Characteristics of Essential Oils of Apiaceae Family: Their Chemical Compositions, in vitro Properties and Effects on Broiler Production

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There has been an upsurge of interest in the phytobiotics coincident with the onset of the potential ban on the use of antibiotic growth promoters (AGPs) in the broiler industry and because many kinds of nutraceuticals play an important role in improving growth performance, feed efficiency, and gut health of broilers. In the previous years, significant biological activities of essential oils (EOs) belonging to phytobiotics were observed, including antibacterial, antifungal, antiviral, and antioxidant properties. We found new perspectives on the roles of EOs, particularly extracts from the Apiaceae family, which is one of the largest plant families, in potential replacement of AGPs, and on the chemical composition involved in regulating microorganism activity and oxidative damage. Furthermore, the positive effects of EOs on broiler production and the possible mechanisms inducing the involvement of gut health and growth performance have been studied.

Key words: Antibiotic growth promoters, Apiaceae, broilers, essential oils, growth performance, gut health

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Introduction

Antibiotic growth promoters (AGPs) have been used in the broiler industry for decades to improve production performance and to minimize morbidity and mortality (Zeng et al., 2015; Broom, 2018). However, the use of antibiotics in broiler production has raised problems in the human population due to bacterial resistance to the agents and
transmission via the food chain (Graham et al., 2009; Chowdhury et al., 2018a). Therefore, the use of AGPs in broilers has been prohibited in several countries. In 2006, the European Union imposed a complete ban on all AGPs. The USA is limiting AGP use and moving towards a significant reduction in general antibiotic usage (Salim et al., 2018). Thereafter, many countries have announced AGP restrictions (Goutard et al., 2017).

In broiler production, AGP supplementation improves body weight gain (BWG) and feed conversion ratio (FCR), indicating that the withdrawal of AGP may increase production costs (Cardinal et al., 2019). This expectation has compelled nutritionists and feed manufacturers to seek the most suitable alternatives to AGPs. Since the early 2000s, researchers have explored the potential of nutraceuticals, such as probiotics, prebiotics, symbiotics, organic acids, and phytobiotics as alternatives to AGPs (Sugiharto, 2016), and the volatile extracts from plant sources have been identified as a new class of phytopgenic feed additives (Zeng et al., 2015).

The volatile extracts obtained from different plant parts, such as flowers, buds, seeds, leaves, twigs, bark, herbs, wood, fruits, and roots by hydro/steam distillation, are referred to as EOs. EOs have been reported to have antibacterial, antifungal, antiviral, and antioxidant properties as biological actions that depend on their chemical constituents (Al Bayati, 2008). Attention to EOs as a replacement for AGPs in poultry has increased because of their positive effects on production performance (Sugiharto, 2016). However, the mode of action of EOs is yet to be fully elucidated (Zeng et al., 2015; Kikusato, 2021).

**Apiaceae** is one of the largest plant families (Pimenov and Leonov, 1993). Its plants have a characteristic pungent smell, whose extracts are EOs. Several constituents of EOs are believed to be precursors of biological compounds that exert beneficial effects on gut morphology, nutrient absorption, microbiota, and oxidative status. Therefore, the EOs extracted from the Apiaceae family have been considered as a possible replacement for AGPs in broiler production (Acimovic et al., 2016).

This review focuses on the characteristics of EOs, particularly the *in vitro* properties of EOs extracted from selected plants of the Apiaceae family, such as coriander (*Coriandrum sativum*), ajwain (*Trachyspermum ammi*), dill (*Anethum graveolens*), fennel (*Foeniculum vulgare*), and anise (*Pimpinella anisum*), and their effects on broiler production and possible machineries. Such an endeavor can never be truly comprehensive; however, this review aims to provide an awareness of the current state of the field for readers both inside and outside the phytobiotics community.

### 1. Chemical compositions and *in vitro* properties of selected essential oils

EOs are synthesized to protect the plant bodies against bacterial and fungal invasions and viruses and protect DNA and photosynthetic apparatus from the oxidative damage caused by ultraviolet radiation (Kikusato, 2021). Therefore, the EOs extracted from the plants of the Apiaceae family can perform various biological activities based on their chemical constituents. The relative concentration and overall yield of the constituents differ among plant types, parts, harvesting season, environmental conditions, soil type, storage conditions, and types of processing (Applegate et al., 2010; Grashorn, 2010; Kiczorowska et al., 2015; Al Yasiry and Kiczorowska, 2016). Most of the published literature describing *in vitro* antibacterial and antifungal properties has focused on the microbial species relevant to food pathogenesis; however, data regarding bacterial species that may influence the intestinal circumstances of broilers are lacking.

In this section, many measurement units are described as used in the literature: minimum inhibitory concentration (MIC) and/or zone of inhibition (ZOI) for antibacterial activity of the EOs. In addition, inhibition of 2,2-diphenyl-1-picrylhydrazyl (DPPH), ferric reducing antioxidant power (FRAP) assay, and Trolox equivalent antioxidant capacity (TEAC) using 2,2'-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid (ABTS), peroxide value (PV), thiobarbituric acid value (TBA), and antioxidant activity in the linoleic acid system are used for antioxidant activity.

#### 1.1. Coriander essential oil (CEO): Table 1

**A. Chemical compositions**

Coriander (*Coriandrum sativum*) is a glabrous, aromatic, and herbaceous annual plant with culinary applications and serves as a source of aroma compounds and EO. Coriander seeds contain 0.03 to 2.6% EO, with linalool being the main chemical constituent (Acimovic et al., 2016; Jeya et al., 2019). Table 1 shows the chemical composition, area of cultivation, extraction method, and yield of CEO from the selected studies. Linalool (Figure 1-1) was the major component of the CEO with a share of 66.3–75.3% of the total composition, whereas α-pinene, γ-terpinene, camphor, geranyl acetate, and cymene were the other major components (Baratta et al., 1998; Delaquis et al., 2002; Singh et al., 2006; Kacaniova et al., 2020). Singh et al. (2006) reported the presence of more than 52 chemical compounds in CEO.

**B. In vitro properties**

#### a) Antibacterial activity

Many studies have shown that the chemical constituents present in CEO have antibacterial properties. Baratta et al. (1998) analyzed the CEO (10 μL/disk) against 25 different bacteria, and the reported ZOI for *Bacillus subtilis*, *Clostridium perfringens*, *Escherichia coli*, *Salmonella pullorum*, and *Staphylococcus* (*Staph.*) *aureus* were 8.5, 4, 6.5, 7.6, and 16.1 mm, respectively. Kacaniova et al. (2020) reported that the ZOI of CEO (10 μL/disk) against *B. subtilis* was 10.7 mm. Delaquis et al. (2002) demonstrated that CEO had antibacterial activity against *E. coli*, *Listeria monocytogenes*, and *Staph. aureus* with MIC 0.2, 0.5, and 0.4 mL/L (% vol/vol), respectively, except for *C. perfringens*. In a recent study, Jeya et al. (2019) reported 0.64 mg/mL as the MIC of CEO against *E. coli*.

#### b) Antifungal activity

The CEO can effectively inhibit the growth of *Aspergillus niger* (inhibition index: 94.8%) at 1 μL/mL concentration (Baratta et al., 1998). Singh et al.
(2006) evaluated the CEO (10 μL) against different fungi and reported good ZOI (more than 70%) against *Curvularia palliscens*, *Fusarium moniliforme*, and *A. terreus*. In addition, Jeya *et al.* (2019) reported that the CEO showed fungicidal effects against *Candida (Can.) albicans* with a MIC of 0.02 mg/mL.

c) Antioxidant activity: The CEO contains natural antioxidants that can prevent or delay the effects of oxidation processes. Baratta *et al.* (1998) analyzed the antioxidant effectiveness of CEO through the modified thiobarbituric acid reactive species (TBARS) assay using two materials rich in lipids as oxidable substrates (egg yolk and rat liver). The results demonstrated that the CEO at 1000 ppm in rat liver exhibited a higher antioxidant index than synthetic antioxidants, α-tocopherol, and butylated hydroxytoluene (BHT) at the same supplementation levels. Singh *et al.* (2006) evaluated the antioxidant capacity of CEO by PV, TBA, and antioxidant activity in the linoleic acid system, revealing that 200 ppm CEO supplementation resulted in a 21% reduction in PV during storage at 80°C for 28 days. Kacaniova *et al.* (2020) analyzed the radical scavenging activity of the CEO using the DPPH test and Trolox (vitamin E analog) as the standard, showing that 25 μL/mL CEO has 51.1% inhibition efficiency for scavenging free radicals. Moreover, Shahwar *et al.* (2012) and Singh *et al.* (2015) performed the radical scavenging activity of CEO at 500 μg/mL and 50 μL/mL.
using the DPPH test and reported 66.5% and 54.6% inhibition in DPPH-derived free radicals, respectively.

### Table 1. Chemical Compositions and in vitro properties of CEO (Coriander Essential Oil) (continued)

| Chemical Composition | Baratta et al., 1998 | Delaquis et al., 2002 | Singh et al., 2006 | Kacaniova et al., 2020 |
|----------------------|----------------------|-----------------------|--------------------|------------------------|
| Antibacterial activity | Species | ZOI (mm) 10 μL/disk | Species | MIC (mL/dL) | Species | ZOI (mm) 10 μL/disk |
| B. subtilis | 8.5 | | | | B. subtilis | 10.7 |
| C. perfringens | 4.0 | L. monocytogenes | 0.5 | | S. maltophilia | 9.2 |
| E. coli | 6.5 | E. coli | 0.2 | | | |
| S. pullorum | 7.6 | S. typhi | No inhibition observed | | | |
| Staph. aureus | 16.1 | Staph. aureus | 0.4 | | | |
| Antifungal activity | Species | % Inhibition index 1 μL/mL | Species | % ZOI 10 μL |
| A. niger | 94.8 | | A. flavus | 31.3 (75% by FPT) |
| A. terreus | 75 (100% by FPT) | | A. niger | 37.5 (100% by FPT) |
| Antioxidant activity | Method | Effects | Method | Effects | Method | Effects |
| Antioxidant index (AI%) using TBARS assay | higher than BHT at 1000 ppm | | Peroxide value (PV) method | PV 248 meq/kg of sunflower oil was reduced to 196 meq/kg during storage at 80°C for 28 days at 200 ppm dose of CEO | DPPH | CEO radical scavenging activity was 39.4 mg TEAC/L (Trolox equivalent antioxidant activity) equivalent to 51.1% of inhibition |
| TBA value | TBA value 4 meq/kg of sunflower was reduced to 2.5 meq/kg during storage at 80°C for 21 days by 200 ppm dose of CEO | | | | |

ZOI = zone of inhibition, MIC = minimum inhibitory concentration, FPT = food poison technique, S. maltophilia = Stenotrophomonas maltophilia

1Mycelial inhibition zone (%) at dose 10 μl by inverted petri plate method

### 1.2. Ajwain essential oil (AjEO: Table 2)

### A. Chemical compositions

Ajwain (Trachyspermum ammi) is an important plant with spice, aromatic, and medicinal properties. It originated in
Egypt and is found worldwide. Ajwain seeds contain 2%-5% EO, with thymol (Fig. 1-2) as a major bioactive compound with a share of 39.1-67.4% of the total composition, followed by p-cymene, γ-terpinene, β-pinene, carvacrol, α-phellandrene, β-phellandrene, α-terpinene, α-pinene, and sabinene (Singh et al., 2004; Vitali et al., 2016; Gradinaru et al., 2018). However, Patil et al. (2016) reported that p-cymene (15.6%) was the major component in AjEO, followed by thymol (15.5%), by analyzing the peak area percentage of GC/MS results.

B. In vitro properties
a) Antibacterial activity: The MIC of AjEO against *Staph. aureus* and *E. coli* were 500 μg/mL (Vitali et al., 2016). However, Paul et al. (2011) showed stronger antibacterial activity against gram-positive bacteria than against gram-negative bacteria. The MIC of AjEO against *Streptococcus*
(Strep.) mutans, E. coli, S. typhi, S. parathyphi, P. vulgaris, and P. aeruginosa was 12.5 μL/mL (Patil et al., 2016). Considering the composition of AjEO, thymol may be the main component to induce antibacterial activity. In a recent study, Gradinaru et al. (2018) revealed that AjEO has the potential to limit the growth of respiratory pathogens (Staph. aureus, Strept. pneumoniae, P. aeruginosa) and discovered the combined effects of AjEO/thymol and conventional antibiotics against multidrug-resistant respiratory pathogens.

b) Antifungal activity: Singh et al. (2004) showed that the AjEO at 6 μL dose rate is 100% fungicidal for all the tested pathogenic fungal species. In contrast, Vitali et al. (2016) reported limited activity of AjEO against Can. albicans with a MIC of 500 μg/mL, which is 125 times higher than nystatin (reference anti-fungal drug).

c) Antioxidant activity: According to Singh et al. (2004), AjEO has good antioxidant properties, as analyzed by the PV, TBA, and linoleic acid system. Patil et al. (2016) demonstrated that AjEO is a strong antioxidant with 71.7% efficacy using the DPPH method, whereas the antioxidant activity of ascorbic acid (standard) was 20.2%. Vitali et al. (2016) evaluated the antioxidant properties of AjEO using DPPH, ABTS, and FRAP assays. The ability of AjEO to scavenge the different radicals in all assays was compared with Trolox (vitamin E analog) and expressed as TEAC. The results revealed that the AjEO showed good antioxidant activity as the TEAC of ABTS, FRAP, and DPPH assays were 266.7 μmol TE/g, 90.6 μmol TE/g, and 72.6 μmol TE/g, respectively. The free radical scavenging activities of AjEO in all the studies mentioned above proved its potential as a natural antioxidant substance, which can be used as an efficient antioxidant agent.

1.3. Dill essential oil (DEO: Table 3)

A. Chemical compositions

Dill (Anethum graveolens) is one of the most useful spices with medicinal properties. It is cultivated worldwide, and its EO has flavoring and medicinal effects. Dill seeds yield 2%-4.2% EO with carvone (Fig. 1–3) as a major chemical component with a share of 47.7–73.6% in total composition, followed by limonene (Fig. 1–4), dill apiole, and α-phellendrene (Singh et al., 2005; Yili et al., 2009; Chalah et al., 2017; Singh et al., 2017). In contrast to previous studies, Kazemi (2015) reported thymol (20.1%) as the major component of DEO, followed by limonene, α-pinene, and carveol. He justified that his results are in contrast with those of other studies because of the genetic, environmental, chemotypes, and nutritional status of the plants. Since the chemical composition of DEO varies considerably between different studies, more comprehensive studies on chemical constituents are required.

B. In vitro properties

a) Antibacterial properties: Singh et al. (2005) analyzed the antimicrobial activity of DEO against six pathogenic bacteria. They reported it as an effective antibacterial agent against P. aeruginosa and E. coli with ZOI 25.3 mm and 18.5 mm, respectively, although ineffective against S. typhi. DEO also showed effective antibacterial activity against Staph. aureus with MIC 0.27 mg/mL (Yili et al., 2009). According to Kazemi (2015), DEO performed best against E. coli at a MIC of 5 μg/mL. In contrast, the MIC for other tested bacteria (B. cereus, Enterococcus (En.) facealis, S. aureus, P. aerogenosa, and S. typhi) ranged between 10–40 μg/mL. In a recent study, DEO showed better inhibitory effects against gram-positive bacteria than gram-negative bacteria at 10 μL dose/disk (Singh et al., 2017). ZOI for B. subtilis, Staph. aureus, E. coli, and P. aerogenosa were 15.6, 20.3, 7.5, and 8.9 mm, respectively.

b) Antifungal activity: DEO has the potential to produce antifungal effects. It has shown 100% fungicidal activity for Penicillium (Pen.) citrinum and A. niger at 6 μL concentration out of eight tested pathogenic fungi. The activity against other fungi was also considerable (Singh et al., 2005). The Can. albican was also found to be very sensitive to DEO with a MIC value of 2.7 μg/mL (Yili et al., 2009). Kazemi (2015) reported the significant fungicidal effects of DEO against Can. albicans and A. fumigatus at MIC 10 and 20 μg/mL, respectively. Singh et al. (2017) reported the significant antifungal activity of DEO against five tested pathogenic fungi. Among the tested fungi, A. flavus was the most sensitive (more than 80% ZOI) to DEO at 10 μL, followed by the other tested fungi. More recently, ten Candida species were examined against DEO and found very significant fungicidal effects with a MIC of 8.75 mg/mL for all tested fungi (Vieira et al., 2019).

c) Antioxidant activity: Singh et al. (2005) evaluated the antioxidant properties of DEO by PV, TBA, and DPPH methods, revealing that 200 ppm DEO supplementation resulted in a 10.6% reduction in PV during storage at 80°C for 28 days. The TBA value of rapeseed oil was also reduced by approximately 50% during this storage period. Moreover, the radical scavenging activity of DEO by the DPPH method was 81.6% compared to butylated hydroxyanisole (BHA) (88.5%) and BHT (90.3%). Kazemi (2015) reported that the DPPH value of DEO (IC50 = 34.4 mg/mL) is comparable to that of Trolox (IC50 = 28.3 mg/mL), suggesting the antioxidant properties of this EO.

In a recent study, Singh et al. (2017) evaluated the antioxidant activity of DEO by PV, TBA, and DPPH methods. They proved that it is a good natural antioxidant, similar to commercial antioxidant products. Briefly, 200 ppm DEO supplementation in mustard oil resulted in a 45% reduction in PV during storage at 60°C for 28 days, and the TBA value was reduced by approximately 50% and 25% on the 21st and 28th day of storage, respectively, compared to the control group. Moreover, DEO showed 75% radical scavenging activity, which was higher than that of the other tested commercial antioxidants. The conclusion of the studies mentioned above indicated the presence of carvone, limonene, and dill apiole in DEO, which may be the main reason for the antioxidant properties.

1.4. Fennel essential oil (FEO: Table 4)

A. Chemical compositions

Fennel (Foeniculum vulgare) is one of the oldest spice plants with considerable medicinal properties. The fennel
contains 4%-6% EOs with more than 30 types of chemical constituents (Kooti et al., 2015). Trans-anethole (Fig. 1-5) was identified as a major component with a share of 56.4-69.9% in total composition, whereas fenchone, estragole, and limonene were the other main components (Anwar et al., 2009; Roby et al., 2013; Diao et al., 2014; Ilic et al., 2019).

**B. In vitro properties**

**a) Antibacterial activity:** According to Anwar et al. (2009), FEO showed considerable antibacterial activity against *B. subtilis* and *E. coli* with ZOI of 29 mm and 14 mm, respectively. Roby et al. (2013) demonstrated the antibacterial effects of FEO against gram-positive bacteria (*B. cereus*, *Staph. aureus*) and gram-negative (*E. coli*, *S. typhi*) bacteria with MICs ranging from 10 to 15 μg/mL. In another study, FEO showed antibacterial activity against *E. coli*, *B. subtilis*, and *S. typhi* at a MIC of 0.25 mg/mL; however, *Staph. aureus* and *P. aerogenosa* did not respond to it even at the highest tested concentration (10 mg/mL) (Diao et al., 2014). More recently, Ilic et al. (2019) reported that the *Can. albicans* was the most sensitive of the seven tested microorganisms in their study, with clear ZOI and 25 μg/mL MIC.

**b) Antifungal activity:** Several studies have reported the significant antifungal properties of FEO, as shown by its activity against various fungal species such as *Can. albicans*, *Aspergillus* species, and *dermatophytes* (Kooti et al., 2015). Anwar et al. (2009) reported FEO as an efficient antifungal against the three tested fungi, particularly *A. niger*, showing the highest sensitivity with 28 mm ZOI and 80.6 mg/mL a MIC value. In another study, *Can. albicans* and *A. flavus* were sensitive to FEO at a MIC of 10 μg/mL (Roby et al., 2013). More recently, Ilic et al. (2019) reported that the *Can. albicans* was the most sensitive of the seven tested microorganisms in their study, with clear ZOI and 25 μg/mL MIC.

**c) Antioxidant activity:** Limited data are available regarding the antioxidant properties of FEO; however, in some studies, it has been proven to be a strong antioxidant agent. Anwar et al. (2009) evaluated the antioxidant properties of FEO using DPPH assay. They concluded that it has good radical scavenging activity with IC50 = 32.32 μg/mL. Moreover, FEO can replace commercial synthetic antioxidants such as BHA and BHT, which are discouraged because of their perceived carcinogenic potential and safety concerns (Anwar et al., 2009).
1.5. Anise essential oil (AnEO: Table 5)

A. Chemical compositions

Anise (Pimpinella anisum) is an annual aromatic spice known for its medicinal and aromatic properties and is found worldwide. The fruit or seed of anise yields 2.1%-3.3% EO, and the important chemical components are trans-anethole (Fig. 1-5), methyl chavicol, and anisaldehyde (Arslan et al., 2004; Sharifi et al., 2008). Trans-anethole was identified as a major component with a share of 79-92.9% of the total composition, whereas estragole, 3,4-dimethoxyxystrene, α-gurjunene, and α-bisabolene were the other main components (Sharifi et al., 2008; Topal et al., 2008; Foroughi et al., 2016; Asadollahpoor et al., 2017). In contrast to the studies mentioned above, De Martino et al. (2009) reported a slightly different chemical composition of AnEO and stated that the major chemical constituent of this EO is cis-anethole (97.1%).

B. In vitro properties

a) Antibacterial activity: Al Bayati (2008) reported the moderate antibacterial activity of AnEO against nine pathogenic bacteria with MIC ranging from 62.5-500 μg/mL, where gram-positive bacteria were more sensitive than gram-negative bacteria. In another study, a wide range of gram-positive and gram-negative bacterial species were found to be sensitive to AnEO, with MICs ranging from 25-100 mg/mL (Al Maofari, 2013). More recently, Foroughi et al. (2016) confirmed the antibacterial effectiveness of AnEO against E. coli and Staph. aureus. The AnEO (31 mg/mL) performed better than the positive controls (kanamycin and cephealexin) in ZOI for E. coli and Staph. aureus. Moreover, the MICs for E. coli and Staph. aureus were 3 mg/mL and 7 mg/mL, respectively.

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**Table 2. Chemical Compositions and in vitro properties of AjEO (Ajwain Essential Oil) (continued)**

| Chemical Composition       | Singh et al., 2004 | Patil et al., 2016 | Vitali et al., 2016 | Gradinaru et al., 2018 |
|----------------------------|-------------------|-------------------|--------------------|------------------------|
| **Antibacterial activity** |                   |                   |                    |                        |
| Species                    | ZOI (mm)          | MIC (μL/mL)       | Species            | ZOI (mm)              | MIC (μg/mL)          | Species | MIC (mg/mL) |
| S. para-typhi A            | 52                | 12.5              | Staph. aureus      | 34.7                  | 500                 | Staph. aureus       | 4       |
| S. typhi                   | 54                | 12.5              | E. coli            | 29.3                  | 500                 |                     |         |
| E. coli                    | 66                | 12.5              |                     |                       |                     |                     |         |
| **Antifungal activity**    |                   |                   |                    |                        |
| Species                    | ZOI (%)           | MIC (μL/mL)       | Species            | ZOI (mm)              | MIC (μg/mL)          |                     |         |
| A. flavus                  | 100               |                   | Can. albicans      | 54.3                  | 500                 |                     |         |
| A. niger                   | 100               |                   |                     |                       |                     |                     |         |
| **Antioxidant activity**   |                   |                   |                    |                        |
| Method                     | Effects           | Method            | Effects            |                         |                     |
| PV method                  |                   | Strongest antioxi-
|                        |                   | dant activity (71.68%) noted at 1000 mg/L concentration and was three times greater than the effect produced by standard; ascorbic acid (20.24%) |
| DPPH                      |                   | ABTS              | AjEO showed strong antioxidant activity with 1C50=22.4 μg/mL and TEAC=266.7 μmol TE/g |
| TBA method                 |                   | FRAP              | AjEO showed anti-
|                        |                   |                   |    oxi-
|                        |                   |                   |    dant activity with TEAC=90.6 μmol TE/g |
| TBA value 3.8 and 5 mg/kg of linseed oil was reduced to 3.0 and 3.8 mg/kg at 80°C during storage for 21 and 28 days, respectively, by 200 ppm addition of EO | | |
| DPPH                      |                   |                   |                   |                         |                     |

ZOI=zone of inhibition, MIC=minimum inhibitory concentration, TEAC=Trolox equivalent antioxidant concentration

1 Concentration of compound that affords a 50% reduction in the assay
### Table 3. Chemical Compositions and in vitro properties of DEO (Dill Essential Oil)

| Chemical Composition | Singh et al., 2005 | Yili et al., 2009 | Kazemi, 2015 | Singh et al., 2017 |
|----------------------|--------------------|-------------------|--------------|-------------------|
| carvone              | 55.2               | 73.6              | 16.3         | 47.7              |
| limonene             | 16.6               | 14.7              | 12.4         | 14.7              |
| thymol               |                   |                   | 20.1         |                   |
| carvacrol            |                   |                   | 8.3          |                   |
| dill ether           | 0.2                |                   | 3.1          |                   |
| dill apiol           | 14.4               |                   | 32.7         |                   |
| α-pinene             | 0.1                |                   | 8.7          |                   |
| linalool             | 3.7                |                   |              |                   |
| trans-dihydrocarvone | 2.8                |                   |              | 2.7               |
| cis-dihydrocarvone   | 2.6                | 5.9               |              | 2.1               |
| α-phellandrene       | 0.03               | 2.4               | 1.3          |                   |
| sabinene             | 0.1                |                   | 1.0          |                   |
| β-pinene             | 0.1                |                   |              |                   |
| myrcene              | 0.1                |                   | 0.7          |                   |
| γ-terpenene          | 0.3                |                   |              |                   |
| terpinen-4-ol        | 0.1                |                   |              |                   |
| iso-dihydrocarveol   | 0.1                |                   |              |                   |
| cis-dihydrocarveol   | 0.1                | 0.2               |              |                   |
| trans-dihydrocarveol | 0.1                |                   |              |                   |
| geranyl acetate      | 0.3                |                   |              |                   |
| β-caryophylene       | 0.6                |                   |              |                   |
| β-bisabolene         | 0.3                |                   |              |                   |
| δ-cadinene           | 0.1                |                   |              |                   |
| trans-isocroweacin   | 0.8                |                   |              |                   |
| 1,2-diethoxyethane   |                   | 1.4               |              |                   |
| dihydrocarvone       |                   | 1.4               |              |                   |
| diplaniol            |                   | 2.2               |              |                   |
| α-thujene            |                   | 0.1               |              |                   |
| neophtadiene         |                   | 1.4               |              |                   |
| n-nonadecane         |                   | 1.0               |              |                   |
| n-eicosane           |                   | 0.9               |              |                   |
| n-heneicosane        |                   | 0.7               |              |                   |
| n-docosane           |                   | 1.0               |              |                   |
| n-tricosane          |                   | 1.0               |              |                   |
| n-tetracosane        |                   | 1.5               |              |                   |
| α-cymenene           |                   |                   | 0.2          |                   |
| menthol              |                   |                   | 0.7          |                   |
| myristicin           |                   |                   | 0.9          |                   |
| Cultivation/experimen- | India             | Uzbekistan       | Iran         | India             |
| tation area          |                    |                   |              |                   |
| Extraction method/source | Hydrodistillation  | Hydrodistillation | Hydrodistillation | Hydrodistillation |
| EO yield (%)         | 2.6                | 4.2               | 3.2          | 2.4               |
Table 3. Chemical Compositions and *in vitro* properties of DEO (Dill Essential Oil) (continued)

| Chemical Composition | Singh et al., 2005 | Yili et al., 2009 | Kazemi, 2015 | Singh et al., 2017 |
|----------------------|--------------------|-------------------|--------------|-------------------|
| Antibacterial activity | Species | ZOI (mm) 6 μL/disk | Species | MIC (μg/mL) | Species | MIC (μg/mL) | Species | ZOI (mm) 10 μL/disk |
| B. subtilis | 16.2 | | | | B. subtilis | 15.6 | |
| Staph. aureus | 13.2 | Staph. aureus | 0.27 | Staph. aureus | 20 | Staph. aureus | 20.3 | |
| S. typhi | No ZOI | S. typhi | 40 | | | |
| E. coli | 18.5 | E. coli | 5 | E. coli | 7.5 | | |
| P. aeruginosa | 25.3 | | | | P. aeruginosa | 8.9 | |
| Antifungal activity | Species | % ZOI 6 μL/disk | Species | MIC (μg/mL) | Species | MIC (μg/mL) | Species | % ZOI (FPT<sup>1</sup>) 10 μL |
| A. niger | 100 | | | | A. niger | 63.9 | |
| A. flavus | 82.5 | Can. albicans | 2.7 | A. fumigates | 20 | A. niger | 89.7 | |
| Pen. citrinum | 100 | | | | Pen. viridicatum | 17.6 | |
| Antioxidant activity | Method | Effects | Method | Effects | Method | Effects | Method | Effects |
| PV | PV 239.2 meq/Kg of rapeseed oil was reduced to 213.9 meq/Kg during storage at 80℃ for 28 days by 200 ppm addition of EO | FRAP | DEO = Antioxidant activity 301 μmol Fe<sup>2+</sup>/g EO, Trolox (standard) = 321 μmol Fe<sup>2+</sup>/g EO | PV | PV 181.8 meq/Kg of mustard oil was reduced to 100 meq/Kg during storage at 60℃ for 28 days by 200 ppm addition of EO |
| TBA value | TBA value 6.9 meq/kg of rapeseed oil was reduced to 3.4 meq/kg during storage at 80℃ for 28 days by 200 ppm addition of EO | NA | TBA value | TBA value 0.18 and 0.21 meq/kg of mustard oil was reduced to 0.092 and 0.16 meq/Kg during storage at 60℃ for 21 and 28 days, respectively, by 200 ppm addition of EO |
| DPPH | DEO showed 81.6% radical scavenging activity in comparison to BHA (88.5%) and BHT (90.3%) | DPPH | DEO scavenging activity IC<sub>50</sub> = 34.41 mg/mL, Trolox (standard) IC<sub>50</sub> = 28.32 mg/mL | DPPH | DEO showed 75% radical scavenging activity |

ZOI = zone of inhibition, MIC = minimum inhibitory concentration, FPT = food poison technique

<sup>1</sup> Concentration of compound that affords a 50% reduction in the assay
b) Antifungal activity: The antifungal activity of AnEO has been proven by many researchers. Elgayyar et al. (2001) reported the antifungal potential of AnEO against *A. niger* with a significant zone of growth inhibition. Ozcan and Chalchat (2006) proved that AnEO is an effective antifungal agent against *A. parasiticus*, *A. niger*, and *Alternaria alternata* at 10–100 ppm doses. In another study, AnEO was reported as an antifungal agent against *A. niger* with a MIC of 2000 μL/L and EC50 of 400 μL/L (half-maximal effective concentration) (Sharifi et al., 2008). In a study by Nanasombat and Wimuttigosol (2011), AnEO produced strong antifungal effects with clear ZOI against the six yeast and four mold species, and the MIC ranged from 1 mg/mL to 6 mg/mL for all the tested microbes.

c) Antioxidant activity: Many studies have proved the antioxidant activity of AnEO and its ability to be used as a replacement for commercial antioxidants. According to Singh et al. (2008), 200 ppm AnEO supplementation in mustard oil resulted in a 28% reduction in PV during storage for 28 days at 70°C, which obtained a better result than commercial antioxidants. In addition, the DPPH assay proved the stronger antioxidant activity of AnEO than BHA.

| Chemical Composition | Anwar et al., 2009 | Roby et al., 2013 | Diao et al., 2014 | Ilic et al., 2019 |
|----------------------|------------------|------------------|------------------|------------------|
| trans-anethole       | 69.9             | 56.4             | 68.5             | 64.9             |
| estragole            | 5.5              | 5.2              | 10.4             | 2.6              |
| limonene             | 5.1              | 4.2              | 6.2              | 2.3              |
| fenchone             | 10.2             | 8.3              | 5.5              | 23.1             |
| δ-3-carene           |                  |                  | 1.2              |                  |
| α-cymene             |                  |                  | 0.6              |                  |
| α-pinene             | 0.6              | 1.6              | 0.4              | 2.0              |
| methyl chavicol      |                  |                  | 5.2              |                  |
| β-farnesene          |                  |                  | 3.0              |                  |
| γ-terpinene          | 0.2              | 1.4              |                  | 0.7              |
| camphene             | 0.1              |                  | 0.2              |                  |
| sabinene             | 0.2              |                  | 0.1              |                  |
| β-pinene             | 0.1              |                  | 0.4              |                  |
| β-myrcene            | 0.9              | 0.6              | 0.2              | 1.0              |
| α-phellandrene       | 0.2              |                  | 0.1              | 0.4              |
| β-ocimene            | 0.6              |                  |                  |                  |
| 1,8-cineol           | 0.2              | 0.9              |                  |                  |
| fenchyl alcohol      | 0.4              |                  |                  |                  |
| fenchyl acetate      | 0.5              |                  | 0.1              |                  |
| cis-anethol          | 0.3              |                  | 0.5              | 0.1              |
| ρ-anisaldehyde       | 0.2              |                  | 0.3              | 0.1              |
| β-caryophyllene      | 0.3              |                  |                  |                  |
| germacrene           | 0.1              |                  | 0.2              |                  |
| α-terpinin           |                  |                  | 0.6              |                  |
| terpin-4-ol          |                  | 2.8              |                  |                  |
| myrcenol             |                  | 1.0              |                  |                  |
| bergamot             |                  | 0.6              |                  |                  |
| 2,5-diethyl phenol   |                  | 0.8              |                  |                  |
| β-farnesene          |                  | 3.0              |                  |                  |
| α-farnesene          |                  | 1.3              |                  |                  |
| camphor              |                  |                  | 0.2              | 0.5              |
| Cultivation/Experimentation area | Pakistan | Egypt | China | Serbia |
| Extraction method/source | Hydrodistillation | Hydrodistillation | Hydrodistillation | Hydrodistillation |
| EO yield (%)         | 2.8              | 2.0              | 1.7              | 4.0              |
and BHT. In a study by Topal et al. (2008), AnEO showed 77.5% free radical scavenging activity using DPPH assay, which was slightly lower than that of BHT (91%). In contrast, De Martino et al. (2009) noted the least free radical scavenging activity of this EO (DPPH inhibition = 19%) and speculated the reason is the low percentage of monoterpenes (1.2%) in its chemical constituents. They discussed that antioxidant activity is directly related to the monoterpene content of EOs. Nanasombat and Wimuttigosol (2011) also reported the antioxidant activity of AnEO measured by DPPH assay with IC50 = 86.88 mg/mL.

Thus, the chemical composition and in vitro properties of EOs are very unstable and depend on the genetic factors, environmental conditions, geographical location, harvest time, plant part used, and method of extraction. The EOs may consist of 20–60 chemical compounds with two or three major components present at high concentrations (70%–80%). The potential antibacterial, antifungal, and antioxidant activities of EOs rely entirely on their major bioactive chemical compounds, functional groups, and synergistic interactions between components (Chouhan et al., 2017). Due to the variable nature of the chemical composition of EOs, it is difficult to determine the exact mechanism of action and dose rates for a specific activity (Kikusato, 2021).

2. Effects of selected essential oils on broiler performance, carcass characteristics and serum traits

Although several in vitro studies have shown the antimicrobial and antioxidant activities of EOs, the in vivo knowledge on the whole body of broiler health and growth performance is relatively less based on their chemical compositions and in vitro properties; however, possible mechanisms underlying the positive effects of EOs on biological actions can be generally hypothesized, including membrane disruption of pathogens, immunity-boosting activities, improvement of beneficial gut microbiota, appetite stimulation, and enhancement in the secretion of endogenous

| Table 4. Chemical Compositions and in vitro properties of FEO (Fennel Essential Oil) (continued) |
|-----------------------------------------------|
| **Chemical Composition** | **Antibacterial activity** | **Antifungal activity** | **Antioxidant activity** |
| Species | **Species** | **Species** | **Species** | **Species** | **Species** | **Species** |
| | **ZOI (mm)** | **MIC (µg/mL)** | **MIC (µg/mL)** | **MIC (µg/mL)** | **MIC (µg/mL)** | **MIC (µg/mL)** |
| | **15 µL/ disk** | | | | | |
| **B. subtilis** | 29 | 62.6 | | | | |
| **E. coli** | 14 | 259.3 | | | | |
| **S. typhi** | 18 | 15 | | | | |
| **E. coli** | 19 | 10 | | | | |
| **S. typhi** | 20.2 | 0.25 | | | | |
| **Staph. aureus** | 11.5 | 0.25 | | | | |
| **k. pneumoniae** | 21 | 75 | | | | |
| **P. aeruginosa** | 12.3 | 10 | | | | |
| **A. niger** | 28 | 80.6 | 22 | 10 | 20 | 10 |
| **Can. albicans** | 100% | 25 | | | | |
| **A. flavus** | NA | NA | NA | NA | NA | NA |

ZOI = zone of inhibition, MIC = minimum inhibitory concentration

1 Concentration of compound that affords a 50% reduction in the assay.
Table 5. **Chemical Compositions and in vitro properties of AnEO (Anise Essential Oil)**

| Chemical Composition | Sharifi et al., 2008 | Topal et al., 2008 | De Martino et al., 2009 | Foroughi et al., 2016 |
|----------------------|----------------------|-------------------|------------------------|-----------------------|
| trans-anethole       | 92.9                 | 79.0              | 89.7                   | 0.9                   |
| cis-anethole         | 0.1                  | 97.1              | 0.4                    | 0.1                   |
| estragole            | 2.2                  | 3.6               | 0.3                    | 0.4                   |
| α-pinene             |                      |                   |                        |                       |
| anisaldehyde         | 0.1                  | 0.7               | 0.1                    | 0.4                   |
| α-himachalene        |                      |                   |                        |                       |
| carvone              | 0.8                  | 0.4               |                        |                       |
| α-bisabolene         | 1.8                  | 0.4               |                        |                       |
| zingiberene          | 0.4                  | 0.4               |                        |                       |
| methyl-chavicol      | 2.2                  | 0.4               |                        |                       |
| 3,4-dimethoxystyrene |                      | 5.2               |                        |                       |
| α-gurjunene          |                      |                   | 4.0                    |                       |
| limonene             | 0.8                  | 0.8               |                        |                       |
| fenchone             | 0.2                  | 4.6               |                        |                       |
| linalool             | 0.3                  | 0.4               |                        |                       |
| α-allyanisole        | 2.2                  | 0.1               |                        |                       |
| cis-dihydrocarvone   | 0.1                  | 0.1               |                        |                       |
| δ-element            | 0.1                  | 0.1               |                        |                       |
| aromadendrene        | 0.1                  | 0.1               |                        |                       |
| α-curcumene          | 0.2                  | 0.2               |                        |                       |
| β-bisabolene         | 0.2                  | 0.2               |                        |                       |
| β-sesquiphellandrene | 0.1                  | 0.2               |                        |                       |
| α-terpinene          | 0.2                  | 0.2               |                        |                       |
| ylangene             | 0.2                  | 0.2               |                        |                       |
| elemene              | 0.2                  | 0.2               |                        |                       |
| α-caryophyllene      | 0.2                  | 0.2               |                        |                       |
| α-cis-himachalene    | 0.5                  | 0.5               |                        |                       |
| α-ethyl-β-anisyl alcohol | 0.3                | 0.3               |                        |                       |
| 1-methylguanaine     | 0.1                  | 0.1               |                        |                       |
| spathulenol          | 0.2                  | 0.2               |                        |                       |
| 3-hydroxyacarbofuran | 0.8                  | 0.8               |                        |                       |
| ethyl oleate         | 0.9                  | 0.9               |                        |                       |
| methyl 1-phenylallyl ether | 1.7              | 1.7               |                        |                       |
| α-phellandrene       |                      | 0.1               | 0.01                   |                       |
| Δ3-carene            |                      | 0.1               |                        |                       |
| α-cymene             |                      | 0.1               |                        |                       |
| ρ-cymene             |                      | 0.1               |                        |                       |
| 1,8-cineole          |                      | 0.1               |                        |                       |
| camphor              |                      | 0.2               |                        |                       |
| β-monopalmitate      |                      | 0.2               |                        |                       |
| Δ2-furylethane       |                      | 0.2               |                        |                       |
| Cultivation/experimentation area | Iran | Turkey | Italy | Iran |
| Extraction method/source | Hydrodistillation | Hydrodistillation | Commercial | Hydrodistillation |
| EO yield (%)         | 3.3                  | Not given         | —                      | Not given             |
digestive enzymes (Williams, 2001; Cross et al., 2007; Hong et al., 2012; Sugiharto, 2016). Thus, some of this information is valuable to the application of EOs in the development of feed additives. It should also be noted that excess supplementation could decrease growth performance, possibly due to the potent nature of EOs, which negatively affects the digestive system by reducing FI and disturbing gut microflora at higher dose rates (Falaki et al., 2016).

2.1. Broiler performance (Table 6)

For CEO supplementation, Ghazanfari et al. (2015) reported a significant decrease in feed intake (FI), increase in body weight gain (BWG), and better feed conversion ratio (FCR) at 0.01, 0.02, and 0.03% of CEO in broiler diets compared to the negative control (NC: no supplementation of any EO or AGP). The highest output was noted with 0.03% supplementation, where a 9% increase in BWG and an 8% decrease in FCR were observed at the end of the experiment. Falaki et al. (2016) showed that the BWG of broilers increased by supplementing the AjEO up to 0.025% in the diet and started to decrease at 0.03% supplementation, although the FI was unchanged. One possible reason why growth performance was reduced by the overdose may be involved in thujone in AjEO, considering that this chemical component in sage oil is responsible for renal and liver dysfunction (Traesel et al., 2011). In contrast, Chowdhury et al. (2018a) reported neither positive nor negative effects of 0.04% AjEO supplementation on growth performance compared to NC, although this supplementation level (0.04%) was even higher than the level at which Falaki et al. (2016) have negative effects on performance. The AjEO used in their study was not extracted by themselves; however, it was procured from a commercial company and was not chemically analyzed to check the purity and chemical composition. This suggests that the purity and chemical composition of EOs should be clarified to determine their effects on growth performance and other parameters in broilers.

Supplementation with FEO improved the BWG by up to 9% and reduced the FCR by up to 6% with 0.025% supplementation in broiler diets (Gharehsheikhlou et al., 2018). In contrast, Stef et al. (2018) reported the non-significant positive effects of FEO on the growth performance supplemented with 0.0125% and 0.025% in broiler diets. In both studies, the authors did not mention the purity and chemical composition of the FEO examined. The discrepancies in the results might be due to differences in the chemical composition and purity of the EOs. More detailed studies are required to clarify the reasons for these differences.

Several studies on AnEO supplementation have been conducted. In the 2000s, 0.04% AnEO-supplemented feed exhibited significantly improved body weight gain (Ciftci et al., 2005; Simsek et al., 2007). These observations were confirmed by Bhandari and Yadav (2013) and Eltazi (2014) using 0.04% AnEO. However, the effects of AnEO on feed intake are controversial: Bhandari and Yadav (2013), Eltazi (2014), and Stef et al. (2018) showed no changes, increases, or decreases in feed intake containing AnEO at 0.02–0.025% of diet, respectively.

Thus, many researchers have reported the positive effects of selected EOs obtained from the Apiaceae family on the

| Chemical Composition | Sharifi et al., 2008 | Topal et al., 2008 | De Martino et al., 2009 | Foroughi et al., 2016 |
|----------------------|----------------------|-------------------|-------------------------|-----------------------|
| Antibacterial activity | NA | NA | Species | ZOI (mm) 490 μg/disk | Species | ZOI (mm) 31 mg/mL | MIC (mg/mL) |
| B. cereus | 6 | | | | | |
| E. coli | 0 | E. coli | 22 | 3 | |
| Stop. aureus | 0 | Stop. aureus | 22 | 7 | |

| Antifungal activity | Species | MIC (μL/L) | NA | NA | NA |
|---------------------|----------|------------|-----|-----|-----|
| A. niger | 2000 (EC50 = 400 μL/L) | | | | |

| Antioxidant activity | Method | Effects | Method | Effects |
|----------------------|--------|---------|--------|---------|
| NA | DPPH | AnEO showed 77.5% free radical scavenging activity, BHT (Standard) showed 91.4% | DPPH | AnEO showed 19% free radical scavenging activity |

ZOI = zone of inhibition; MIC = minimum inhibitory concentration

1/ Half maximal effective concentration
Table 6. Effects of selected essential oils on broiler performance

| EO  | Actual data | Percent increase (+) or decrease (-) VS NC | Age | References       |
|-----|-------------|---------------------------------------------|-----|------------------|
|     | Dose rate (%)* | FI (g) | BWG (g) | FCR | FI | BWG | FCR |
| CEO | 0 (control)  | 4082  | 2122  | 1.92 | 1.05 | 8.91 | -7.29 |
|     | PC1         | 4125  | 2311  | 1.78 | -2.67 | 2.21 | -4.69 |
|     | 0.01        | 3973  | 2169  | 1.83 | -1.69 | 4.57 | -6.25 |
|     | 0.02        | 4013  | 2219  | 1.8  | 0   | 8.81 | -8.33 |
|     | 0.03        | 4082  | 2309  | 1.76 | 0   | 8.81 | -8.33 |
| AjEO| 0 (control)  | 4317  | 2277  | 1.89 | 0   | 8.91 | -7.29 |
|     | PC2         | 4243  | 2305  | 1.84 | -3.41 | 2.28 | -5.29 |
|     | 0.015       | 4170  | 2329  | 1.79 | -0.12 | 0.48 | -0.53 |
|     | 0.025       | 4132  | 2288  | 1.88 | 0.14 | -0.4 | 0.53 |
|     | 0.035       | 4323  | 2268  | 1.9  | 0   | 8.81 | -8.33 |
| AjEO| 0 (control)  | 3721  | 2304  | 1.79 | 0.75 | 2.73 | -1.86 |
|     | PC4         | 3650  | 2164  | 1.73 | -2.50 | 1.07 | -1.69 |
|     | 0.01        | 3457  | 2186  | 1.58 | 0.14 | -0.4 | 0.53 |
|     | 0.02        | 3449  | 2186  | 1.58 | -0.03 | 1.91 | -1.86 |
|     | 0.04        | 3470  | 2462  | 1.41 | 0.58 | 14.76 | -12.42 |
| FEO | 0 (control)  | 4633  | 2606  | 1.78 | 0   | 8.81 | -8.33 |
|     | 0.0125      | 4437  | 2537  | 1.75 | -4.23 | 2.65 | -1.69 |
|     | 0.025       | 4517  | 2587  | 1.75 | -2.50 | 1.07 | -1.69 |
|     | 0.04        | 4302  | 2578  | 1.75 | 0   | 8.81 | -8.33 |
| FEO | 0 (control)  | 3450  | 2146  | 1.61 | 0.20 | 7.36 | -6.83 |
|     | PC5         | 3457  | 2304  | 1.50 | -0.49 | 2.07 | -2.48 |
|     | 0.01        | 3433  | 2190  | 1.57 | -0.03 | 1.91 | -1.86 |
|     | 0.02        | 3449  | 2186  | 1.58 | 0.14 | 14.76 | -12.42 |
|     | 0.04        | 3470  | 2462  | 1.41 | 0.58 | 14.76 | -12.42 |
| AnEO| 0 (control)  | NA    | 2256  | 1.78 | 0   | 8.81 | -8.33 |
|     | PC6         | NA    | 2414  | 1.78 | 0   | 8.81 | -8.33 |
|     | 0.01        | NA    | 2296  | 1.77 | 0   | 8.81 | -8.33 |
|     | 0.02        | NA    | 2572  | 14.04 | 0   | 8.81 | -8.33 |
| AnEO| 0 (control)  | 4633  | 2606  | 1.78 | 0   | 8.81 | -8.33 |
|     | 0.0125      | 4326  | 2690  | 1.61 | -6.63 | 3.22 | -9.55 |
|     | 0.025       | 4302  | 2672  | 1.61 | -7.14 | 2.53 | -9.55 |
|     | 0.04        | 3470  | 2105  | 1.83 | 10.72 | 23.98 | -10.73 |
| AnEO| 0 (control)  | 3394  | 1796  | 1.97 | 0   | 8.81 | -8.33 |
|     | PC6         | 3437  | 1835  | 1.97 | 1.27  | 2.17 | -1.02 |
|     | 0.01        | 3408  | 1809  | 1.96 | 0.41  | 0.72 | -0.51 |
|     | 0.02        | 3412  | 1825  | 1.95 | 0.53  | 1.61 | -1.02 |
|     | 0.04        | 3471  | 1883  | 1.92 | 2.27  | 4.84 | -2.54 |
|     | 0.06        | 3450  | 1847  | 1.94 | 1.65  | 2.84 | -1.52 |

PC = positive control

All values are in percentage except PC.

1 Flavophospholipol, 600 mg/kg; 2 Virginiamycin, 200 mg/kg; 3 Bacitracin methylene disalicylate, 500 mg/kg; 4 Avilamycin, 1000 mg/kg; 5 Neomycin sulfate, 1000 mg/kg; 6 Chloretetracycline, 5 mg/kg

Mean values sharing a common superscript letter are not statistically different at \( P < 0.05 \).
growth performance of broilers. Therefore, detailed data from animal experiments should be carefully understood considering the dose rate and purity of each EO examined and their active compounds, including phenolics, terpenoids, glycosides, and alkaloids present as secondary plant metabolites.

2.2. Carcass characteristics (Table 7)

Although the CEO having linalool as a major chemical component has been proved a potent antimicrobial and growth promoter in broilers (Çabuk et al., 2003; Ghazanfari et al., 2015), there is a lack of published data regarding its effects on carcass characteristics of broilers according to the authors’ knowledge. No positive effects of AjEO and FEO supplementation have been observed on the carcass characteristics of broilers, as well (Falaki et al., 2016; Chowdhury et al., 2018a).

Simsek et al. (2007) reported improved hot and cold carcass yields by supplementing broiler diets with 0.04% AnEO. This observation was confirmed by the results of Eltazi (2014) using the same level of supplementation. Moreover, the relative percentages of breast, thigh, and drumstick and the weight of the liver and gizzard were also improved by supplementing broiler diets with 0.04% AnEO (Simsek et al., 2007; Eltazi, 2014). The highest FI was noted with 0.04% AnEO supplementation in the study by Eltazi (2014), which may be a possible reason for the improved liver and gizzard weight. The positive effects on carcass characteristics may be related to the effects of anethol, a major bioactive compound in AnEO, on the digestive system and liver metabolism of broilers.

2.3. Serum traits (Table 8)

Supplementation of CEO at 0.01% to 0.03% in broiler diets did not lead to significant changes in serum traits in broilers, including total cholesterol, triglycerides, glucose, high-density lipoproteins (HDL), low-density lipoproteins (LDL), and very low-density lipoproteins (VLDL) (Ghazanfari et al., 2015). Chowdhury et al. (2018b) reported a reduced blood total cholesterol level of up to 19% in comparison to NC by the diet supplemented with 0.04% AjEO. The concentrations of triglycerides, glucose, and total proteins, however, remained unaffected in their study. The decrease in total cholesterol levels may be due to thymol, a major component of AjEO, which can act as an inhibitor of hepatic 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase activity, which is a key regulatory enzyme in cholesterol synthesis (Lee et al., 2003).

3. Effects of selected essential oils on intestinal microbiota and gut morphology of broilers

The status of intestinal microbiota and gut morphology are important factors for evaluating gut health, including different aspects of the gastrointestinal tract (GIT), such as effective digestion of feed, absence of GIT ailment, normal and stable intestinal microbiota, and effective immune status.

### Table 7. Effects of selected essential oils on carcass characteristics

| EO    | dietary dose %* | Hot dressing | Breast | Thigh | Wing | Gizzard | Liver | Heart | Abdominal fat | Spleen | Bursa | Age | Reference       |
|-------|-----------------|--------------|--------|-------|------|---------|------|-------|---------------|--------|-------|-----|----------------|
|       | % of slaughter body weight | % of live body weight |       |       |      |         |      |       |               |        |       |     |                 |
| AjEO  | 0 (control)     | 65.5         | 22.2   | 8.73  | 5.30 | 2.27    | 1.71 | 0.48  | 1.71          | 0.11   | 0.11  | 0–39| Chowdhury et al., 2018ab |
|       | PC              | 66.0         | 21.0   | 8.99  | 5.61 | 2.24    | 1.78 | 0.48  | 2.11          | 0.13   | 0.08  |     |                 |
|       | 0.04            | 66.6         | 22.4   | 9.65  | 5.79 | 2.22    | 1.77 | 0.48  | 2.15          | 0.10^b | 0.07  |     |                 |
| AnEO  | 0 (control)     | 67.5         | 24.6c  | 15.0c |      |         |      |       |               |        |       |     | Simsek et al., 2007 |
|       | PC              | 68.8b        | 25.0b  | 15.8b |      |         |      |       |               |        |       |     |                 |
|       | 0.015           | 68.7^b       | 25.0b  | 15.8b |      |         |      |       |               |        |       |     |                 |
|       | 0.025           | 68.8^b       | 25.5b  | 15.9b |      |         |      |       |               |        |       |     |                 |
|       | 0.040           | 69.1^a       | 26.5a  | 16.8a |      |         |      |       |               |        |       |     |                 |
| AjEO  | 0 (control)     | 63.8         | 19.9   | 17.4  |      |         |      |       |               |        |       |     | Falaki et al., 2016 |
|       | PC              | 65.3         | 21.8   | 18.1  |      |         |      |       |               |        |       |     |                 |
|       | 0.015           | 66.0         | 21.2   | 18.2  |      |         |      |       |               |        |       |     |                 |
|       | 0.025           | 64.6         | 21.4   | 17.6  |      |         |      |       |               |        |       |     |                 |
|       | 0.035           | 64.0         | 20.0   | 17.8  |      |         |      |       |               |        |       |     |                 |
| AnEO  | 0 (control)     | 73.7^ab      | 28.5   | 22.2  | 10.8^h | 2.06^b | 2.4^h | 0.51  | 2.34          | 0.13   |       |     | Simsek et al., 2007 |
|       | PC              | 72.9^b       | 29.0   | 21.31 | 10.7^h| 2.12^c | 2.27^b | 0.51  | 2.45          | 0.14   |       |     |                 |
|       | 0.01           | 74.5^ab      | 28.8   | 21.36 | 11.3^a| 2.48^c | 2.43^b | 0.49  | 2.44          | 0.14   |       |     |                 |
|       | 0.02           | 73.1^ab      | 28.7   | 21.11 | 10.6^h| 2.36^bc | 2.42^b | 0.47  | 2.62          | 0.13   |       |     |                 |
|       | 0.04           | 74.6^a       | 29.5   | 21.46 | 9.8^a | 2.53^a | 2.67^a | 0.41  | 2.75          | 0.12   |       |     |                 |

PC = positive control
* All values are in percentage except PC.
1 Bacitracin methylene disalicylate, 500 mg/kg; 2 Neomycin sulfate, 1000 mg/kg; 3 Virginiamycin, 200 mg/kg; 4 Avilamycin, 1000 mg/kg
^ Mean values sharing a common superscript letter are not statistically different at P<0.05.
Intestinal microbiota play a crucial role in maintaining the health of broilers by altering several physiological functions, including digestion, metabolism, and immune responses (Carrasco et al., 2019). Broilers are vulnerable to potentially harmful bacteria such as E. coli, Salmonella species, and C. perfringens, which compete with the host in GIT for nutrients, ultimately leading to poor growth performance and greater risk of disease incidence (Gunal et al., 2006). The EO supplements can probably control intestinal microbiota, as these phytochemicals perform beneficial functions in the intestine, similar to prebiotics, even remaining less absorbed in the small intestine (Martel et al., 2020). It should be noted that the absorption of phytobiotics, including EOs, is very low in the small intestine, as only 2%-15% of the compounds can be absorbed. This fact has been supported by recent studies revealing that phytochemicals may not need to be absorbed in the body to perform beneficial functions (Kikusato, 2021). Iqbal et al. (2020) claimed that the intestinal microbiota would convert the phytochemicals into simpler metabolites to some extent to make them absorbable compounds, which may increase their bioavailability and improve the health-promoting effects in the intestine and inside the body. Furthermore, along with microbial community structure, EO supplementation could also be related to the microbial metabolites that improve the nutritional status of birds as well as GIT function and health (Ghazanfari et al., 2015). Thus, the way of phytochemicals where to work on the host or microbiota is discussed.

### 3.1. Intestinal microbiota (Table 9)

Decreased numbers of pathogenic bacteria and an increased number of beneficial bacteria in the gut may improve the ability of epithelial cells to regenerate villi and thus enhance intestinal absorptive capacity (Mourao et al., 2006). Considering the properties of phytobiotics, it is reasonable to expect such an effect by EOs due to their well-documented inhibitory effects against pathogenic microbiota. However, to the best of our knowledge, studies regarding the effects of selected EOs on the gut microbiota are limited.

CEO supplementation with 0.03% in broiler diets reduced the concentration of E. coli (log cfu/g) in caecum content by

| Table 9. Effects of selected essential oils on intestinal microbiota |
|-------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| EO | Intestine part | Dietary dose %* | Lactobacillus (log cfu/g) | E. coli (log cfu/g) | Clostridium (log cfu/g) | Age | Reference |
| CEO | Caecum content | 0 (control) | 4.46 | 4.44* | NA | 0-42 | Ghazanfari et al., 2015 |
| | | PC2 | 4.47 | 4.23b | | |
| | | 0.01 | 4.47 | 4.36b | | |
| | | 0.02 | 4.46 | 4.29b | | |
| | | 0.03 | 4.51 | 4.25b | | |
| AjEO | Pre-caecal digesta | 0 (control) | 7.77 | 7.91* | 7.27a | 0-39 | Chowdhury et al., 2018b |
| | | PC1 | 4.40 | 7.29b | 6.63b | |
| | | 0.04 | 7.74 | 7.97a | 7.26a | |

PC=positive control
* All values are in percentage except PC.
1 Flavophospholipol, 600 mg/kg; 2 Bacitracin methylene disalicylate, 500 mg/kg

| Table 8. Effects of selected essential oils on serum traits |
|------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| EO | Dietary dose %* | Cholesterol (mg/dl) | Triglyceride (mg/dl) | glucose (mg/dl) | HDL (mg/dl) | LDL (mg/dl) | VLDL (mg/dl) | Total Protein (mg/dl) | Age | Reference |
| CEO | 0 (control) | 129 | 138 | 280 | 55 | 47 | 27 | | 0-42 | Ghazanfari et al., 2015 |
| | PC2 | 114 | 82 | 240 | 52 | 46 | 16 | | |
| | 0.01 | 111 | 112 | 237 | 49 | 41 | 22 | | |
| | 0.02 | 130 | 119 | 229 | 53 | 54 | 23 | | |
| | 0.03 | 121 | 114 | 235 | 54 | 44 | 23 | | |
| AjEO | 0 (control) | 184* | 91 | 216 | | | | | 2780 | Chowdhury et al., 2018b |
| | PC1 | 194* | 90 | 220 | | | | | 2660 | |
| | 0.04 | 148b | 100 | 238 | | | | | 2810 | |

PC=positive control
* All values are in percentage except PC.
1 Flavophospholipol, 600 mg/kg; 2 Bacitracin methylene disalicylate, 500 mg/kg

Mean values sharing a common superscript letter are not statistically different at P<0.05.
4% in comparison to NC; however, the concentration of \textit{Lactobacillus} (log cfu/g) remained unchanged (Ghazanfari et al., 2015). Previously, it was observed that linalool, a major bioactive compound of CEO, inhibits the pathogenic microorganisms in the digestive system, which is possibly related to the reduction in the concentration of \textit{E. coli} in the gut (Çabuk et al., 2003; Lee et al., 2004). Chowdhury et al. (2018b) reported no significant reduction in the concentration of \textit{E. coli}, \textit{Clostridium}, and \textit{Lactobacillus} bacteria in pre-caecal digesta by supplementing broiler diets with 0.04% AjEO. They suggested that the low dose rate (0.04%) might be the reason for the unaffected concentration of \textit{E. coli} and \textit{Clostridium} bacteria; otherwise, thymol, a major bioactive compound of AjEO, is a potent antibacterial agent for these bacteria.

### 3.2. Gut morphology (Table 10)

The intestinal mucosal status and its microscopic structure may be a good indicator of the response of the GIT to active substances present in feed and the intestinal content (Viveros et al., 2011). This mucosa is one of the main barriers in the intestine that prevent the invasion of pathogens and toxins in the GIT; therefore, these barriers can be destroyed by environmental, dietary, and oxidative stress, which results in systemic and intestinal inflammation (Kikusato, 2021). According to Huang and Lee (2018), phytobiotics, including EOs, have the potential to modulate inflammation-inducing factors in the intestine and can alleviate the inflammation cascade (For detail: Kikusato, 2021) and support gut health. Regarding changes in mucosal microscopic structure with EOs, the increased VH was reported to be related to enhanced digestive and absorptive functions of the intestine due to larger absorptive surface area and higher expression of brush border enzymes and nutrient transport systems (Pluske et al., 1996).

Supplementation of CEO in broiler diets significantly affected VH, CD, and the VH/CD ratio in the duodenum, jejunum, and ileum parts of the intestine (Ghazanfari et al., 2015). VH and CD increased significantly, whereas the VH/CD ratio decreased with CEO supplementation compared to NC. Çabuk et al. (2003) demonstrated that linalool, a major component of CEO, can enhance VH in the intestine of broilers, and the activity of digestive enzymes, possibly improving digestibility and absorption of nutrients. Moreover, amylase concentration in the broiler intestine increases

### Table 10. Effects of selected essential oils on gut morphology

| EO | Intestinal site | Dietary dose %* | VH (μm) | % change VS NC | CD (μm) | % change VS NC | VH/CD Ratio | % change VS NC | Age | Reference |
|----|----------------|----------------|--------|----------------|--------|----------------|--------------|----------------|-----|-----------|
|    | Doudenum       |                |        |                |        |                |              |                |     |           |
|    | 0 (control)    |                | 1759^c | 11.91^a       | 147.8^c| 11.91^a       |              |                |     | Ghazanfari et al., 2015 |
|    | PC^2           |                | 1912^a | 6.6           | 157.6^ab| 6.6           | 12.15^a      | 2.0            |     |           |
|    | 0.01           |                | 1798^b | 2.0           | 150.8^b| 2.0           | 11.94^a      | 0.3            |     |           |
|    | 0.02           |                | 1810^b | 6.4           | 157.2^ab| 6.4           | 11.53^ab     | -3.2           |     |           |
|    | 0.03           |                | 1805^b | 9.3           | 161.6^a| 9.3           | 11.18^b      | -6.1           |     |           |
|    | CEO            | Jejunum        |        |                |        |                |              |                |     |           |
|    | 0 (control)    |                | 849^d  | 6.58^a        | 107.6^d| 6.58^a        | 7.9^a        | -16.7          | 0-42|           |
|    | PC^2           |                | 877^a  | 24.0          | 133.4^a| 24.0          | 7.85^b       | -0.6           |     |           |
|    | 0.01           |                | 858^cd | 1.7           | 109.4^cd| 1.7           | 7.59^b       | -3.9           |     |           |
|    | 0.02           |                | 866^bc | 6.1           | 114.2^bc| 6.1           | 7.53^b       | -4.7           |     |           |
|    | 0.03           |                | 872^ab | 7.8           | 116.0^b| 7.8           | 7.53^b       | -4.7           |     |           |
|    | CEO            | Ileum          |        |                |        |                |              |                |     |           |
|    | 0 (control)    |                | 757^d  | 6.4^a         | 97^d   | 6.4^a         | 7.88^a       | -18.8          |     | Chowdhury et al., 2018b |
|    | PC^2           |                | 829^a  | 33.8          | 129.8^a| 33.8          | 6.5^b        | -18.8          |     |           |
|    | 0.01           |                | 770^bc | 9.7           | 106.4^b| 9.7           | 7.26^b       | -7.9           |     |           |
|    | 0.02           |                | 799^ab | 19.8          | 116.2^b| 19.8          | 6.89^bc      | -12.6          |     |           |
|    | 0.03           |                | 783^bc | 22.0          | 118.4^b| 22.0          | 6.61^bc      | -16.1          |     |           |
|    | AjEO           | Jejunum        |        |                |        |                |              |                |     |           |
|    | 0 (control)    |                | 1307   | 19.5^b        | 70.6   | 19.5^b        | 17.0^b       | 10             | 0-39|           |
|    | PC^1           |                | 1426   | 22.9^a        | 64.8   | -8.2          | 18.7^a       | 10             |     |           |
|    | 0.04           |                | 1230   | -9.2          | 64.1   | -9.2          | 19.4^b       | -0.5           |     |           |
|    | AjEO           | Ileum          |        |                |        |                |              |                |     |           |
|    | 0 (control)    |                | 1070^b | 17.0^a        | 63.7   | 17.0^b        | 14.3^b       | 14              |     |           |
|    | PC^1           |                | 1261^a | 6.0           | 67.5   | 6.0           | 14.5^b       | 1.4            |     |           |
|    | 0.04           |                | 1036^b | 8.3           | 69.0   | 8.3           | 15.9^b       | -6.5           |     |           |
|    | AjEO           | Ileum          |        |                |        |                |              |                |     |           |
|    | 0 (control)    |                | 865^b  | 14.3^b        | 62.1^b | 14.3^b        | 14.3^b       | 23.8           |     |           |
|    | PC^1           |                | 1012^a | 10.1          | 68.4^a | 10.1          | 14.5^b       | 1.4            |     |           |
|    | 0.04           |                | 959^b  | -11.8         | 54.8^b | -11.8         | 17.7^a       | 23.8           |     |           |

PC = positive control
* All values are in percentage except PC.
1 Flavophospholipol, (600mg/kg); 2 Bacitracin methylene disalicylate, 500mg/kg
a-b Mean values sharing a common superscript letter are not statistically different at P<0.05.
after dietary supplementation with CEO, which induces the villi to grow longer. According to Chowdhury et al. (2018b), AjEO supplementation at 0.04% of the diet in broilers increased the VH and VH/CD ratio in the ileum by up to 27% and 24%, respectively. However, the morphology of the duodenum and jejunum remained unaffected.

How do EOs, such as CEO or AjEO, work on the mucosal structure? Windisch et al. (2008) suggested that EOs increase VH due to their antioxidant properties. EOs can exhibit antioxidant effects through several mechanisms. These compounds contribute to the elimination of the reactive oxygen species (ROS) produced due to oxidative stress, not only by direct antioxidant action, but also by inducing the expression of antioxidant enzymes, such as catalase and superoxide dismutase (Windisch et al., 2008; Kikusato, 2021). These antioxidant enzymes neutralize the ROS released during digestive processes, which can cause damage to the intestinal mucosa and ultimately shorten the villi. The EOs may protect the villi from oxidative damage by stimulating the activity of the antioxidant enzymes, and the phenolic group of the EOs may act as hydrogen donors showing antioxidant activity (Windisch et al., 2008). The involvement of antioxidants was confirmed by Valenzuela-Grijalva et al. (2017), who speculated that the supplemented EOs can enhance the production performance not only by better FI, possibly due to improved flavor and palatability of diet, better intestinal functions, and activation of the endocrine system, but also by anti-oxidative defense mechanisms.

Based on the discussed literature, it is clear that all the EOs are not equally effective in the antimicrobial, antioxidant, and growth-promoting effects inside the body of broilers. The benefits of EOs in terms of growth performance may depend on their biological activities. Moreover, it is difficult to determine the precise and invariant effects of each EO, as they constitute variable percentages of mixtures in EOs for each plant. In addition to effectiveness, EOs are safe to be used as growth promoters for broilers and for the user (feed manufacturers/farm managers) and the consumers of the meat products compared to AGPs. In any case, as long as antimicrobial resistance will never emerge in response to their usage, EOs can be supplemented to the broiler diet throughout the rearing period without following the withdrawal period to guarantee food safety.

Conclusions

The potential ban on the use of AGPs in the broiler industry has highlighted the development of alternatives to supplement in broiler diets to support gut health and growth performance. We have endeavored to demonstrate several key themes.

1. The published data suggest that the chemical composition and yield of EOs from selected members of the Apiaceae family are quite variable depending on the geographical origin, environmental conditions, sowing/harvesting time of the plants, and the extraction method.

2. The *in vitro* antibacterial, antifungal, and antioxidant properties vary between the same EO of different origins. The relative percentage of bioactive compounds in EOs determines the extent and type of biological activity.

3. The results of the literature regarding supplementation of selected EOs in broiler diets are arbitrary and suggest ambiguous results regarding growth performance and feed efficiency.

4. The EOs extracted from the plant parts of the Apiaceae family have the potential to be utilized as a replacement for AGPs in broiler production.

5. Although these EOs have proven beneficial effects in broilers, the literature is so limited that further investigations regarding dose rate, combination of different EOs, and possible mechanisms of action are required.

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Conflicts of Interest

The authors declare no conflict of interest.

References

Acimovic MG, Kostadinovic LM, Puvaca NM, Popovic SJ and Urosevic MI. Phytochemical constituents of selected plants from Apiaceae family and their biological effects in poultry. Food and Feed Research, 43: 35-47, 2016.

AI Maofari A, El Hajjaji S, Debbah A, Zaydoun S, Ouaki B, Charof R, Bennane Z, Hakiki A and Mosaddak M. Chemical composition and antibacterial properties of essential oils of Pimpinella Anisum L. growing in Morocco and Yemen. Scientific Study & Research, 14: 11-16, 2013.

Al Bayati FA. Synergistic antibacterial activity between Thymus vulgaris and Pimpinella anisum essential oils and methanol extracts. Journal of Ethnopharmacology, 116: 403-406, 2008.

Al Yasiry ARM and Kiczorowska B. Frankincense-therapeutic properties. Advances in Hygiene & Experimental Medicine, 70; 380-391. 2016.

Anwar F, Ali M, Hussain AI and Shahid M. Antioxidant and antimicrobial activities of essential oil and extracts of fennel (Foeniculum vulgare Mill.) seeds from Pakistan. Flavour and Fragrance Journal, 24: 170-176, 2009.

Applegate T, Klose V, Steiner T, Ganner A and Schatzmayer G. Probiotics and phytoecogenics for poultry: Myth or reality? Journal of Applied Poultry Research, 19: 194-210. 2010.

Arslan N, Gurbuz B, Sarihan EO, Bayrak A and Gumuscu A. Variation in essential oil content and composition in Turkish anise (Pimpinella anisum L.) populations. Turkish Journal of Agriculture and Forestry, 28: 173-177, 2004.

Asadollahpoor A, Abdollahi M and Rahimi R. Pimpinella anisum L.
fruit: Chemical composition and effect on rat model of nonalcoholic fatty liver disease. Journal of Research in Medical Sciences, 22: 37. 2017.

Baratta MT, Dorman HD, Deans SG, Biondi DM and Ruberto G. Chemical composition, antimicrobial and antioxidative activity of laurel, sage, rosemary, oregano and coriander essential oils. Journal of Essential Oil Research, 10: 618–627. 1998.

Bhandari BB and Yadav JL. Anise oil (Pimpinella Anisum L.) as a natural feed supplement for growth in broiler chicken. Proceedings of the 9th National Workshop on Livestock and Fisheries Research in Nepal, 30: 101–109. 2013.

Bischoff SC. ‘Gut health’: a new objective in medicine? BMC Medicine, 9: 24. 2011.

Broom LJ. Gut barrier function: effects of (antibiotic) growth promoters from broiler diets: performance indexes and economic impact. Poultry Science, 98: 6659–6667. 2019.

Ciftci M, Guler T, Dalkilic B and Ertas ON. The effect of anise oil (Pimpinella anisum) on broiler performance. International Journal of Poultry Science, 5: 149–155. 2006.

Diao WR, Hu QP, Zhang H and Xu JG. Chemical composition, antibacterial activity and mechanism of action of essential oil from seeds of fennel (Foeniculum vulgare Mill.). Food Control, 35: 109–116. 2014.

Elgayyar M, Draughon F, Golden D and Mount J. Antimicrobial activity of essential oils from plants against selected pathogenic and saprophytic microorganisms. Journal of Food Protection, 64: 1019–1024. 2001.

Eltazi S. Effect of using dietary antibiotic and anise oil as feed additives on performance and carcass quality of broiler chicks. Assiut Veterinary Medical Journal, 60: 1–9. 2014.

Falaki M, Shams Shargh M, Dastar B, Hashemi SR and Sadeghi Mahoonak AR. Growth performance, carcass characteristics and intestinal microflora of broiler chickens fed diets containing carum copticum essential oil. Poultry Science Journal, 4: 37–46. 2016.

Foroughi A, Pournaghi P, Najafi F, Zangeneh A, Zangeneh MM and Moradi R. Antibacterial effect and phytochemical screening of essential oil of Pimpinella anisum against Escherichia coli O137: H7 and Staphylococcus aureus. International Journal of Current Pharmaceutical Research, 7: 367–371. 2016.

Gharehashkelhou H, Chamani M, Seidavi A, Sadeghi A and Mohiti-Asli M. Effect of fennel and savory essential oils on performance, carcass characteristics and blood parameters of broilers. Journal of Livestock Science, 9: 23–31. 2018.

Ghazanfari S, Mohammadi Z and Adib Moradi M. Effects of coriander essential oil on the performance, blood characteristics, intestinal microbiota and histological of broilers. Brazilian Journal of Poultry Science, 17: 419–426. 2015.

Gourtard FL, Bordier M, Calba C, Erlacher-Vindel E, Gochez D, de Balogh K, Benigno C, Kalpravidh W, Roger F and Yong S. Antimicrobial policy interventions in food animal production in South East Asia. BMJ, 358: j3544. 2017.

Gradimiru A, Trifan A, Spac A, Brebu M, Miron A and Aprotosoaie A. Antibacterial activity of traditional spices against lower respiratory tract pathogens: combinatorial effects of Trachyspermum ammi essential oil with conventional antibiotics. Letters in Applied Microbiology, 67: 449–457. 2018.

Graham JP, Evans SL, Price LB and Silbergeld EK. Fate of antimicrobial-resistant enterococci and staphylococci and resistance determinants in stored poultry litter. Environmental Research, 109: 682–689. 2009.

Grashorn M. Use of phytotherapeutics in broiler nutrition – an alternative to in feedantibiotics? Journal of Animal and Feed Sciences, 19: 338–347. 2010.

Gunal M, Yayli G, Kaya O, Karahan N and Sulak O. The effects of antibiotic growth promoter, probiotic or organic acid supplement on performance, intestinal microflora and tissue of broilers. International Journal of Poultry Science, 5: 149–155. 2006.

Hong JC, Steiner T, Auffy A and Lien TF. Effects of supplemental essential oil on growth performance, lipid metabolites and immunity, intestinal characteristics, microbiota and carcass traits in broilers. Livestock Science, 144: 253–262. 2012.

Huang CM and Lee TT. Immunomodulatory effects of phytochemicals in chickens and pigs - A review. Asian-Australasian Journal Animal Science, 31: 617–627. 2018.

Ilic DP, Stanojevic LP, Troter DZ, Stanojevic JS, Danilovic BR, Nikolic VD and Nikolic LB. Improvement of the yield and antimicrobial activity of fennel (Foeniculum vulgare Mill.) essential oil by fruit milling. Industrial Crops and Products, 142: 111854. 2019.

Iqbal Y, Cottrell JJ, Suleria HAR and Dunshea FR. Gut microbiota-polyphenol interactions in chicken: A review. Animals, 10: 1391–1409. 2020.
Jeya K, Veerapagu M and Sangeetha V. Antimicrobial and anti-
Kazemi M. Chemical composition and antimicrobial, antioxidant
Kacaniova M, Galovicova L, Ivanisova E, Vukovic NL, Stefanikova
Roby MHH, Sarhan MA, Selim KAH and Khalel KI. Antioxidant
Ozcan MM and Chalchat JC. Chemical composition and antifungal
Lee KW, Everts H and Beynen AC. Essential oils in broiler nu-
Kiczorowska B, Klebaniuk R, Bakowski M and Al Yasiry ARMH.
06
application in foods. Foods, 9: 282–300. 2020.
Kazemi M. Chemical composition and antimicrobial, antioxidative
activities and anti-inflammatory potential of A. millefo-
lL., Anethum graveolens L., and Carum cypitcim L.
Kacaniova M, Galovicova L, Ivanisova E, Vukovic NL, Stefanikova
Smart R, Kiani H, Farzaneh M and Ahmadzadeh M. Chemical
composition of essential oils of Iranian P. anisum L. and Foeniculum
Coriandrum sativum L. seed essential oil. American Journal of Essential Oils and Natural Products, 7: 60–10. 2019.
Kacaniova M, Galovicova L, Ivanisova E, Vukovic NL, Stefanikova
J., Valkova V, Borotova P, Ziarovska J, Terentjeva M and Felsociova S. Antioxidant, antimicrobial and antibiofilm ac-
tivity of coriander (Coriandrum sativum L.) essential oil for its
application in foods. Foods, 9: 282–300. 2020.
Kazemi M. Chemical composition and antimicrobial, antioxidative
activities and anti-inflammatory potential of A. millefoil-
ium L., Anthemum graveolens L., and Carum cypitcim L.
Kiczorowska B, Klebaniuk R, Bakowski M and Al Yasiry ARMH.
Culinary herbs—the nutritive value and content of minerals.
Journal of Elementology, 20: 599–608. 2015.
Kikusato M. Phytobiotics to improve health and production of
broiler chickens: functions beyond the antioxidative activity.
Animal BioScience, 34: 345–355. 2021.
Kooti W, Moradi M, Ali-Akbari S, Sharafi-Ahvazi N, Asadi-
Samani M and Ashtary-Larky D. Therapeutic and pharmaco-
ological potential of Foeniculum vulgare Mill. a review. Journal of HerbMed Pharmacology, 4: 1–9. 2015.
Lee KW, Everts H and Beynen AC. Essential oils in broiler nu-
trition. International Journal of Poultry Science, 3: 738–752.
Lee KW, Everts H, Kappert H, Frehner M, Losa R and Beynen A.
Effects of dietary essential oil components on growth per-
formance, digestive enzymes and lipid metabolism in female
broiler chickens. British Poultry Science, 44: 450–457. 2003.
Martel J, Ojcius DM, Ko YF and Young JD. Phytochemicals as pre-
biotics and biological stress inducers. Trends Biochemical
Sciences, 45: 462–471. 2020.
Mourao JL, Pinheiro V, Alves A, Guedes C, Pinto L, Saavedra MJ,
Spring P and Kocher A. Effect of mannann oligosaccharides on
the performance, intestinal morphology and cecal fermentation
of fattening rabbits. Animal Feed Science and Technology,
126: 107–120. 2006.
Nanasombat S and Wimuttigosol P. Antimicrobial and antioxidant
activity of spice essential oils. Food Science and Bio-
technology, 20: 45–53. 2011.
Ozcan MM and Chalchat JC. Chemical composition and antifungal
effect of anise (Pimpinella anisum L.) fruit oil at ripening stage.
Annals of Microbiology, 56: 353–358. 2006.
Patil SD, Maknikar PP, Wankhade SJ, Ukesh CS and Rai MK.
Chemical composition, antimicrobial and antioxidant activity
of essential oils from cumin and ajowan. Nusantrasa Bioscience,
8: 60–65. 2016.
Paul S, Dubey R, Maheswari D and Kang SC. Trachyspermum
ammi (L.) fruit essential oil influencing on membrane perme-
ability and surface characteristics in inhibiting food-borne
pathogens. Food Control, 22: 725–731. 2011.
Pimenov MG and Leonov MV. The genera of the Umbelliferae: a
nomenclator. Royal Botanic Gardens, Kew and Botanical
Garden of Moscow University, Russia, 1993.
Pluske JR, Thompson MJ, Atwood CS, Bird PH, Williams IH and
Hartmann PE. Maintenance of villus height and crypt depth,
and enhancement of disaccharide digestion and monosacchar-
ide absorption, in piglets fed on cows’ whole milk after
weaning. British Journal of Nutrition, 76: 409–422. 1996.
Roby MHH, Sarhan MA, Selim KAH and Khalel KI. Antioxidant
and antimicrobial activities of essential oil and extracts of
fennel (Foeniculum vulgare L.) and chamomile (Matricaria
chamomilla L.). Industrial Crops and Products, 44: 437–445.
2013.
Salim HM, Huque KS, Kamaruddin KM and Haque Beg A. Global
restriction of using antibiotic growth promoters and alternative
strategies in poultry production. Science Progress, 101: 52–75.
2018.
Shahwar MK, El Ghorab AH, Anjum FM, Butt MS, Hussain S
and Nadeem M. Characterization of coriander (Coriandrum
sativum L.) seeds and leaves: volatile and non-volatile extracts.
International Journal of Food Properties, 15: 736–747. 2012.
Sharifi R, Kiani H, Farzaneh M and Ahmadzadeh M. Chemical
composition of essential oils of Iranian P. anisum L. and
Foeniculum vulgare Miller and their antifungal activity against
postharvest pathogens. Journal of Essential Oil Bearing
Plants, 11: 514–522. 2008.
Simek U, Ciftci M, Dalkilic B, Guler T and Ertas O. The effects
of dietary antibiotic and anise oil supplementation on body
weight, carcass characteristics and organoleptic analysis of
meat in broilers. Veterinary Medicine Review, 158: 514–518.
2007.
Singh G, Kapoor I, Singh P, De Heluani C and Catalan C. Chemical
composition and antioxidant potential of essential oil and
oleoresins from anise seeds (Pimpinella anisum L.). Inter-
national Journal of Essential Oil Therapeutics, 2: 122–130.
2008.
Singh G, Maurya S, Catalan C and De Lampasona M. Chemical
constituents, antifungal and antioxidative effects of ajwain
essential oil and its acetone extract. Journal of Agricultural and
Food Chemistry, 52: 3292–3296. 2004.
Singh G, Maurya S, De Lampasona M and Catalan C. Chemical
constituents, antimicrobial investigations, and antioxidative
potentials of Anethum graveolens L. essential oil and acetone
extract; Part 52. Journal of Food Science, 70: 208–215.
2005.
Singh G, Maurya S, De Lampasona M and Catalan CA. Studies on
essential oils, Part 41. Chemical composition, antifungal,
antioxidant and sprout suppressant activities of coriander
(Coriandrum sativum) essential oil and its oleoresin. Flavour
and Fragrance Journal, 21: 472–479. 2006.
Singh K, Rani R, Bansal P, Medhe S and Srivastava M. Antioxidant
activity of essential oil of Coriandrum sativum and standardiza-
tion of HPTLC method for the estimation of major phyto-
markers. Journal of Analytical Chemistry, 70: 220–224. 2015.
Singh S, Das S, Singh G, Perotti M, Schuff C and Catalán C.
Comparative studies of chemical composition, antioxidant
and antimicrobial potentials of essential oils and oleoresins
obtained from seeds and leaves of Anethum graveolens L.
Toxicology: Open Access, 3: 2–9. 2017.
Stef L, Simiz E, Marcu A, Stef D, Gherasim V, Pet I, Patruica S,
Ahmadi M, Manciu A and Julean C. The Effect of Essential
Oils on the Bioproductive Performance of Broilers. Animal
Sciences and Biotechnologies, 51: 43–53. 2012.
Sugiharto S. Role of nutraceuticals in gut health and growth per-
formance of poultry. Journal of the Saudi Society of Agri-
cultural Sciences & Biotechnologies, 51: 43–53. 2012.
Topal U, Sasaki M, Goto M and Otles S. Chemical compositions
and antioxidant properties of essential oils from nine species of
Turkish plants obtained by supercritical carbon dioxide
extraction and steam distillation. International Journal of Food
Sciences and Nutrition, 59: 619–634. 2008.
Traesel CK, Wolkmer P, Schmidt C, Silva CB, Paim FC, Rosa AP,
Alves SH, Santurio JM and Lopes ST. Serum biochemical
profile and performance of broiler chickens fed diets containing essential oils and pepper. Comparative Clinical Pathology, 20: 453–460. 2011.

Valenzuela-Grijalva NV, Pinelli-Saavedra A, Muhlia-Almazan A, Dominguez-Diaz D and Gonzalez-Rios H. Dietary inclusion effects of phytochemicals as growth promoters in animal production. Journal of Animal Sciences and Technology, 59: 8. 2017.

Vieira JN, Goncalves C, Villarreal J, Goncalves V, Lund R, Freitag R, Silva A and Nascente P. Chemical composition of essential oils from the Apiaceae family, cytotoxicity, and their antifungal activity in vitro against Candida species from oral cavity. Brazilian Journal of Biology, 79: 432–437. 2019.

Vitali LA, Beghelli D, Nya PCB, Bistoni O, Cappellacci L, Damiano S, Lupidi G, Maggi F, Orsomando G and Papa F. Diverse biological effects of the essential oil from Iranian Trachyspermum ammi. Arabian Journal of Chemistry, 9: 775–786. 2016.

Viveros A, Chamorro S, Pizarro M, Arija I, Centeno C and Brenes A. Effects of dietary polyphenol-rich grape products on intestinal microflora and gut morphology in broiler chicks. Poultry Science, 90: 566–578. 2011.

Windisch W, Schedle K, Plitzner C, and Kroismayr A. Use of phytogenic products as feed additives for swine and poultry. Journal of Animal Science, 86: E140–E148. 2008.

Williams P. The use of essential oils and their compounds in poultry nutrition. World Poultry, 17: 14–15. 2001.

Yili A, Aisa H, Maksimov V, Veshkurova O and Salikhov SI. Chemical composition and antimicrobial activity of essential oil from seeds of Anethum graveolens growing in Uzbekistan. Chemistry of Natural Compounds, 45: 280–281. 2009.

Zeng Z, Zhang S, Wang H and Piao X. Essential oil and aromatic plants as feed additives in non-ruminant: a review. Journal of Animal Science and Biotechnology, 6: 7. 2015.