

Effects of Essential Oils on Enhancing Animal Performance

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ABSTRACT

This article aimed to provide an overview of the promising potentials of essential oils derived from plants as a natural strategy based on their antioxidative properties to enhance animal productive performance and reproductive function, prevent parasite infection, reduce transport stress, hypoxia stress, and heat stress, as well as the underlying mechanism. It highlighted the potential of essential oils to improve productivity while promoting animal welfare and environmental sustainability.

Keywords: Essential Oils; Productive Performance; Reproductive Function; Stress

Abbreviations: ADG: Average Daily Gain; CRP: C-Reactive Protein; BW: Body Weight; ETEC: Enterotoxigenic *Escherichia Coli*; T-SOD: Total Superoxide Dismutase; TLR: Toll-Like Receptor; PEO: Peppermint Essential Oil; TTO: Tea Tree Oil; AWMA: Adult Worm's Motility Assay; CLE: Curcuma Longa Essential Oil; HPI: Hypothalamic-Pituitary-Interrenal; LP: Lipid Peroxidation; TAN: Total Ammonia Nitrogen; CEO: Clove Essential Oil

Introduction

Essential oils, volatile compounds extracted from plants, have gained significant attention in animal nutrition due to their potential to enhance animal performance and overall health [1]. These natural substances are rich in bioactive compounds such as terpenes, phenolics, and aldehydes, which exhibit versatile biological properties, such as antioxidant, antimicrobial, and anti-inflammatory effects [2]. Research has demonstrated that essential oils can improve feed efficiency, growth rates, and immune function in various animal species [3-5]. They achieve these benefits through several mechanisms, including modulating gut microbiota, enhancing nutrient absorption, and reducing oxidative stress. This article provides an overview of the growing interest in essential oils as a natural strategy based on their antioxidative properties to enhance animal productive performance and reproductive function, prevent parasite infection, reduce

transport stress, hypoxia stress, and heat stress, highlighting their potential to improve productivity while promoting animal welfare and environmental sustainability.

Benefits on Productive Performance

Oxidative stress is a harmful process associated with decreased nutrient absorption and growth performance [6]. Maintaining gastrointestinal health is crucial for animal growth and production. Research has shown that dietary supplementation with essential oil including carvacrol positively impacts the gut flora, leading to improved growth performance in broilers [7]. Additionally, preincubation of intestinal epithelial cells (Caco-2 cells) with citral, cinnamaldehyde, or a combination of both compounds could attenuate LPS-induced LP and restore GSH and SOD activities [6,8]. These findings suggest that essential oils can act as growth promoters by existing beneficial effect

against oxidative stress-induced gut injury in animals. Sows experience elevated systemic oxidative stress during early lactation and late gestation [9]. Weaning is a critical period in pig production that impacts the productivity commercial farms. *Origanum vulgare* L. (oregano) EO could significantly reduce TBARS and 8-OHdG levels in sow on day 1 of lactation [9]. By the third week of lactation, administration of this EO could enhance sow's feed intake, resulting in a higher average daily gain (ADG) in piglets, suggesting that OEO supplementation in sows' diet could improve piglet performance, likely due to reduce oxidative stress [9]. Supplementation with OEO and garlic (*Allium sativum*) powder at 0.4% yielded better ADG, higher C-reactive protein (CRP) levels, and better overall productive performance compared to the control group [10].

Additionally, dietary supplementation with a mixed EO blend, primarily consisting of cinnamaldehyde (aliphatic aldehyde, from cinnamon) and thymol (phenolic compound from oregano and thyme), increased ADG and body weight (BW), while decreasing the feed conversion ratio (F:G) in weaning piglets [11]. This improvement is associated with enhanced global antioxidative capacity and reduced systemic inflammation [11]. Further, this mixed EO blend could improve growth performance and protect gut health against Enterotoxigenic *Escherichia coli* (ETEC) infection in weanling pigs through modulating intestinal integrity, as evidenced by a reduction in gut permeability, an improvement in villus morphology, an enhancement in tight junction proteins expression, a modulation in oxidative stress such as increasing total superoxide dismutase (T-SOD) activities, reducing MDA levels in the serum and jejunal mucosa [11]. This EO blend also alleviated ETEC-induced inflammatory response through diminishing pro-inflammatory cytokine contents (IL-6, IL-8, TNF- α) and inhibiting the activation of toll-like receptor (TLR)4/NF- κ B signaling pathway [11]. Furthermore, it modulated immune function by elevating serum immunoglobulin (Ig) M, IgG and jejunal sIgA concentrations, and im-

proved gut microbiota by increasing the abundance of beneficial bacteria (e.g., Lactobacillus) and reducing pathogenic bacteria (e.g., Proteobacteria) [11]. The effects of EOs on growth performance provide new insights into improving the health of weanling pigs.

In laying hens, treatment with 200 mg/kg of OEO significantly improved performance, egg weight, feed intake, and intestinal morphology while reducing oxidative stress [12]. Similarly, dietary supplementation with *Prosopis Africana* essential oils (PAOs) tended to stimulate appetite, improve digestive enzyme generation and weight gain, and reduce free radicals in broiler chickens [13]. With the rapid development of intensive aquaculture, several factors contribute to enteritis, including increased aquaculture density, pathogenic microorganism invasion, nutrient deficiencies in feed, and the aquaculture environmental deterioration, all of which impact the growth of aquaculture [14,15]. *Artemisia vulgaris* essential oil could effectively alleviate enteritis in zebrafish. This action might be involved in improving intestinal function and architecture including diminishing oxidative damage and histopathological lesions, repairing immune system, and regulating IL-1 β , IL-10 and other gene expressions through the MyD88/TRAF6/NF- κ B pathway [16]. Additionally, this EO showed greater protective action against enteritis in male zebrafish compared to females [16]. Digestive issues and stress negatively impact productivity in Nile tilapia (*Oreochromis niloticus*) [17]. Peppermint essential oil (PEO) in the diet could improve feed intake, weight gain, and feed conversion by increasing SOD and CAT activities, while reducing MDA and NO production in the liver of juvenile Nile tilapia [17].

The findings clarify the benefits of EOs on health and growth in aquaculture, providing an advantageous theoretical foundation for delving into the feasibility of using EO as a replacement for feed antibiotics. The effects of OEs on productive performance were listed in Table 1.

Table 1: Essential oils plant name and their antioxidative effects on productive performance.

Plants	Experiment model	The dose/concentration of administration	Main action	References
Productive Performance				
<i>Origanum vulgare</i> L. (Oregano)	Sows	15 mg/kg dietary supplementation	Increasing sow's feed intake and improving performance of their piglets	[9]
<i>Origanum vulgare</i> L. (Oregano)	Lohmann white laying hens	200, 400, 600 mg/kg in the diets	Improving performance, egg weight, feed intake and intestinal morphology	[12]
<i>Prosopis Africana</i>	Broiler chickens	800 mg/kg dietary supplementation	Arousing their appetite, translating to higher weight gain	[13]
<i>Mentha piperita</i> (peppermint)	Juvenile Nile tilapia	0.6 g/kg dietary supplementation	Improving the feed intake, weight gain, and feed conversion	[17]
Reproductive Function				
<i>Satureja Khuzestanica</i>	Busulfan-treated genotoxicity mice	225 mg/kg for 28 days	Reducing oxidative stress, apoptosis, cytotoxicity, and genotoxicity in sperm and testes	[18]
<i>khozestanica</i>	Nonophenol treated female mice	225 mg/kg for 28 days	Increasing the quality and number of oocytes	[19]

Note: AWMA, adult worm's motility assays; b. w., body weight.

Table 2: Essential oils plant name and their antioxidative effects on parasite infection and stress.

Plants	Experiment model	The dose/concentration of administration	Main action	References
Parasite Infection				
<i>Melaleuca alternifolia</i>	Pacu infected with <i>Ichthyophthirius multifiliis</i>	bath treatment of 1 h/day for 4 days	Eliminating 100% of parasites present in the gills and skin, minimizing the hepatic oxidative stress	[21]
<i>Mentha pulegium</i>	The AWMA assay	400 mg/kg, exposure 6 h	Nematicidal effect	[24]
<i>Curcuma longa</i>	<i>Toxoplasma gondii</i> tachyzoites infected mice	1.25, 2.5, and 5 mg/kg/day for 2 weeks	Inhibitory effects on <i>Toxoplasma gondii</i> tachyzoites, increasing the survival rate	[26]
Transport Stress				
<i>Lippia alba</i>	Cururu stingray were transported for 24 h or 48 h	200mg/L	protecting brain against transport stress and oxidative stress	[30]
<i>Ocimum gratissimum</i> L.	<i>L. alexandri</i> were subjected to 4 h transport	5 and 10mg/L	Reducing animal metabolism and opercular beats, and resulting in no mortality	[31]
<i>Ocimum basilicum</i>	Fish were transported for 72 h	5 and 10mg/L	Reducing oxidative stress, alleviating the morphological damage of gill tissue	[35]
<i>Melissa officinalis</i> L.,	Sea bass subjected to 72 h transport	10, 20, 40 mg/L	Increasing the total antioxidant capacity and protecting liver	[36]
Hypoxia Stress				
<i>Melaleuca alternifolia</i>	Fish subjected to 96-h hypoxia stress trial	89.63 and 90.25mg/kg diets for 60 days	Reducing ERS, oxidative damage, and autophagy	[41]
Heat Stress				
<i>Syzygium aromaticum</i> L. (clove)	broiler subjected to cyclic heat stress	250, 350, and 450 ppm dietary supplementation	Improving feed intake, body weight gain, and carcass component weight	[44]
<i>Lippia origanoides</i> <i>Rosmarinus officinalis</i>	broiler chickens undergo heat stress	45 ppm + 45 ppm dietary supplementation	Improving growth performance, body weight, intestinal permeability	[45]

Note: ERS, endoplasmic reticulum stress; ppm, parts per million.

Benefits on Reproductive Function

Animal experiments have shown the positive effects of EOs on protecting reproductive function in both male and female models (Table 1). Animal experiments have shown the positive effects of EOs on protecting reproductive function in both male and female models. In normal male rats, *Satureja Khuzestanica* essential oil (SKEO) SKEO has demonstrated an obvious elevation in serum testosterone level, spermatozoa, and Leydig cells [18]. Furthermore, pre-treatment with this EO played an important role in diminishing genotoxicity, cytotoxicity, apoptosis and oxidative stress in sperm and testes, indicated by increased activities of SOD and GPx, along with significantly decreased TBA and LDH levels in the G4 compared with the G3 and G2, via the regulation of Bcl-2 family gene expressions in busulfan-treated genotoxicity mice [18]. In addition, prescribing *Satureja khuzestanica* essential oil protected oocytes maturity against nanophenol. It presented its androgenic and antioxidative benefits, resulting in an obvious elevated number and quality of oocytes, thus preventing reproductive toxicity and significantly increasing the fertilization rate in nonophenol-treated female mice models [19]. It could be concluded that the beneficial properties of EOs, whether used as pretreatment or co-treatment, may prevent reproductive damage.

Preventing Parasite Infection

Ichthyophthirius multifiliis is a widespread holotrichous obligate parasite that causes ichthyophthiriasis or “white spot disease”, a major parasitic disease affecting freshwater fish species [20]. During *Ichthyophthirius multifiliis* infection, oxidative stress is one of the underlying mechanisms involved in the onset and development of liver damage [21]. EO from *Melaleuca alternifolia*, usually well-known as tea tree oil (TTO) has proven effective in eliminating all *I. multifiliis* existing in the skin and gills of Pacu following a TTO bath treatment for one hour per day over four days [21]. Furthermore, TTO offered a therapeutic alternative for silver catfish during *I. multifiliis* infection, helping to avoid or minimize hepatic oxidative stress. It was indicated by the reduction of trophonts on the skin, decreased protein carbonylation content and hepatic TBARS, along with an enhanced antioxidant system and preservation of hepatocytes morphology in infected fish [21]. Furthermore, the research results explored that TTO could be a new method for preventing or treating liver injury in fish infected by bacteria, including *Aeromonas hydrophila* [22] and *Pseudomonas aeruginosa* [23]. *Mentha pulegium* essential oil has shown its antioxidant potential by total antioxidant capacity, DPPH quenching effect, ABTS activity, and FRAP effect *in vitro* [24]. In the adult worm's motil-

ity assay (AWMA), worms exposed to *M. pulegium* EO showed paralysis after six hours [24].

Regarding *in vivo* studies, this essential oil presented a nematicidal through elevating endogenous antioxidant activities (GPx, CAT, SOD), thus reducing morphological damages during *Haemonchus contortus polygyrus* infection in a rodent model [24]. Toxoplasmosis affects human and warm-blooded animal infected with the *Toxoplasma gondii* parasite [25]. *Curcuma longa* essential oil (CLE) demonstrated an inhibitory effect on *Toxoplasma gondii* tachyzoites, evidenced by an elevated survival, apoptosis, NO level, activity of antioxidant enzymes and proinflammatory cytokines, and a reduced mean number of peritoneal tachyzoites [26]. CLE also decreased hepatic oxidative stress markers without significant toxicity to vital organs in infected mice [26]. The findings reveal the significant *in vivo* and *in vitro* inhibitory effects of EOs on parasite infections, improving survival rate and minimizing hepatic oxidative stress. The protective effects of OEs on parasite infection were listed in Table 2.

Effects on Stress Syndrome

Transport Stress

Transport is considered a stressful situation. Transporting live fish plays an important role in the aquaculture supply chain and commonly involves placing fish in bags or containers, which can cause metabolic disorder, mechanical damage, and elevated mortality, resulting in a significant economic loss for the aquaculture industry [27]. Transport could stimulate the hypothalamic-pituitary-interrenal (HPI) axis, activating catecholamines releasing into the bloodstream, and causing changes in blood components [28,29]. Moreover, transport stress leads to elevated lactate levels in the brain and plasma upon arrival and triggers GSH oxidation and lipid peroxidation (LP), as indicated by increased levels of H_2O_2 and superoxide anion ($O_2^{\cdot-}$), elevated HSP70 levels, and decreased activities of GR, GPx 1, CAT, SOD 1 and SOD 2 in the brain though the activation of NF- κ B [30]. Juveniles transported with *Ocimum gratissimum* L. essential oil (EOOG) appeared to reduce metabolism, opercular beats, hemoglobin levels, blood AST, Glu, lipid and protein oxidation concentrations in tissues [31]. This ensured lower ammonia and higher dissolved oxygen levels, prevented LP and the formation of carbonyl protein, and resulted in no mortality 24 hours after transport [31]. The essential oil extracted from basil (*Ocimum gratissimum* L.) (EOOG) presented various chemotypes (geraniol, thymol, eugenol) and had promise as an anesthetic in fish [32,33]. During transport, an increased GST in the brain and liver of fish treated with EOOG appeared its efficiency in mitigating stress [34].

It has been recommended to add essential oils into transport water as a good method to reduce stress. Treatment with EO from *Lippia alba*, principally the linalool chemotype, protected the brain against transportation and oxidative stress upon arrival, as evidenced by reduced levels of HSP70, lactate, H_2O_2 and $O_2^{\cdot-}$, activation of NF- κ B,

elevated GR, GPx1, CAT, and SOD, reduced oxidized GSH ratio, and regulated physiological responses to stress, thus avoiding the elevation of TBARS in the brain [30]. It was found that adding *Ocimum basilicum* EO to transport water resulted in a decrease in water pH, dissolved oxygen (DO), and total ammonia nitrogen (TAN) levels, as well as reductions in serum GLU and cortisol (COR), and liver GPX, MDA, CAT, and SOD levels [35]. It also increased serum LDH, AST, and ALT, muscle total free amino acid (TFAA) levels, and alleviated morphological damage to gill tissue [35]. *Melissa officinalis* L., treatment decreased levels of cortisol, urea nitrogen (BUN), UA, GSH-Px, MPO, CAT, HSPs, lactic acid (LD), and Glu in live fish after 72 hours transport, and increased total antioxidant capacity (T-AOC) [36]. These findings indicate appropriate addition of EOs to transport water is helpful to improve water quality, relieve stress, lower energy metabolism, reduce oxidative damage, and improve survival rate even for long-term transport, thus decreasing economic losses in the fishery industry.

Hypoxia Stress

Intensive aquaculture tends to increase the risk of hypoxia for fish, causing reduced quantity and quality of fish. Hypoxia stress will induce gill ROS overexpression, leading to autophagy, endoplasmic reticulum stress (ERS), apoptosis, and oxidative stress [37-39]. Baldissera, et al. [21] showed that TTO, obtained from *Melaleuca alternifolia*, could ameliorate silver catfish (*Rhamdia quelen*)'s antioxidative ability [40]. Hu et al. [41] elaborated on TTO's protective effects against hypoxia stress in grass carp (*Ctenopharyngodon idellu*), as reflected in mending serum biomarkers (lactic acid, lactic dehydrogenase, Glu, and cortisol) and suppressing apoptosis [41]. This suppression is likely related to the hypoxia-inducible factor (HIF)/Bcl-2/adenovirus E1B 19-kDa interacting protein (BNIP3) pathway, as evidenced by decreased LC3-II protein and ATG gene expressions, reducing oxidative damage to the gills, relieved gill ERS by reducing glucose-regulated protein 78 (GRP78) protein expression and stimulating the related signaling cascade reaction, thereby improving the structural integrity of the gills [41]. Thus, EOs are capable of mitigating hypoxia stress-induced damage, and the possible mechanism may involve reducing ERS, oxidative damage, and autophagy in fish.

Heat Stress

Animals under heat stress exhibit simultaneously metabolic alterations and oxidative/nitrosative stress, causing further cellular damage and negatively influencing their performance, growth and production [42-44]. Heat stress is an inevitable challenge for the poultry industry, particularly in tropical and subtropical countries, due to the global increase in environmental temperature. Treatments with orange peel EO in Japanese quails exposed to acute heat stress indicated that supplementation could decrease MDA level and prevent tissues from lipid peroxidation [43]. Clove essential oil (CEO) contributed to the normalization of oxidative/nitrosative biomarkers, significantly improving body weight gain, feed intake, and carcass component weight in broiler subjected to cyclic heat stress [44]. In addition, in

broiler chickens under heat stress, EOs from *Rosmarinus officinalis*, *Lippia origanoides*, and natural betaine or either beetroot in diet improved intestinal permeability, body weight, growth performance, antioxidative and anti-inflammatory activities [45]. These findings indicate that supplementing natural EOs as antioxidants could mitigate heat stress. The effects of OEs on transport stress, hypoxia stress and heat stress were listed in Table 2.

Conclusion

Essential oils have shown their promising potentials on enhancing animal productive performance and reproductive function, preventing parasite infection, reducing transport stress, hypoxia stress, and heat stress, due to their remarkable antioxidative properties. Future studies should focus on understanding the specific mechanisms through which different essential oils exert their effects, as well as the optimal dosages and combinations for various animal species and production systems. Additionally, long-term research is necessary for assessing the sustainability and economic viability of incorporating essential oils into animal diets on a larger scale. Further research should also explore the potential interactions between essential oils and other dietary components to optimize their efficacy.

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