3.125 µl/ml) (Table 7). The corresponding concentrations for essential oils from cardamom, lemongrass and lemongrass combined with cardamom were 12.5, 0.781 and 3.125 µl/ml, respectively, representing a 16-fold increase from their MBC. Furthermore, complete antibacterial activity could be observed within 30 min for all tested essential oils except for lemongrass oil for which complete antibacterial activity was observed after 3 hr. The result could imply that temperature has an effect on the antibacterial activity of essential oils, namely, that the MBC at a lower temperature is greater than the MBC at a higher temperature to maintain the greatest efficacy for antibacterial

activity. The temperature dependency of MBC was also reported by Govaris,⁴² who studied the inhibitory effect of 0.6% and 0.9% essential oil from oregano

on *S. Enteritidis* in ground lamb; their result indicated that keeping ground lamb at 10 °C inhibited germs better than at 4 °C.

Table 7: Inhibitory activity of essential oils from spices against *Salmonella typhimurium* at 4 °C

Essential oils	Concentration (µl/ml)	Microorganisms (log CFU/ml) Time (hr)												
		0	0.5	1	1.5	2	2.5	3	3.5	4	4.5	5	5.5	6
Finger root oil	MBC (0.781)	6.63	6.35	6.29	6.26	6.22	6.16	6.10	6.05	6.03	6.05	6.03	5.96	5.94
	4MBC (3.125)	6.56	0	0	0	0	0	0	0	0	0	0	0	0
	16MBC (12.50)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
	64MBC (50.0)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
Clove oil	MBC (0.781)	6.52	6.19	6.25	6.21	6.17	6.19	6.03	6.04	5.92	5.90	5.89	5.90	5.87
	4MBC (3.125)	6.55	0	0	0	0	0	0	0	0	0	0	0	0
	16MBC (12.50)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
	64MBC (50.0)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
Lemon grass oil	MBC (0.0488)	6.71	6.66	6.68	6.61	6.58	6.65	6.65	6.65	6.55	6.54	6.55	6.48	6.53
	4MBC (0.195)	6.58	5.92	5.92	5.92	5.90	5.88	5.86	5.84	5.85	5.80	5.80	5.79	5.78
	16MBC (0.781)	6.50	2.99	2.67	2.45	1.85	1.54	1	0	0	0	0	0	0
	64MBC (3.125)	6.49	0	0	0	0	0	0	0	0	0	0	0	0
Cardamom oil	MBC (0.781)	6.72	6.40	6.36	6.34	6.31	6.26	6.23	6.22	6.21	6.19	6.18	6.16	6.14
	4MBC (3.125)	6.82	4.63	4.39	4.33	4.23	4.19	4.12	4.12	4.08	4.02	3.91	3.86	3.84
	16MBC (12.50)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
	64MBC (50.0)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
Lemon grass oil + cardamom oil	MBC (0.195)	6.56	5.98	5.91	5.87	5.86	5.86	5.84	5.84	5.84	5.82	5.81	5.80	5.80
	4MBC (0.781)	6.72	5.70	5.59	5.51	5.36	5.40	5.25	5.14	4.97	4.87	4.77	4.77	4.76
	16MBC (3.125)	6.74	0	0	0	0	0	0	0	0	0	0	0	0
	64 MBC (12.50)	6.74	0	0	0	0	0	0	0	0	0	0	0	0

The essential oils employed in this study exhibited inhibitory effects on *S. typhimurium*; however, further investigation is necessary to elucidate the specific constituents of essential oils responsible for this activity. Future research should aim to identify and evaluate the individual components of essential oils that effectively exert an effect on *S. typhimurium*.

Mathematical Models Predicting the Survival of *Salmonella typhimurium* using Essential Oils

Tables 8 and 9 present the parameters of various models employed to predict the survival of *S. typhimurium* in the presence of essential oils, as well as the coefficients of determination (R^2) and root mean square error (RMSE). The results indicated that the Weibull and modified Gompertz models were more suitable for describing the reduction in the number of surviving *S. typhimurium* compared to the first-order kinetic model. These findings are

consistent with a previous study by Oliveira,³⁵ which utilized the Weibull model to describe the number of *S. Enteritidis* in raw mashed beef treated with essential oils from oregano and lemongrass during storage at 4 ± 2 °C for 6 days. Nikkiah⁴⁵ studied the ability of essential oils from basil, cinnamon, rosemary and marjoram to kill *Penicillium expansum* and *Botrytis cinerea*; their results showed that the modified Gompertz model could explain the reduction of number of *P. expansum* and *B. cinerea*.

The parameters presented in Table 8 offer quantitative insights into the behaviour of the models and aid in comprehending how the response variable evolves over time or under specific conditions. These parameter values are estimated through data fitting or statistical analysis to determine the optimal fit of the model to the observed data. In the context of the first-order kinetic model, the

parameter “k” denotes the rate at which a reaction occurs. A higher “k” value signifies a faster reaction, while a lower value suggests a slower rate. For the Weibull model, the parameter “b” influences the shape of the curve and determines the rate of change of the response variable over time. A higher “b” value leads to a steeper increase or decrease in the response variable, resulting in a more rapid change. Conversely, a lower “b” value yields a more gradual change. Additionally, the “n” parameter in the Weibull model determines the time required for the response variable to reach a certain proportion. It affects the duration needed to achieve the specified level. A higher “n” value indicates a longer time

for reaching the desired proportion, while a lower value suggests a shorter duration. The modified Gompertz model incorporates the “M” and “A” parameters. The maximum value of “M” represents the highest level that the response variable can reach. It characterizes the maximum growth or decay attainable by the response variable. On the other hand, the initial value of “A” denotes the starting point of the response variable before it begins to change or evolve. It signifies the initial level of the response at the onset of the process or reaction. A higher “A” value corresponds to a higher starting point, whereas a lower value indicates a lower initial level.

Table 8: Modelling parameters for first-order kinetic, Weibull, and Modified Gompertz models

Treatment	First-order kinetic model	Weibull model	Modified Gompertz model		
	k	b	n	M	A
Finger root oil (0.781 µl/ml)	0.045	0.22	0.409	5.969	0.567
Clove oil (0.781 µl/ml)	0.093	0.019	0.5	5.785	0.351
Lemongrass oil (0.049 µl/ml)	0.03	< 0.001	2.856	6.425	0.207
Lemongrass oil (0.195 µl/ml)	0.071	0.046	0.099	5.844	3.905
Lemongrass oil (0.781 µl/ml)	0.848	0.406	0.739	0.015	0.154
Cardamom oil (0.781 µl/ml)	0.066	0.024	0.262	6.195	1.072
Cardamom oil (3.125 µl/ml)	0.274	0.186	0.151	4.059	2.335
Lemongrass oil + cardamom oil (0.781 µl/ml)	0.066	0.045	0.099	5.836	2.902
Lemongrass oil + cardamom oil (3.125 µl/ml)	0.249	0.077	0.367	4.847	0.672

Therefore, mathematical models that accurately describe *S. typhimurium* survival offer numerous advantages across various domains. These models possess predictive power, enabling us to understand how *S. typhimurium* behaves under different conditions. Additionally, they facilitate the optimization of strategies for controlling *S. typhimurium* survival in healthcare and agriculture settings. By using mathematical models, we gain insights into the underlying mechanisms governing *S. typhimurium* survival. This knowledge can be

utilized to enhance cost and time efficiency by reducing the necessity for extensive experimental work. Overall, mathematical models provide a quantitative framework that aids in understanding and managing *S. typhimurium* populations. They guide interventions, predict outcomes, and contribute to the development of effective measures for combating the threat of *S. typhimurium*.

However, in the further study, it is crucial to broaden the investigation by exploring a wider

range of concentrations of spice essential oils. This comprehensive approach allows for a thorough examination of the factors influencing the survival rates of *S. typhimurium*. To gain a holistic understanding, it is recommended to incorporate

mathematical modelling approaches in these investigations. This integration will provide valuable insights into the complex dynamics and interactions involved.

Table 9: Comparison of coefficients of determination and root mean square error of first-order kinetic, Weibull, and Modified Gompertz models

Treatment	First-order kinetic model		Weibull model		Modified Gompertz model	
	R ²	RMSE	R ²	RMSE	R ²	RMSE
Finger root oil (0.781 µl/ml)	0.873	0.480	0.963	2.956	0.952	0.043
Clove oil (0.781 µl/ml)	0.893	1.288	0.921	0.053	0.887	0.068
Lemongrass oil (0.049 µl/ml)	0.715	0.545	0.696	0.073	0.676	0.041
Lemongrass oil (0.195 µl/ml)	0.474	0.802	0.990	0.020	0.952	0.044
Lemongrass oil (0.781 µl/ml)	0.927	0.515	0.947	0.427	0.931	0.489
Cardamom oil (0.781 µl/ml)	0.747	0.930	0.994	0.012	0.927	0.041
Cardamom oil (3.125 µl/ml)	0.615	1.534	0.998	0.037	0.968	0.134
Lemongrass oil + cardamom oil (0.781 µl/ml)	0.445	0.742	0.998	0.009	0.986	0.023
Lemongrass oil + cardamom oil (3.125 µl/ml)	0.884	2.092	0.976	0.080	0.906	0.168

Anti-Salmonella typhimurium Activity of Essential Oils from Spices in Salad Cream

In the present study, during storage at 4 °C, no *S. typhimurium* was found in salad cream samples incorporating essential oils from spices (finger root, clove, lemongrass, cardamom and lemongrass combined with cardamom) and to which *S. typhimurium* had been added (as shown in Table 10). However, *S. typhimurium* was detected in salad cream samples with added *S. typhimurium* but without essential oils during a storage period of 7 days at 4 °C. *Salmonella spp.* are able to adapt to an inappropriate environment. For example, *Salmonella spp.* can grow or survive in salad cream samples during refrigerated storage 2–4 °C.³⁵ In addition, *Salmonella spp.* can survive for months at low temperatures (< 5 °C) and adapt to inappropriate

external pH, responding to acid resistance by the process called acid tolerance response (ATR).⁴⁶ For the reasons above, the addition of essential oils from spices could inhibit the growth of *S. typhimurium* in salad cream samples during storage for 7 days at 4 °C.

Despite the significant impact on bacterial growth observed when incorporating specific components, namely ground pepper, and mustard, during the salad cream preparation process, these ingredients were deliberately included in the control sample of the salad cream. Consequently, the addition of essential oils to the salad cream in this study resulted in a considerable reduction in bacterial counts compared to the control sample, which lacked any essential oils.

Table 10: *Salmonella typhimurium* in salad cream with and without essential oils from spices during storage at 4 °C

Samples	Concentration of essential oil (µl/ml)	S. typhimurium							
		Duration of storage (in days)							
		0	1	2	3	4	5	6	7
1. Salad cream without essential oils from spices and without S. typhimurium	0	x	x	x	x	x	x	x	x
2. Salad cream with finger root essential oil and with added S. typhimurium	3.125	/	x	x	x	x	x	x	x
3. Salad cream with clove essential oil and with added S. typhimurium	3.125	/	x	x	x	x	x	x	x
4. Salad cream with lemongrass essential oil and with added S. typhimurium	0.781	/	x	x	x	x	x	x	x
5. Salad cream with cardamom essential oil and with added S. typhimurium	6.250	/	x	x	x	x	x	x	x
6. Salad cream with lemongrass and cardamom essential oil and with added S. typhimurium	3.125	/	x	x	x	x	x	x	x
7. Salad cream without essential oils and with added S. typhimurium	0	/	/	/	/	/	/	/	/

x means S. typhimurium was not detected; / means S. typhimurium was detected.

Assessment of Antimicrobial Activity of Spice-Derived Essential Oils in Salad Cream

Salad cream samples containing essential oils from spices (finger root, clove, lemongrass, cardamom and lemongrass combined with cardamom), salad cream from a local market and supermarket, salad cream from Mae Hia market (shop 1), salad cream from Mae Hia market (shop 2) and salad cream from the Royal Project were used to determine the number of microorganisms and pH value. Additionally, the salad cream samples were packed, stored at 4 °C, and subjected to analysis on days 0, 7, and 15 to assess the survival of S. typhimurium. Total microorganisms and pH were analyzed at 3-day intervals between 0 and 15 days of storage. The results showed that S. typhimurium was not found in any of the salad cream samples containing essential oils from spices (finger root, clove, lemongrass, cardamom and lemongrass combined with cardamom) during 15 days of storage. The total number of microbes in salad cream from Mae Hia market (shop 1 and shop 2) and salad cream without essential oils from spices was higher than that in salad cream containing essential oils from spices and salad cream from the Royal Project Foundation at 12 and 15 days. However, all salad cream samples were in accordance with the Thai Community Products Standard for salad dressing products in Thailand (< 10,000 CFU/g). The approximate range of pH of all salad cream samples was 3.52–4.24. The salad cream from Mae Hia market (shop 1) had a higher range of pH (4.02–4.24) compared to other salad cream samples.

Sensory Evaluation of Salad Cream Incorporating Essential Oils from Spices

Based on the consumer preference data presented in Table 11, there were no statistically significant differences (p ≤ 0.05) observed between salad cream without essential oils from spices, salad cream with essential oils from spices (finger root, clove, lemongrass, cardamom and lemongrass combined with cardamom) and salad cream from the Royal Project Foundation. Furthermore, the colour and overall liking ratings for all salad cream samples did not demonstrate any significant differences (p ≤ 0.05). In addition, the salad cream from the Royal Project Foundation had the lowest scores for appearance and texture. The salad cream without essential oils from spices had the highest odour scores.

Table 11: Mean scores for sensory acceptability of salad cream with and without essential oils from spices and salad cream from the Royal Project Foundation

Samples	Appearance	Colour	Odour	Texture	Overall liking
Salad cream without essential oil	6.50 ± 0.97 ^a	6.17 ± 1.23 ^a	5.43 ± 1.61 ^a	6.43 ± 1.14 ^a	6.17 ± 1.34 ^a
Salad cream with finger root oil	6.43 ± 0.86 ^a	6.53 ± 0.94 ^a	4.37 ± 1.67 ^b	6.60 ± 1.10 ^a	5.80 ± 1.27 ^a
Salad cream with clove oil	6.40 ± 1.04 ^a	6.50 ± 1.14 ^a	4.73 ± 2.08 ^{ab}	6.37 ± 1.27 ^a	5.73 ± 1.74 ^a
Salad cream with lemongrass oil	6.43 ± 1.01 ^a	6.43 ± 1.04 ^a	5.07 ± 1.62 ^{ab}	6.30 ± 1.09 ^a	5.87 ± 1.10 ^a
Salad cream with cardamom oil	6.33 ± 0.88 ^a	6.27 ± 1.05 ^a	4.60 ± 1.52 ^{ab}	6.67 ± 1.03 ^a	5.73 ± 0.98 ^a
Salad cream with lemongrass and cardamom oils	6.37 ± 1.00 ^a	6.73 ± 1.14 ^a	5.13 ± 1.33 ^{ab}	6.40 ± 1.00 ^a	5.77 ± 1.19 ^a
Salad cream from the Royal Project Foundation	5.27 ± 1.89 ^b	6.50 ± 1.70 ^a	5.10 ± 2.38 ^{ab}	4.97 ± 1.59 ^b	5.57 ± 1.91 ^a

Note: Values are the average ± standard deviation. Statistically significant differences within the same column are denoted by different lowercase letters in superscript, with a significance level set at $p \leq 0.05$.

Conclusions

The essential oils from finger root, clove, lemongrass, cardamom, and lemongrass combined with cardamom showed strong inhibitory activity against *S. typhimurium*. At 4 °C, the required concentrations of finger root and clove essential oils to kill *S. typhimurium* were four times higher than the MBC at 37 °C (3.125 µl/ml). Cardamom, lemongrass, and lemongrass combined with cardamom essential oils exhibited corresponding concentrations of 12.5, 0.781, and 3.125 µl/ml, representing a 16-fold increase from their MBC. Complete inactivation occurred within 30 minutes for most essential oils, except for lemongrass oil, which took 3.5 hours. The modified Gompertz model fit the surviving number of *S. typhimurium* when using finger root essential oil, while the Weibull and modified Gompertz models best described the decrease in surviving bacteria when inhibited by clove, lemongrass, cardamom, and lemongrass combined with cardamom essential oils.

During 15-day storage at 4 °C, salad cream containing essential oils from spices (finger root, clove, lemongrass, cardamom, and lemongrass combined with cardamom) and *S. typhimurium* exhibited no detectable presence of the bacteria, and the overall microbe count decreased. These findings suggest the potential of spice essential oils as natural preservatives. Sensory evaluations

comparing appearance, colour, odour, texture, and overall liking between salad creams without essential oils, with essential oils, and from the Royal Project Foundation showed no significant differences ($p \leq 0.05$). Hence, salad cream containing essential oils from spices can be used for commercial production.

The obtained results demonstrate the significant inhibitory and antiseptic effects of essential oils derived from spices against *S. typhimurium*. However, further investigations are necessary to delve into the chemical composition of finger roots, cloves, lemongrass, and cardamom essential oils. This research should prioritize the identification and characterization of specific bioactive constituents responsible for the inhibitory and eradication properties against *S. typhimurium*. Additionally, it is essential to conduct studies utilizing a broader range of concentrations for spice essential oils in order to comprehensively assess their impact on the survival rates of *S. typhimurium*. By employing mathematical modelling approaches, a more thorough understanding of the underlying factors influencing these survival rates can be achieved.

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Conflicts of interest

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