Meta-Analysis on Effect of Essential Oils and Extracts of Spices on the Microbiological Quality of Meat and Poultry Products

(Analisis Meta Kesan Minyak Pati dan Ekstrak Rempah ke atas Kualiti Mikrobiologi Produk Daging dan Poltri)

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ABSTRACT

Different spices have been reported containing antimicrobial compounds that can prevent or reduce the microbiological spoilage or pathogenic bacteria in meat or poultry products with various results. This study evaluated the effect of essential oils and extracts of spices in reducing bacteria in meat/poultry products using a meta-analysis approach by comparing the Hedges'd effect size (standardized mean difference, SMD, and 95% confidence interval, CI). A total of 240 data, extracted from 10 articles that were selected by Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) from 121 full-text articles, were analyzed using Meta Essential tools. The results showed that essential oils had a better cumulative significant effect in reducing bacterial loads of meat/poultry products (SMD=-4.37, 95%CI=-5.63 to -3.10) in comparison to the extracts (SMD=-3.66, 95%Cl=-4.56 to -2.76). As essential oils, cassia (SMD=-58.17, 95%CI=-109.88 to -6.47) showed the best effect size, whereas as extract, ganghwayassuk (SMD=-4.19, 95%CI=-6.22 to -2.16) was the most significant. Furthermore, the total plate count was significantly affected by cassia (SMD=-58.17, 95%CI=-109.88 to -6.47), Enterobacteriaceae by sage (SMD=-5.93, 95%CI=-8.32 to -3.54), and coliform also by sage (SMD=-3.79, 95%CI=-6.76 to -0.82). In general, Salmonella spp. was found as pathogenic bacterium that was the most affected (SMD=-19.68 and 95%CI=-39.01 to -0.35). In the form of essential oils, dipping was the best way in reducing microorganisms, while as extracts, adding them in the products was the best method. This study provided reliable data in selecting spices for applications to improve the quality and safety of meat and poultry products.

Keywords: Antimicrobial; meat/poultry-based products; meta-analysis; spices

ABSTRAK

Rempah yang berbeza telah dilaporkan mengandungi sebatian antimikrob yang boleh menghalang atau mengurangkan kerosakan mikrobiologi atau bakteria patogen dalam daging atau produk poltri dengan keputusan yang pelbagai. Kajian ini menilai kesan minyak pati dan ekstrak rempah dalam mengurangkan bakteria dalam produk daging/poltri menggunakan pendekatan analisis meta dengan membandingkan saiz kesan Hedges'd (perbezaan min standard, SMD dan selang keyakinan 95%, CI). Sebanyak 240 data yang diekstrak daripada 10 artikel yang dipilih melalui Item Pelaporan Pilihan untuk Ulasan Sistematik dan Analisis Meta (PRISMA) daripada 121 artikel teks penuh, telah dianalisis menggunakan alat Meta Perlu. Keputusan menunjukkan bahawa minyak pati mempunyai kesan ketara kumulatif yang lebih baik dalam mengurangkan beban bakteria produk daging/poltri (SMD=-4.37, 95%CI=-5.63 hingga -3.10) berbanding ekstrak (SMD=-3.66, 95% Cl=-4.56 hingga -2.76). Sebagai minyak pati, cassia (SMD=-58.17, 95%CI=-109.88 hingga -6.47) menunjukkan saiz kesan terbaik, manakala sebagai ekstrak, ganghwayassuk (SMD=-4.19, 95%CI=-6.22 hingga -2.16) adalah paling ketara. Tambahan pula, jumlah kiraan plat terjejas dengan ketara oleh cassia (SMD=-58.17, 95%CI=-109.88 hingga -6.47), Enterobacteriaceae oleh sej (SMD=-5.93, 95%CI=-8.32 hingga -3.54) dan koliform juga oleh sej (SMD=-3.79, 95%CI=-6.76 hingga -0.82). Secara umum, Salmonella spp. didapati sebagai bakteria patogen yang paling terjejas (SMD=-19.68 dan 95%CI=-39.01 hingga -0.35). Dalam bentuk minyak pati, pencelupan adalah cara terbaik dalam mengurangkan mikroorganisma, manakala sebagai ekstrak, menambahnya dalam produk adalah kaedah terbaik. Kajian ini menyediakan data yang boleh dipercayai dalam memilih rempah untuk digunakan dalam meningkatkan kualiti dan keselamatan produk daging dan poltri.

Kata kunci: Analisis meta; antimikrob; produk berasaskan daging/poltri; rempah

INTRODUCTION

Foodborne disease is the main outbreak throughout the world caused by consuming food that contains pathogenic microorganisms. Meat and poultry products are rich in protein, and it is quickly deteriorated by the growth of microorganisms (Dewi et al. 2016; Hernando et al. 2015) that causes the products to be recalled frequently. USDA for 2021 reported that there were at least 27 cases of recalls of meat and poultry products due to microbial contamination, such as *Listeria*, *Salmonella*, and *Escherichia coli* (USDA 2021).

The presence of microbial contamination in a product does not only affect safety but is also closely related to the quality of a food product. Studies on spices and their applications as antimicrobial agents on meat products have been widely carried out, and they showed different results. Spices such as cinnamon, onion, oregano, thyme, rosemary, cloves, and sage as essential oils and extracts have shown antimicrobial activity against *Escherichia coli, Staphylococcus aureus, Salmonella, Listeria*, and some yeast (Abdel-Wahab et al. 2020; Pedrós-Garrido et al. 2020; Van Haute et al. 2017).

The antimicrobial effectiveness of spices also depends on the method of application to the product. The addition of spices extracts to meatballs (Nugboon & Intarapichet 2015), chicken nugget (Martínez, Ros & Nieto 2020) and chicken patties (Gao et al. 2019) have been shown reduction on microorganisms loads in various level, as well as spraying of spices extracts on fresh mutton (Kumudavally et al. 2011). Similarly, various effect has been found on the applications of essential oils of spices on meat/poultry products. Different studies have been reported the used essential oil of spices such as by spraying on fresh slices of beef (Sirocchi et al. 2017) and on fresh meat (Mehdizadeh et al. 2020), and by adding to the product such as beef burgers (Sharafati-Chaleshtori et al. 2014), pork sausage (Bortolotto et al. 2018), mutton nugget (Kumar et al. 2018), and chicken sausage (Sharma et al. 2018).

Further analysis is needed to examine the significance of various spices sources as antimicrobial and their application. Meta-analysis is a quantitative scientific synthesis consisting of various research results that can analyze evidence-based and resolve research results that seem contradictory (Gurevitch et al. 2018). This research aimed to evaluate the effect of essential oils and extracts of spices in reducing the number of bacteria in meat and poultry-based products that poses the safety and quality of the products.

MATERIALS AND METHODS

MATERIALS AND TOOLS

The materials were articles from reputable international journals obtained from various online databases, including Google Scholar, ProQuest, Pubmed, Science Direct, and Wiley Online Library. The tools used were Mendeley software version 1.19.4.0, Zotero version 5.0.0.0, Microsoft Excel version 2016, and Meta Essentials version 1.5.

SEARCHING STRATEGIES

Collecting of articles were started by specifying research questions using the PICO criteria (Population, Intervention, Comparison, Outcome) and the inclusion as well as the exclusion criteria (Tawfik et al. 2019). The inclusion criteria were articles that have been published in reputable journals for the last 10 years (2011-2021), the primary subject was meat or poultry products, the treatments included extracts and or essential oils derived from spices, control data were available, and reported the quantitative data as research results. The exclusion criteria are articles in the form of reviews, books, or patents and articles with incomplete data. Then, related articles were searched from reputable online databases, i.e., Google Scholar, ProQuest, Pubmed, Science Direct, and Wiley Online Library (Jayanegara, Wina & Takahashi 2014), using the keywords 'essential oil', 'extract', 'spice', 'meat products', and 'poultry products' with boolean operator terms OR, AND, and NOT (Jahan et al. 2016).

SELECTION OF STUDY

The related articles were then selected for eligibility following the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) approach (Moher et al. 2009), which included identification, selection, and suitability to obtain selected articles for a meta-analysis. Articles from several databases were collected during the identification step, and the duplicate articles were then removed by reference managers Mendeley and Zotero. Furthermore, the articles were selected based on the abstract, title, and completeness that met the inclusion criteria to be processed for meta-analysis.

DATA EXTRACTION

Articles that met the conformity selection are subjected to data extraction. Information from the entire contents of the articles were tabulated on Microsoft Excel, including the

author's name, year of publication, article origin, article index, type of spices, the form of spices used (essential oil or extract), method of obtaining, concentration, method of application, target microorganisms, control, number of microorganisms (log CFU/g), number of replications, standard deviation, and standard error.

STATISTICAL ANALYSIS

Within the data that were collected from different studies, the untreated samples were put into the control group (C), while the samples treated with essential oils or extracts from spices were included in the experimental group (E). The weighted analysis using Hedges'd (Standard Mean Difference/SMD) was used for statistical analysis using the Meta Essential tools. The mean, standard deviation or standard error, and the number of replicate experiments were extracted from the selected articles. The SMD with corresponding 95%CI values in this research were pooled using the random-effects model. The effect size is statistically significant if the CI does not reach the null effect size. The heterogeneity across studies was explored using the I² index, where the I² > 50% indicated sufficient heterogeneity (Afandi et al. 2021).

The collected data was calculated for the pooled standard deviation (S), the correction factor for small sample size (J), and the effect size (d) (Palupi et al. 2012) with the following formulas:

$$S = \sqrt{\frac{(N^{E}-1)(s^{E})^{2} + (N^{C}-1)(s^{C})^{2}}{(N^{E}+N^{C}-2)}}$$

$$J=1-\frac{3}{(4(N^C+N^E-2)-1)}$$

$$d = \frac{(X^E + X^C)}{S}J$$

where X^E is the mean value from the experimental group; X^C is the mean value of the control group; S^C is the standard deviation of the control group; S^C is the sample size of the control group; and S^C is the sample size of the experimental group. Cohen's benchmarks were used as the standard judgment borders to indicate the large of the effect size, i.e., 0.2 for small, 0.5 for medium, and 0.8 for large effect size. The meta-analysis results are interpreted as a forest plot to show the cumulative effect size. The variables used for subgroup analysis in this research were the type of spices, the form of spice (essential oils or extracts), the group of microorganisms and the application method to the products.

RESULTS AND DISCUSSION

A total of 1256 studies were identified from the online databases, including Science Direct, Pubmed, ProQuest, and Wiley Online Library, indicated that the effect of essential oils and extracts of spices were widely studied. All articles were stratified to exclude duplicates (337 articles) and inappropriate articles (e.g., without control, standard deviation, standard error, or replication). From 121 full-text articles, 10 final studies (Table 1) were fit

TABLE 1. List of studies used in meta-analysis

Study	Place	Product	Spices	Essentials oils/ extract
Sarıçoban & Yilmaz (2014)	Turkey	Chicken patties	Thyme	Essential oils
			Cumin	Essential oils
Khanjari et al. (2013)	Greece	Raw chicken fillet	Oregano	Essential oils
Sharma et al. (2017)	India	Chicken sausage	Clove	Essential oils
			Holy Basil	Essential oils
			Cassia	Essential oils
			Thyme	Essential oils
Triki et al. (2013)	Spain	Fresh Sausage	Olive	Essential oils
de Melo et al. (2017)	Brazil	Chicken breast	Oregano	Essential oils
de Oliveira et al. (2013)	Brazil	Raw beef	Thyme	Essential oils
			Rosemary	Essential oils
Sharafati-Chaleshtori et al. (2014)	Iran	Beef burger	Tarragon	Essential oils
Cegiełka et al. (2019)	Poland	Chicken meat	Sage	Essential oils
			-	Water extract Ethanolic extract
Hwang et al. (2013)	Korea	Raw chicken patties	Ganghwa-yakssuk	Ethanolic extract
Kumudavally et al. (2011)	India	Fresh mutton	Clove	Ethanolic extract

for meta-analysis with a total of 240 data. A total of 10 essential oils of spices were implicated, i.e., basil, cassia, clove, cumin, olive, oregano, rosemary, sage, tarragon, and thyme. Three extracts of spices were involved, i.e., clove, ganghwayakssuk, and sage.

OVERALL EFFECT SIZE OF ESSENTIAL OILS AND EXTRACTS OF SPICES ON MICROBIOLOGICAL QUALITY Significant effect of essential oils and extracts of spices in reducing bacterial counts in meat/poultry products

was found based on the standardized mean difference (SMD) and 95% confident interval of the studies. The forest plot of cumulative effect size of essential oils showed SMD =-4.37, 95%CI = -5.63 to -3.10, I^2 = 83.32%, and p = 0.000 (Figure 1(A)). The extracts of spices also showed a significant effect on decreasing bacteria in meat/poultry-based products (Figure 1(B)) with SMD = -3.66, 95%Cl= -4.56 to -2.76, I^2 = 84.85%, p = 0.000. The reduction of microorganisms by extract was slightly less than by essential oils.

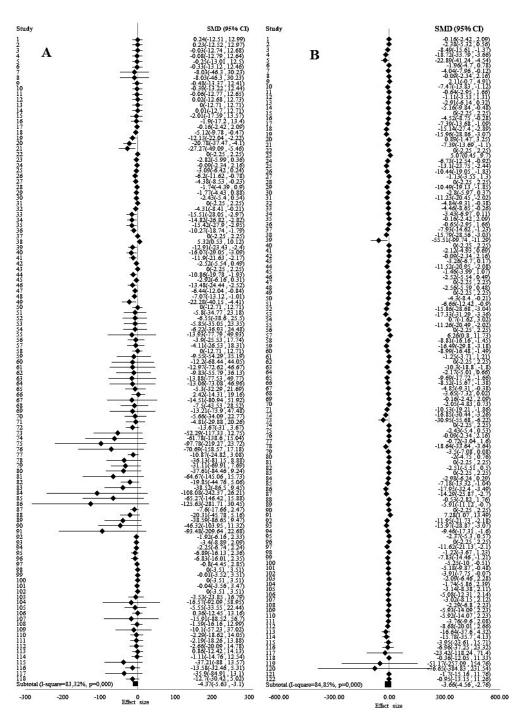


FIGURE 1. Forest plot of cumulative effect size and 95% confidence interval (CI) of (A) essential oils and (B) extracts

Essential oils are secondary metabolites derived from plant parts such as flowers, leaves, roots, stems, or bark which have volatile characteristics and produce a distinctive aroma and odor that can act as antimicrobials (Macwan et al. 2016; Valdivieso-Ugarte et al. 2019). On the other hand, the extract is any substance obtained from the extracting process or separating active substances from part of plants or natural materials by mixing them with various suitable solvents (Mukhtarini 2014). In general, essential oils and extracts of spices significantly reduced the microbial loads, hence enhancing the microbiological quality of meat/ poultry products, as shown in Figure 1(A) and 1(B). For spices extracts analyses, although only three studies were met the inclusion criteria, however, a lot of data have been generated. In these three studies, three different extracts were tested against many microbial groups, i.e., total mesophilic count, psychotropic bacteria, Enterobacteriaceae, coliform, enterococci, and

Salmonella (Cegiełka et al. 2019; Hwang et al. 2013; Kumudavally et al. 2011).

Based on the subgroup analyses for the type of spices (Figure 2(A)), the combined effect size on reducing bacteria was not significant since its 95%CI crossing the null effect size (SMD=-3.33, 95%CI =-6.79 to 0.12, I^2 = 83.32%, and p = 0.000). However, the significant effects were demonstrated by essentials oils of cassia, clove, holy basil, olive, oregano and sage. The best three significant effects were showed, in order, by cassia (SMD=-58.17, 95%CI=-109.88 to -6.47), cloves (SMD=-54.6, 95%CI=-92.91 to -16.3), and basil (SMD=-32.13, 95%CI= -54.55 to -9.71). Based on the microorganism affected, the most significant effect was found against Salmonella spp. (SMD=-19.68, 95%CI=-39.01 to -0.35) followed by the total plate counts (SMD=-8.73, 95%CI=-13.09 to -4.38). Furthermore, dipping the product in essential oils was found as the best application method (SMD=-6.07, 95%CI=-8.17 to -3.96) in comparison to adding to the batter or vaporing the essential oils to the product.

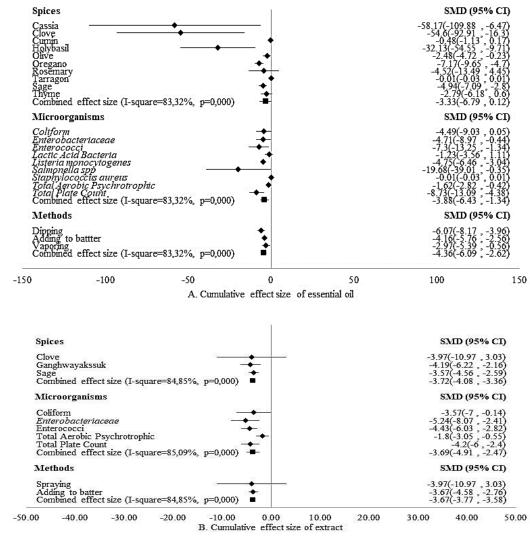


FIGURE 2. Forest plot of subgroup analysis of cumulative effect size and 95% confidence interval (CI) of (A) essential oils, extracted from 118 data and (B) extract, from 122 data, for the spices involved, the microorganisms affected, and the application methods

Cassia bark is derived from Cinnamomum cassia, a plant belonging to the Lauraceae family that is widely used for seasoning meat products, poultry products, sauces, to confectionery as a flavoring agent (Prasetya & Ngadiwiyana 2006; Sharma et al. 2017). Essential oil of cassia bark is dominated by volatile compounds, including cinnamaldehyde, trans-4-methoxycinnamaldehyde, cinnamyl acetate, and o-hydroxycinnamic acid, which act as antibacterial against S. aureus, E. coli, B. subtilis, Salmonella and Listeria monocytogenes (Huang et al. 2014; Kacaniova et al. 2021; Prasetya & Ngadiwiyana 2006). Clove (Syzygium aromaticum) is largely used as a food preservative due to its phenolic compounds such as flavonoids, hydroxybenzoic acids, hydroxyphenyl propens, hydroxycinnamic acids and eugenol, which are potential as antioxidant and antimicrobial agents (Cortés-Rojas, de Souza & Oliveira 2014). The application of clove essential oils on meat and poultry products reduces undesirable reactions and decreased bacterial counts, including Listeria monocytogenes, Salmonella, and Escherichia coli (Haro-González et al. 2021; Kammon, Almaeyoufi & Asheg 2019; Vrinda Menon & Garg 2001). Furthermore, holy basil (Ocimum sanctum Linn.) is an aromatic plant of the Labiate family that is being used as spices, seasoning for cooking, and antimicrobial agents for various food products (Amor et al. 2021; Mittal, Kumar & Chahal 2018). This plant contains saponins, tannins, flavonoids, carvacrol, eugenol, methyl cinnamate, methyl eugenol, citral, and linalool which have antibacterial properties (Jaggi, Madaan & Singh 2003; Mittal, Kumar & Chahal 2018; Pattanayak et al. 2010; Sharma et al. 2017). As an essential oil, basil contains eugenol, eucalyptol, camphor, alpha bisabolene, beta bisabolene, and beta-caryophyllene, which are responsible for antimicrobial properties (Yamani et al. 2016). Previous studies showed basil as an essential oil or extract was effective against E. coli and Staphylococcus aureus (Angelina, Turnip & Khotimah 2015; Fachriyah, Wibawa & Awaliyah 2020).

On the other hand, when the spices were applied as extract, the extract of ganghwayassuk (SMD=-4.19, 95%CI=-6.22 to -2.16) had the most significant effect on reducing the number of bacteria in meat/poultry products (Figure 2(B)), followed by sage. The group of bacteria that was the most significantly affected was *Enterobacteriaceae* (SMD=-5.24, 95%CI=-8.07 to -2.41). The application of spices extracts by adding them to the batter has been shown to reduce the number of bacteria (SMD=-3.67, 95%CI=-4.58 to -2.76).

Ganghwayassuk (Artemisia princeps Pamp.) is a native plant from East Asia (Japan, Korea, and China) whose young leaves and seeds are often used as a flavoring and food coloring (Hwang et al. 2011; Trendafilova et al. 2021; Toda 2005; Umano et al. 2000; Verloove & Andeweg 2020). The leaf extract of this plant has volatile compounds which are dominated by monoterpenoid compounds followed by aromatic components, aliphatic esters, aliphatic alcohols, monoterpenes, sesquiterpenes, sesquiterpenoids, aliphatic aldehydes, aliphatic hydrocarbons, aliphatic ketones, and polyphenols that can act as antibacterial (Hwang et al. 2011; Umano et al. 2000). Furthermore, sage (Salvia officinalis) is an aromatic plant originating from the Lamiaceae family which is generally used in cooking as culinary herbs, food preparation components, and flavorings that have been shown antibacterial activity against foodborne pathogenic bacteria (Ghorbani & Esmaeilizadeh 2017; Mitic-Culafic et al. 2005; Moghimi et al. 2016; Ouahida, Amina & Halima 2021; Šojić et al. 2021). The chemical components of sage essential oil are dominated by volatile compounds such as terpenoids, terpenes, and flavonoids (Ghorbani & Esmaeilizadeh 2017; Mitic-Culafic et al. 2005). These components can act as antibacterial by causing leakage of ions, metabolites, and enzymes and are effective against pathogenic food bacteria, both Gram-negative and positive (Ghorbani & Esmaeilizadeh 2017; Mitic-Culafic et al. 2005; Moghimi et al. 2016).

EFFECT OF ESSENTIAL OILS AND EXTRACTS OF SPICES ON TOTAL PLATE COUNT (TPC)

The combined effect size of essential oils and extracts, based on subgroup analysis, indicated a significantly decrease of TPC, with SMD = -5.95, 95%CI= -11.75 to -0.16, I² = 85.60%, and p = 0.000 (Figure 3). The most significant effect on decreasing the TPC was shown by the cassia, holy basil, and clove. The use of essential oils resulted in a higher effect on TPC (SMD=-8.73, 95%CI=-13.09 to -4.38) than extracts (SMD=-4.2, 95%CI=-6 to -2.4), although the use of extracts of spices has been shown to reduce the number of Gram-positive and Gram-negative bacteria (Shan et al. 2009; Zhang, Wu & Guo 2016).

The application method that has the most significant effect on TPC is by adding essential oils or extracts of spices in food products (SMD=-5.48, 95%CI=-7.66 to -3.31) in comparison to spraying the product. The addition of extracts to the batter has been shown to reduce the number of bacteria and extend the shelf

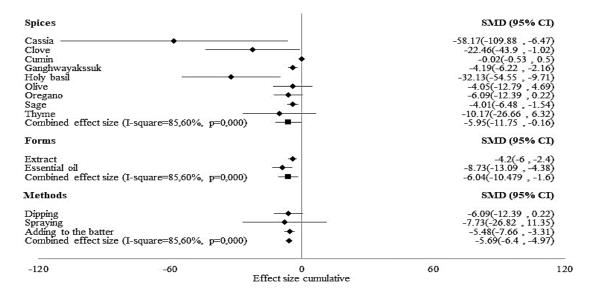


FIGURE 3. Forest plot of subgroup analysis of cumulative effect size and 95% confidence interval (CI) of essential oils and extract of spices on the TPC, extracted from 77 data

life of the products (Cegiełka et al. 2019; Hwang et al. 2013). Volatile organic compounds evaporate easily at room temperature, it is therefore expected that its use by spraying provides low effectiveness in comparison to the other methods (Ariyani, Setiawan & Soetaredjo 2008).

EFFECT OF ESSENTIAL OILS AND EXTRACTS OF SPICES ON Enterobacteriaceae AND COLIFORM

Based on the subgroup analysis, the type of spices and the method of application did not significantly affect the reduction of *Enterobacteriaceae* (Figure 4(A)). The

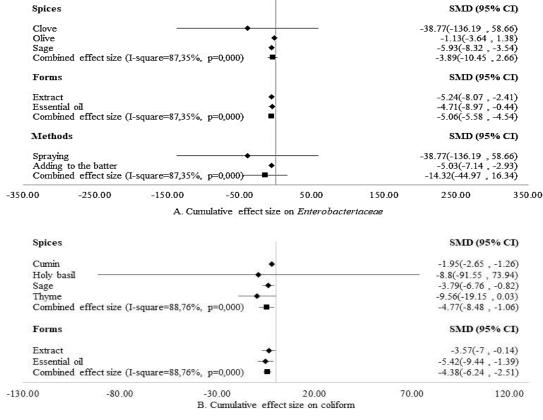


FIGURE 4. Forest plot of subgroup analysis of cumulative effect size and 95% confidence interval (CI) of essential oils and extracts of spices on (A) *Enterobacteriaceae* (35 data) and (B) coliform (32 data)

combined effect size of the type of spices on reducing *Enterobacteriaceae* was found with SMD= -3.89, 95%CI= -10.45 to 2.66, whereas the application method with SMD= -14.32, 95%CI= -44.97 to 16.34. However, sage demonstrated significant effect with SMD=-5.93 (95%CI=-8.32 to -3.54. Furthermore, the form of spices, as extracts as well as essential oils, reduced significantly the numbers of *Enterobacteriaceae* with SMD=-5.24 (95%CI=-8.07 to -2.41) and SMD=-4.7 (95%CI=-8.97 to -0.44), respectively.

Enterobacteriaceae is a large family of Gramnegative bacteria, including Escherichia, Shigella, and Salmonella (Darna et al. 2018; Octavia & Lan 2014). In a previous study, Sun et al. (2018) reported that the spices extracts (cinnamon and anise) reduced Enterobacteriaceae on dry pork sausage. The antimicrobial activity of spices extract is likely derived from the phenolic and flavonoid compounds that play a role in disrupting membranes and chelating metal on microorganisms (Zhang, Wu & Guo 2016).

The effect of different types of spices and their forms (essential oils or extracts) on the coliform of meat/poultry products is presented in Figure 4(B). Based on the forest plot subgroup, the use of various spices and their forms showed a significant effect on the reduction of coliform in meat/poultry products. Sage showed the biggest and most significant effect size on coliform reduction with SMD=-3.79 (95%CI= -6.76 to -0.82.) Previous results were reported by Veličković et al. (2003) for *in vitro* evaluation and by Karpin'skatymoszczyk (2007) on turkey sausage, showing that the

use of sage extract reduced the coliform loads. In the form of essential oil and extract, this plant is dominated by monoterpene compounds such as cineole, borneol, and thujone that can act as antimicrobials on Gram-negative and Gram-positive bacteria (Palareti et al. 2016). Since among the studies only used one application method, namely by adding to the batter, the application method was not included in the forest plot.

EFFECT OF ESSENTIAL OILS AND EXTRACTS OF SPICES ON PATHOGENIC BACTERIA

The pathogenic bacteria that were involved in this study were L. monocytogenes, S. aureus, and Salmonella, as shown in Figure 5. The combined effect size against pathogenic bacteria as subgroup, indicated that no significantly effect was found (SMD= -4.45, 95%CI = -13.28 to 4.37, $I^2 = 77.50\%$, and p = 0.000). However, Salmonella was significantly affected with SMD= -19.68 and 95%CI = -39.01 to -0.35. Likewise, the combined effect size of spices on pathogenic bacteria did not show significant effect (SMD=-3.17, 95%CI = -6.51 to 0.17, $I^2 = 77.50\%$, and p = 0.000), but oregano exhibited a significant effect with SMD= -7.63 (95%CI= -10.56 to -4.71). On the other hands, the application methods showed a significant effect on reducing pathogenic bacteria as indicated with SMD= -3.85, 95%CI= -6.51 to -1.19, $I^2 = 77.50\%$, and p = 0.000. The dipping method was found as the best method with SMD=-6.17 (95%CI= -8.53 to -3.81). In the forest plot, the form of application was not included, because among the studies only used the essential oils.

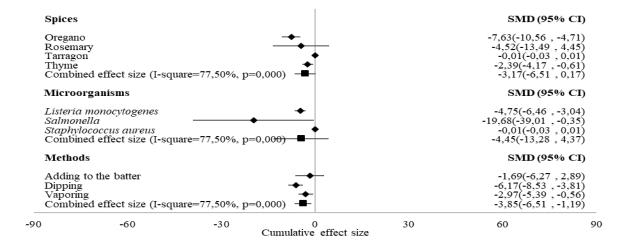


FIGURE 5. Forest plot of subgroup analysis of cumulative effect size and 95% confidence interval (CI) of spices on pathogenic bacteria, extracted from 32 data

In a previous study, the essential oil of oregano showed bactericidal effect against *Salmonella* Enteritidis on mince sheep meat (Govaris et al. 2010). Oregano (*Oreganum vulgare*) is an aromatic herb from the Lamiaceae family which is generally used to preserve food such as fresh chicken meat. The main components of oregano as essential oil were carvacrol, thymol, γ -terpinene, and β -Fenchyl. The presence of carvacrol and thymol act as an antibacterial activity to inhibit pathogenic bacteria such as *L. monocytogenes* and *E. coli* (Teixeira et al. 2013).

Based on the application methods, adding to the batter had no significant effect. Instead, dipping the product showed significant effect on reducing pathogenic bacteria in meat/poultry products. The dipping into (thyme) essential oils was effective in reducing *Salmonella* on chicken breast meat and wings (Thanissery & Smith 2014).

In contrast to *Salmonella*, the spices showed no significant cumulative effect on *S. aureus* (SMD=-0.01, 95%CI=-0.03 to 0.01). This result was different from some previous studies which showed significant effect against *S. aureus* (García-Díez et al. 2017; Hernández-Ochoa et al. 2012; Semeniuc, Pop & Rotar 2017). No cumulative effect size that was found in this study was likely affected by the use of limited data that was met the inclusion criteria, with SMD crossing the null effect size line (x=0) (Higgins & Green 2008).

CONCLUSION

The results of this meta-analysis indicated that the use of spices as essential oils was better than the extract on enhancing the microbiological quality of raw and processed meat or poultry products. The cumulative significant effect of essential oils in reducing bacterial loads of meat/poultry products showed a lower standardized mean difference (-4,37) than that showed by the extracts (-3.66). As essential oils, cassia showed the best significant effects in reducing microorganisms, followed by clove and holy basil, whereas as extracts, significant effects were showed by ganghwayassuk followed by sage. Furthermore, dipping is the best application in reducing microorganisms when essential oils were used, while as extracts, adding into products was the best method. Further identical meta-analysis may be best applicable for evaluating the use of spices on enhancing microbiological quality and safety of the other commodities.

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REFERENCES

- Abdel-Wahab, M., El-Sohaimy, S.A., Ibrahim, H.A. & El-Makarem, H.S.A. 2020. Evaluation the efficacy of clove, sage, and kiwifruit peels extracts as natural preservatives for fish fingers. *Annals of Agricultural Sciences* 65(1): 98-106.
- Afandi, F.A., Wijaya, C.H., Faridah, D.N., Suyatma, N.E. & Jayanegara, A. 2021. Evaluation of various starchy foods: A systematic review and meta-analysis on chemical properties affecting the glycemic index values based on *in vitro* and *in vivo* experiments. *Foods* 10(2): 1-28.
- Amor, G., Porta, R., Sabbah, M., Caputo, L., Idbella, M., de Feo, V., Fechtali, T. & Mauriello, G. 2021. Basil essential oil: Composition, antimicrobial properties, and microencapsulation to produce active chitosan films for food packaging. *Foods* 10(121): 1-16.
- Angelina, M., Turnip, M. & Khotimah, S. 2015. Antibacterial activity test of basil leaf ethanol extract (*Ocimum sanctum* L.) against *Escherichia coli* and *Staphylococcus aureus* bacterial growth. *Protobiont* 4(1): 184-189.
- Ariyani, F., Setiawan, L.E. & Soetaredjo, F.E. 2008. Extraction of essential oils from lemongrass plants with methanol, acetone, and n-hexane solvents. *Journal of Widya Engineering* 7(2): 124-133.
- Bortolotto, F.C.K., Ceccoti, S.P.C., Orso, P.B., Wolupeck, H.L., Holley, R.A., Luciano, F.B. & de Macedo, R.E.F. 2018. Combined effects of garlic essential oil and allyl isothiocyanate against *Escherichia coli* O157:H7 *in vitro* and in pork sausage. *Ciência Rural* 48(10): 1-7.
- Cegiełka, A., Hać-Szymańczuk, E., Piwowarek, K., Dasiewicz, K., Słowiński, M. & Wrońska, K. 2019. The use of bioactive properties of sage preparations to improve the storage stability of low-pressure mechanically separated meat from chickens. *Poultry Science* 98(10): 5045-5053.
- Cortés-Rojas, D.F., de Souza, C.R.F. & Oliveira, W.P. 2014. Clove (*Syzygium aromaticum*): A precious spice. *Asian Pacific Journal of Tropical Biomedicine* 4(2): 90-96.
- Darna, Turnip, M. & Rahmawati. 2018. Identification of Enterobacteriaceae members in traditional foods of pangkong cuttlefish. Labora Medika Journal 2(2): 6-12.
- de Melo, A.N.F., de Souza, G.T., Schaffner, D., de Oliveira, T.C.M., Maciel, J.F., de Souza, E.L. & Magnani, M. 2017. Changes in thermo-tolerance and survival under simulated gastrointestinal conditions of *Salmonella enteritidis* PT4 and *Salmonella typhimurium* PT4 in chicken breast meat after exposure to sequential stresses. *International Journal* of Food Microbiology 251: 15-23.

- de Oliveira, M.M.M., Brugnera, D.F. & Piccoli, R.H. 2013. Essential oils of thyme and rosemary in the control of *Listeria monocytogenes* in raw beef. *Brazilian Journal of Microbiology* 44(4): 1181-1188.
- Dewi, E.S., EL. Latifa, S., Fawwarahly. & Kautsar, R. 2016. Kualitas mikrobiologis daging unggas di RPA dan yang beredar di pasaran. *Jurnal Ilmu Produksi dan Teknologi Hasil Peternakan* 4(3): 379-385.
- Fachriyah, E., Wibawa, P.J. & Awaliyah, A. 2020. Antibacterial activity of basil oil (*Ocimum basilicum* L.) and basil oil nanoemulsion. *Journal of Physics Conference Series* 1524(1): 1-11.
- Gao, Y., Zhuang, H., Yeh, H.Y., Bowker, B. & Zhang, J. 2019. Effect of rosemary extract on microbial growth, pH, color, and lipid oxidation in cold plasma-processed ground chicken patties. *Innovative Food Science and Emerging Technologies* 57: 102168.
- García-Díez, J., Alheiro, J., Pinto, A.L., Falco, V., Fraqueza, M.J. & Patarata, L. 2017. Synergistic activity of essential oils from herbs and spices used on meat products against food borne pathogens. *Natural Product Communications* 12(2): 281-286.
- Ghorbani, A. & Esmaeilizadeh, M. 2017. Pharmacological properties of Salvia officinalis and its components. Journal of Traditional and Complementary Medicine 7(4): 433-440
- Govaris, A., Solomakos, N., Pexara, A. & Chatzopoulou, P.S. 2010. The antimicrobial effect of oregano essential oil, nisin and their combination against *Salmonella enteritidis* in minced sheep meat during refrigerated storage. *International Journal of Food Microbiology* 137(2-3): 175-180.
- Gurevitch, J., Koricheva, J., Nakagawa, S. & Stewart, G. 2018. Meta-analysis and the science of research synthesis. *Nature* 555(7695): 175-182.
- Haro-González, J.N., Castillo-Herrera, G.A., Martínez-Velázquez, M. & Espinosa-Andrews, H. 2021. Clove essential oil (*Syzygium aromaticum* L. myrtaceae): Extraction, chemical composition, food applications, and essential bioactivity for human health. *Molecules* 26(21): 6387.
- Hernández-Ochoa, L., MacÍas-Castañaeda, C.A., Nevárez-Moorillón, G.V., Salas-Muñaoz, E. & Sandoval-Salas, F. 2012. Antimicrobial activity of chitosan-based films including spices' essential oils and functional extracts. CyTA Journal of Food 10(2): 85-91.
- Hernando, D., Septinova, D. & Adhianto, K. 2015. Water content and microbial quality of the meat in Bandar Lampung abattoirs. *JIPT* 3(1): 61-68.
- Higgins, J.P.T. & Green, S. 2008. Cochrane Handbook for Systematic Reviews of Interventions: Cochrane Book Series. Chichester: John Wiley & Sons Ltd.
- Huang, D.F., Xu, J.G., Liu, J.X., Zhang, H. & Hu, Q.P. 2014. Chemical constituents, antibacterial activity and mechanism of action of the essential oil from *Cinnamomum cassia* bark against four food-related bacteria. *Microbiology* 83(4): 357-365.

- Hwang, K.E., Kim, H.W., Choi, Y.S., Lee, S.Y., Yeo, E.J., Ham, Y.K., Choi, S.M., Lee, M.A. & Kim, C.J. 2013. Evaluation of the antioxidant effect of ganghwayakssuk (*Artemisia princes* Pamp.) extract alone and in combination with ascorbic acid in raw chicken patties. *Poultry Science* 92(12): 3244-3250.
- Hwang, K.E., Choi, Y.S., Choi, J.H., Kim, H.Y., Kim, H.W., Lee, M.A., Chung, H.K. & Kim, C.J. 2011. The antioxidative properties of ganghwayakssuk (*Artemisia princeps* Pamp.) extracts added to refrigerated raw chicken nugget batter against lipid oxidation. *Korean Journal for Food Science* of *Animal Resources* 31(2): 166-175.
- Jaggi, R.K., Madaan, R. & Singh, B. 2003. Anticonvulsant potential of holy basil, *Ocimum sanctum* Linn., and its cultures. *Indian Journal of Experimental Biology* 41(11): 1329-1333.
- Jahan, N., Naveed, S., Zeshan, M. & Tahir, M.A. 2016. How to conduct a systematic review: A narrative literature review. *Cureus* 8(11): 1-8.
- Jayanegara, A., Wina, E. & Takahashi, J. 2014. Meta-analysis on methane mitigating properties of saponin-rich sources in the Rumen: Influence of addition levels and plant sources. *Asian-Australasian Journal of Animal Sciences* 27(10): 1426-1435.
- Kacaniova, M., Galovicova, L., Valkova, V., Tvrda, E., Terentjeva, M., Ziarovska, J., Kunova, S., Savitskaya, T., Grinshpan, D., Stefanikova, J., Felsöciova, S., Vukovic, N. & Kowalczewski, P.L. 2021. Antimicrobial and antioxidant activities of *Cinnamomum cassia* essential oil and its application in food preservation. *Open Chemistry* 19(1): 214-227.
- Kammon, A., Almaeyoufi, A. & Asheg, A. 2019. *In vitro* antimicrobial activity of clove oil against Gram negative bacteria isolated from chickens. *Approaches in Poultry, Dairy & Veterinary Sciences* 6(2): 542-546.
- Karpin'ska-tymoszczyk, M. 2007. Effect of sage extract (Salvia officinalis L.) mixture of sage extract and sodium isoascorbate on the quality and shelf life of vacuum-packed turkey meatballs. Journal of Muscle Foods 18(4): 420-
- Khanjari, A., Karabagias, I.K. & Kontominas, M.G. 2013. Combined effect of N, O-carboxymethyl chitosan and oregano essential oil to extend shelf life and control *Listeria* monocytogenes in raw chicken meat fillets. Food Science and Technology 53(1): 94-99.
- Kumar, S., Mendiratta, S.K., Agrawal, R.K., Sharma, H. & Singh, B.P. 2018. Anti-oxidant and antimicrobial properties of mutton nuggets incorporated with blends of essential oils. *Journal of Food Science and Technology* 55(2): 821-832.
- Kumudavally, K.V., Tabassum, A., Radhakrishna, K. & Bawa, A.S. 2011. Effect of ethanolic extract of clove on the keeping quality of fresh mutton during storage at ambient temperature (25±2 °C). Journal of Food Science and Technology 48(4): 466-471.

- Macwan, S.R., Dabhi, B.K., Aparnathi, K.D. & Prajapati, J.B. 2016. Essential oils of herbs and spices: Their antimicrobial activity and application in preservation of food. *International Journal of Current Microbiology and* Applied Sciences 5(5): 885-901.
- Martínez, L., Ros, G. & Nieto, G. 2020. Effect of natural extracts obtained from food industry by-products on nutritional quality and shelf life of chicken nuggets enriched with organic Zn and Se provided in broiler diet. *Poultry Science* 99(3): 1491-1501.
- Mehdizadeh, T., Tajik, H., Langroodi, A.M., Molaei, R. & Mahmoudian, A. 2020. Chitosan-starch film containing pomegranate peel extract and *Thymus kotschyanus* essential oil can prolong the shelf life of beef. *Meat Science* 163: 108073.
- Mittal, R., Kumar, R. & Chahal, H. 2018. Antimicrobial activity of *Ocimum sanctum* leaves extracts and oil. *Journal of Drug Delivery and Therapeutics* 8(6): 201-204.
- Mitic-Culafic, D., Vukovic-Gacic, B., Knezevic-Vukcevic, J., Stankovic, S. & Simic, D. 2005. Comparative study on the antibacterial activity of volatiles from sage (Salvia officinalis L.). Archives of Biological Sciences 57(3): 173-178
- Moghimi, R., Aliahmadi, A., McClements, D.J. & Rafati, H. 2016. Investigations of the effectiveness of nanoemulsions from sage oil as antibacterial agents on some food borne pathogens. *Food Science and Technology* 71: 69-76.
- Moher, D., Liberati, A., Tetzlaff, J., Altman, D.G., Altman, D., Antes, G., Atkins, D., Barbour, V., Barrowman, N., Berlin, J.A., Clark, J., Clarke, M., Cook, D., D'Amico, R., Deeks, J.J., Devereaux, P.J., Dickersin, K., Egger, M., Ernst, E., Gøtzsche, P.C., Grimshaw, J., Guyatt, G., Higgins, J., Ioannidis, J.P.A., Kleijnen, J., Lang, T., Magrini, N., McNamee, D., Moja, L., Mulrow, C., Napoli, M., Oxman, A., Pham, B., Rennie, D., Sampson, M., Schulz, K.F. Shekelle, P.G., Tovey, D. & Tugwell, P. 2009. Preferred reporting items for systematic reviews and meta-analyses: The PRISMA statement. *PLoS Medicine* 6(7): 1-6.
- Mukhtarini. 2014. Extraction, separation of compounds, and identification of active compounds. *Jurnal Kesehatan* 8(2): 361-367.
- Nugboon, K. & Intarapichet, K. 2015. Antioxidant and antibacterial activities of thai culinary herb and spice extracts, and application in pork meatballs. *International Food Research Journal* 22(5): 1788-1800.
- Octavia, S. & Lan, R. 2014. The family *Enterobacteriaceae*. In *The Prokaryotes*. 4th ed., edited by Rosenberg, E., DeLong, E.F., Lory, S., Stackebrandt, E. & Thompson, F. German: Springer Berlin.
- Ouahida, B., Amina, B. & Halima, M.I. 2021. Antibacterial activity of sage leaves against pathogenic bacteria as affected by different drying temperature. *Arabian Journal of Medicinal & Aromatic Plants* 7(1): 74-92.

- Palareti, G., Legnani, C., Cosmi, B., Antonucci, E., Erba, N., Poli, D., Testa, S. & Tosetto, A. 2016. Comparison between different D-Dimer cutoff values to assess the individual risk of recurrent venous thromboembolism: Analysis of results obtained in the DULCIS study. *International Journal of Laboratory Hematology* 38(1): 42-49.
- Palupi, E., Jayanegara, A., Ploeger, A. & Kahl, J. 2012. Comparison of nutritional quality between conventional and organic dairy products: A meta-analysis. *Journal of* the Science of Food and Agriculture 92(14): 2774-2781.
- Pattanayak, P., Behera, P., Das, D. & Panda, S. 2010. Ocimum sanctum Linn. A reservoir plant for therapeutic applications: An overview. Pharmacognosy Reviews 4(7): 95-105.
- Pedrós-Garrido, S., Clemente, I., Calanche, J.B., Condón-Abanto, S., Beltrán, J.A., Lyng, J.G., Brunton, N., Bolton, D. & Whyte, P. 2020. Antimicrobial activity of natural compounds against *Listeria* spp. and their effects on sensory attributes in salmon (*Salmo salar*) and cod (*Gadus morhua*). Food Control 107: 106768.
- Prasetya, N.B.A. & Ngadiwiyana, N. 2006. Identification of compounds compounding cinnamon bark oil (*Cinnamomum cassia*) using GC-MS. *Journal of Scientific & Applied Chemistry* 9(3): 81-83.
- Sarıçoban, C. & Yilmaz, M.T. 2014. Effect of thyme cumin essential oils and butylated hydroxyl anisole butylated. *Poultry Science* 93: 456-463.
- Semeniuc, C.A., Pop, C.R. & Rotar, A.M. 2017. Antibacterial activity and interactions of plant essential oil combinations against Gram-positive and Gram-negative bacteria. *Journal of Food and Drug Analysis* 25(2): 403-408.
- Shan, B., Cai, Y.Z., Brooks, J.D. & Corke, H. 2009. Antibacterial and antioxidant effects of five spice and herb extracts as natural preservatives of raw pork. *Journal of the Science of Food and Agriculture* 89(11): 1879-1885.
- Sharafati-Chaleshtori, R., Rokni, N., Rafieian-Kopaei, M., Drees, F., Sharafati-Chaleshtori, A. & Salehi, E. 2014. Use of Tarragon (*Artemisia dracunculus*) essential oil as a natural preservative in beef burger. *Italian Journal of Food Science* 26(4): 427-432.
- Sharma, H., Mendiratta, S.K., Agrawal, R.K., Talukder, S. & Kumar, S. 2018. Studies on the potential application of various blends of essential oils as antioxidant and antimicrobial preservatives in emulsion based chicken sausages. *British Food Journal* 120(7): 1398-1411.
- Sharma, H., Mendiratta, S.K., Agarwal, R.K., Kumar, S. & Soni, A. 2017. Evaluation of anti-oxidant and antimicrobial activity of various essential oils in fresh chicken sausages. *Journal of Food Science and Technology* 54(2): 279-292.
- Sirocchi, V., Devlieghere, F., Peelman, N., Sagratini, G., Maggi, F., Vittori, S. & Ragaert, P. 2017. Effect of *Rosmarinus officinalis* L. essential oil combined with different packaging conditions to extend the shelf life of refrigerated beef meat. *Food Chemistry* 221: 1069-1076.

- Šojić, B., Tomović, V., Savanović, J., Kocić-Tanackov, S., Pavlić, B., Jokanović, M., Milidrag, A., Martinović, A., Vujadinović, D. & Vukić, M. 2021. Sage (*Salvia officinalis* L.) essential oil as a potential replacement for sodium nitrite in dry fermented sausages. *Processes* 9(3): 1-15.
- Sun, Q., Zhao, X., Chen, H., Zhang, C. & Kong, B. 2018. Impact of spice extracts on the formation of biogenic amines and the physicochemical, microbiological and sensory quality of dry sausage. *Food Control* 92: 190-200.
- Tawfik, G.M., Dila, K.A.S., Mohamed, M.Y.F., Tam, D.N.H., Kien, N.D., Ahmed, A.M. & Huy, N.T. 2019. A step by step guide for conducting a systematic review and metaanalysis with simulation data. *Tropical Medicine and Health* 47(1): 1-9.
- Teixeira, B., Marques, A., Ramos, C., Serrano, C., Matos, O., Neng, N.R., Nogueira, J.M.F., Saraiva, J.A. & Nunes, M.L. 2013. Chemical composition and bioactivity of different oregano (*Origanum vulgare*) extracts and essential oil. *Journal of the Science of Food and Agriculture* 93(11): 2707-2714.
- Thanissery, R. & Smith, D.P. 2014. Marinade with thyme and orange oils reduces *Salmonella enteritidis* and *Campylobacter coli* on inoculated broiler breast fillets and whole wings. *Poultry Science* 93(5): 1258-1262.
- Toda, S. 2005. Antioxidative effects of polyphenols from leaves of *Artemisia princeps* Pamp. on lipid peroxidation *in vitro*. *Journal of Food Biochemistry* 29(3): 305-312.
- Trendafilova, A., Moujir, L.M., Sousa, P.M.C. & Seca, A.M.L. 2021. Research advances on health effects of edible *Artemisia* species and some sesquiterpene lactones constituents. *Foods* 10(1): 1-38.
- Triki, M., Herrero, A.M., Jiménez-Colmenero, F. & Ruiz-Capillas, C. 2013. Storage stability of low-fat sodium reduced fresh merguez sausage prepared with olive oil in konjac gel matrix. *Meat Science* 94(4): 438-446.

- Umano, K., Hagi, Y., Nakahara, K., Shoji, A. & Shibamoto, T. 2000. Volatile chemicals identified in extracts from leaves of Japanese mugwort (*Artemisia princeps Pamp.*). *Journal* of Agricultural and Food Chemistry 48(8): 3463-3469.
- USDA. 2021. Food Safety Information: Recalls & Public Health Alerts. www.fsis.usda.gov/recalls.
- Valdivieso-Ugarte, M., Gomez-Llorente, C., Plaza-Díaz, J. & Gil, Á. 2019. Antimicrobial, antioxidant, and immunomodulatory properties of essential oils: A systematic review. *Nutrients* 11(11): 1-29.
- Van Haute, S., Raes, K., Devlieghere, F. & Sampers, I. 2017. Combined use of cinnamon essential oil and MAP/vacuum packaging to increase the microbial and sensorial shelf life of lean pork and salmon. *Food Packaging and Shelf Life* 12: 51-58.
- Veličković, D.T., Randjelović, N.V., Ristić, M.S., Veličković, A.S. & Šmelcerović, A.A. 2003. Chemical constituents and antimicrobial activity of the ethanol extracts obtained from the flower, leaf and stem of Salvia officinalis L. Journal of the Serbian Chemical Society 68(1): 17-24.
- Verloove, F. & Andeweg, R. 2020. *Artemisia princeps* L. (Asteraceae), an overlooked invasive Far Eastern weed in Western Europe. *Gorteria* 42(1): 1-18.
- Vrinda, M.K. & Garg, S.R. 2001. Inhibitory effect of clove oil on *Listeria monocytogenes* in meat and cheese. *Food Microbiology* 18(6): 647-650.
- Yamani, H.A., Pang, E.C., Mantri, N. & Deighton, M.A. 2016. Antimicrobial activity of Tulsi (*Ocimum tenuiflorum*) essential oil and their major constituents against three species of bacteria. *Frontiers in Microbiology* 7(681): 1-10.
- Zhang, H., Wu, J. & Guo, X. 2016. Effects of antimicrobial and antioxidant activities of spice extracts on raw chicken meat quality. *Food Science and Human Wellness* 5(1): 39-48.

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