Essential Oils as Phytogenic Feed Additive: Potential Benefits on Environment, Livestock Health and Production

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Abstract

Essential oils (EOs) are volatile secondary plant metabolites, which provide characteristic aroma and flavour, specific to many plants. They are mainly derived from herbs and spices, and used traditionally by humans for many centuries for their antimicrobial and preservative properties. The term ''essential oil'' is misleading as neither they are ''essential'' for nutrition and metabolism nor ''oils'' as glycerol based lipids. They are aromatic compounds with oily appearance. The phrase "essential" derives from "essence," which means smell or taste, and relates to the property of these substances of providing specific flavours and odours to many plants. EOs and their components reduce entric methane and ammonia emission by modulating rumen microbial community structure and could be used as phytogenic feed additive. However, their effects need to be studied by long term *in vivo* experimentation, as adaptation of rumen microbes could lead to ineffectiveness. Optimum dose of bioactive essential oil components and their appropriate combinations needs to be standardized in relation to dietary composition of animals to achieve consistent benefit of their feeding for livestock health and performance before their application in large scale livestock production systems.

Keywords: Environment, essential oils, feed additive, health, phytogenic

1. Introduction

Methane, one of the important greenhouse gases, produced normally during the enteric fermentation of feeds in ruminants mainly, has global warming potential of 21 times than of carbon dioxide. Although there is intense debate on contribution of domesticated ruminants on global methane emission, it is thought to be responsible for about 15% of total methane emission due to anaerobic enteric fermentation of feeds (FAO, 2010). Currently, India possesses the world's largest livestock population (13% of the global livestock population, with 57% of the world's buffalo and 16% cattle). Enteric methane emission from Indian livestock contributed 15.1% of total global enteric methane emission (Patra, 2014). Besides, ruminant animals lose a substantial (2-15% of gross energy) amount of feed energy through methane emission, which otherwise could be converted to metabolizable energy for productive purposes. Therefore, reduction in methanogenesis is of interest of ruminant nutritionists since long for efficient utilization of feed and more recently from the perspective of global warming.

Ionophore antibiotics (monensin, lasalocid) and many other chemical substances have been used as feed additives in animals' diet in recent past for improving feed efficiency in ruminants with lesser environmental pollution. However, an increasing public awareness on hazards associated with these feed additives, i.e. appearance of residues in animal products and development of bacterial resistance to antibiotics has encouraged the research towards exploiting natural products as feed additives. Feed additives derived from plants, also called phytogenic feed additives, can be included in animals' diets to improve their productivity, properties of animal products and reducing environmental pollution through enteric methane and ammonia abatement. There are number of plant secondary metabolites (essential oils, tannins, saponins) which have been studied extensively for their potential application as feed additives. Out of these, essential oils are gaining importance in ruminant nutrition for reducing enteric methanogenesis and improvement in health and productivity of animals.

2. Essential Oils and Origin

Essential oils (EOs) are volatile secondary plant metabolites, which provide characteristic aroma and flavour, specific to many plants. They are mainly derived from herbs and spices, and used traditionally by humans for many centuries for their antimicrobial and preservative properties. The term ''essential oil'' is misleading as neither they are ''essential'' for nutrition and metabolism nor ''oils'' as glycerol based lipids. They are aromatic compounds with oily appearance. The phrase "essential" derives from "essence," which means smell or taste, and relates to the property of these substances of providing specific flavours and odours to many plants (Greathead, 2003).

3. Chemistry and Composition of Essential Oils

Essential oils can be structurally classified as alcohol, ester or aldehyde derivatives of terpenoids and phenylpropanoids. Terpenoids (monoterpenes and sesquiterpenes) are more diversified group of plant bioactive compounds abundantly available in many plants and derived from acetyl-CoA via either the deoxy-xylulose or melavonate pathways (Hart et al., 2008). These compounds are derived from a basic structure of 5 carbons (C_5H_8), commonly called an isoprene unit, and are classified depending on the number of these units in its skeleton. Phenylpropanoids are derived from aromatic amino acid, phenylalanine via shikimate pathways and less abundant family of EOs compared to terpenoids (Gershenzon et al., 2000) (Table 1).

EOs can be extracted from many parts of a plant, including the leaves, flowers, stem, seeds, roots and bark. The composition of essential oils varies among different parts of the same plant. They are very complex natural mixtures which can contain about 20–60 components at quite different concentrations and characterized by two or three major components at fairly high concentrations (20–70%) compared to others present in trace amounts. These major components determine the biological activities.

Table 1: Common essential oils with their major components

Essential oils	Plants	Major essential oil components
Cinnamon	Cinnamomum cassia	Cinnamaldehyde, Eugenol
Eucalyptus	*Eucalyptus citriodora*	Citronellal, Citronellol
Capsicum	*Capsicum annum*	Capsaicin
Oregano	*Origanum vulgare*	Carvacrol, Thymol
Clove	*Syzygium aromaticum*	Eugenol
Thyme	*Thymus vulgaris*	Thymol, Carvacrol
Lemon grass	*Cymbopogon citratus*	Citral, Limonene, Citronellal, Geraniol
Garlic	*Allium sativum*	Allicin, Diallyl sulphite
Ginger	*Zingiber officinale*	Zingiberene, Zingerone

4. Mode of Antimicrobial Activities of Essential Oils

Antimicrobial activities of EOs have been demonstrated against a wide variety of microorganisms, including Gram-positive and Gram-negative bacteria. The number of terpenoid and phenolic compounds, as well as the chemical constituents and functional groups contained in the EOs, and the interactions between them determines the antimicrobial properties. Antagonistic and synergistic effects have also been observed between components of EOs. EOs interacts with the microbial cells which involve several targets of cellular components and modulate the response of these target components. Most EOs exert their antimicrobial activities by interacting with processes associated with the bacterial cell membrane, including electron transport, ion gradients, protein translocation, phosphorylation, and other enzyme-dependent reactions. Essential oils, due to their hydrophobic nature, have a high affinity for lipids of bacterial cell membrane and accumulate in the lipidic bilayer of bacteria, occupying a space between the chains of fatty acids. The efficiency to destroy bacteria largely depends on the constituents of essential oils and their functional group. The conformational changes in membrane structure due to accumulation of fluid and expansion result in loss of membrane stability. A decrease in trans-membrane ionic gradient due to leakage of ions across the cell membrane situates pressure on the bacteria to maintain the cell integrity causing either reduced growth rate or cell death. The antimicrobial activity is higher in essential oils having phenolic structures (viz. thymol, carvacrol) due to formation of hydrogen bridges in interacting with water or trans-membrane transport of monovalent cations and protons by hydroxyl group of phenols (Ultee et al., 2002) as the way of ionophore antibiotics. Gram-positive bacteria are more susceptible, because of direct interaction of cell membrane to hydrophobic compounds of EOs.

5. Essential Oils in Reducing Environmental Pollution From Livestock

Essential oils, because of their antimicrobial properties, affect the rumen microbial ecosystem by selective inhibition of specific group of microbes depending on the dose and constituents of essential oils as well as diet of animals.

5.1. Essential oils and ruminal protein degradation

The plant bioactive compounds have the ability to improve efficiency of protein utilization by reducing protein degradation in rumen (Min et al., 2004; Dey et al., 2008)). Essential oils also have inhibitory effect on ruminal protein degradation, resulting lower ammonia production. Wallace et al., (2002) suggested that only the final stage of protein degradation, deamination of amino acids to ammonia is reduced by essential oils probably due to inhibition of hyper-ammonia (HAP) producing bacteria (*Clostridium sticklandii, Peptostreptococcus anaerobius*) in rumen. Hyper-ammonia producing bacteria are present in low numbers in the rumen (<0.01 of the rumen bacterial population), but they possess a very high deamination activity. Essential oils also alter the attachment and colonisation of plant materials entering rumen without affecting fibre digestion. Inhibition of total fermentation by supplementation of high concentration of essential oils to the rumen, suggesting shift from a selective effect on the rumen microbial population to a more general inhibition if essential oils are added in excess (Fernandez-Lopez et al., 2005; Dey et al., 2016).

The addition (1µl ml^{-1}) of eucalyptus oil (ECO) in *in vitro* fermentation media, reduced the ammonia-N concentration, however at lowest dose (0.5 µl ml^{-1}) no effect on ammonia-N was evident (Singh et al., 2017). Similarly, lemongrass essential oil (LGO) was also found to reduce ammonia-N concentration in a dose dependent manner, with reduction at a dose of 0.5 µl ml^{-1} under *in vitro* condition (Singh et al., 2016). In a recent study, Kumar (2017) observed a significant reduction in ammonia-N concentration while fermentation of oats hay supplemented with either extracts of essential oil rich eucalyptus leaves, poplar leaves or clove buds, suggesting inhibitory effect on HAP bacteria. The researcher also reported synergistic effects of blends these extracts on ruminal ammonia production. In a study with garlic oil, Dey et al. (2016) observed that the NH_3-N concentration in the *in vitro* fermentation culture was linearly decreased (p<0.05) at increasing doses (1 µl 30 ml^{-1} onwards) with reduction of feed digestibility at higher doses. These studies demonstrated the role of essential oils in reducing deamination process and potential of their application in improving efficiency of protein utilization by ruminants, thereby reducing environmental pollution by lowering ammonia emission.

5.2. Essential oils and methanogenesis

The antimicrobial effect of EOs suggests their relevance as antimethanogenic feed additive but challenge is to maintain feed digestibility. It is indicated that essential oil reduces methane production either by inhibiting methanogenic archaea, changes in the phylogenetic distribution of archeal population or activity of methane producing genes. Macheboeuf et al. (2008) reported an inhibition of methane (0.93-89%) production on incubation of corn-soybean and hay based feed with graded (1-5mM) doses of cinnamaldehyde, however, feed degradability was also reduced at higher doses. Singh et al. (2017) observed a linear decrease in methane production with increasing doses of eucalyptus oils (ECO) with a reduction in methane production (82.60% to 85.30%) with a dose of ECO at 2-3 ml l^{-1}. A reduction in methane production was reported by Kumar (2017), on incubation of oats hay with petroleum ether extracts (1 ml 30 ml^{-1}) of eucalyptus leaves, poplar leaves or clove buds. The blends of these extracts were observed to have more pronounced effect on methane reduction at lower doses.

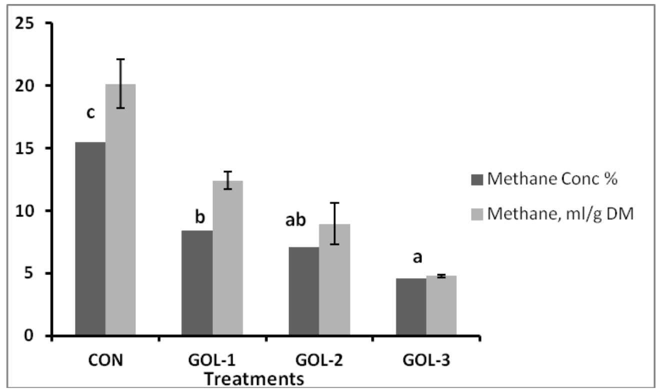

Figure 1: Dey et al., 2016

Garlic oils contain various bio-active components like allicin, diallyl sulfide, diallyl disulfide, allyl marcaptan which have direct antimicrobial effects to a group of organisms (Busquet et al., 2005). In a study with graded doses of garlic oil supplementation (Dey et al., 2016), total gas production was increased with supplementation of GOL-1 (33.3 ml l^{-1}) however, it remains unaffected (p>0.05) with higher doses (GOL-2 and GOL-3, 83.33 and 166.66 µl l^{-1}, respectively) of supplementation. Methane concentration in the head space gas was less and thus, total methane production was reduced (p<0.001) with the increasing doses of garlic oil (Fig 1). The organosulfur compounds present in the garlic oil has direct inhibitory effects on methanogenic archaea by inhibiting HMG-CoA reductase enzyme, which is required for the synthesis of membrane lipid of the archaeal community.

Wanapat et al. (2008) supplemented lemongrass powder at 100g/d to beef cattle and found improved digestibilities of nutrients, rumen microbial population, and microbial protein synthesis efficiency. A linear decrease in methane production was reported by Singh et al. (2016) with *in vitro* incubation of oat hay or mixed feed with graded doses (10- 120 µl 40 ml^{-1}) lemongrass oil (*Cymbopogon citrates*). However, feed digestibility and volatile fatty acids production were reduced at higher doses, suggesting general inhibitory effect on rumen microbes. The essential oil components (citral, limonene, citronellal, geraniol) present in lemongrass oil are the key factors for inhibiting methanogenesis. Therefore, essential oils could be used as antimethanogenic feed additive to reduce enteric methane emission from livestock, however, long term *in vivo* experiments with different feeding regimes need to be conducted before their use in animal ration.

5.3. Essential oil in improving health status of animals

Recently, essential oils are considered to have positive effects on health by reducing risk of cardiovascular diseases, inflammatory processes, and excessive production of free radicals in cell. Supplementation (0.05%) of *Salvia officinalis* essential oils, which contains mainly rosmarinic acid, diterpene phenolics carnosol and carnosic acid to broiler birds increased total antioxidant status, increased glutathione peroxidase enzyme with reduced lipid peroxidation (Ryzner et al., 2013). Seifzadeh et al. (2016), in a study with supplementation of medicinal plant mix (1.5%) to Holstein calves reported increased total antioxidant activity with reduced weaning age.

Unique study by Kumar (2017) demonstrated the increased antioxidant status of buffalo calves supplemented with a mixture of essential oil rich poplar and eucalyptus leaves. Increased concentration of major antioxidant enzymes (reduced glutathione, catalase, superoxide dismutase) and reduced lipid peroxidation as measured by MDA formation in erythrocyte were reported. The researcher also observed improved cell mediated immunity (CMI) in supplemented buffaloes by measuring skin fold thickness in delayed type of hypersensitivity (DTH) reaction (Fig. 2). Humoral immune response as measured by antibody titre against *Pasteurella multocida* was also higher, suggesting positive role of essential oil rich feed additive in improving overall immune status of buffalo calves.

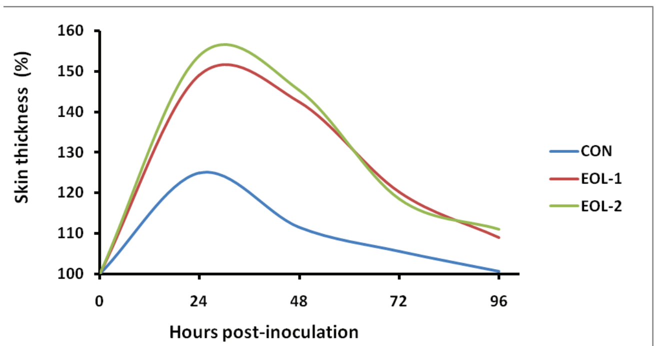

Figure 2

5.4. Essential oil in improving livestock production and product quality

The reports on feeding of essential oils on livestock performance is not consistent and largely depends on feed intake and the components of essential oils. Yang et al. (2007) observed that addition of garlic (*Allium sativa*, 5 g day^{-1}) and juniper berry (*Juniperus communis*, 2 g day^{-1}) oils to dairy cow diets had no effect on feed intake, milk production or milk composition. The increased in milk fat percent without affecting milk yield was demonstrated by Santos et al. (2010) by feeding an essential oil mixture containing eugenol, geranyl acetate and coriander oil. However, Santos et al. (2010) also reported increased milk yield on feeding of commercial essential oil mixture (Crina). Rumen microbes involved in biohydrogenation of fatty acids could be affected by feeding of essential oils, due to specific antimicrobial activities of some essential oils. Benchaar et al. (2007) reported an increased concentration of health promoting, conjugated linoleic acids (CLA) in milk fat by feeding of a mixed essential oils (MEO) at 2g per day, however, no changes reported at lower dose of supplementation. Singh (2016) fed a mixture of *Ficus bengalensis* (Bargad) leaves, *Sapindus mukrossi* (Reetha) fruits and eucalyptus essential oils to buffalo calves and observed increased growth rate and feed efficiency. However, Kumar (2017) reported no effects on body weight gain in buffalo calves fed a blend of essential oil rich poplar *(Populus deltoids)* and eucalyptus *(Eucalyptus citriodora)* leaves. These studies suggest the factors like feed composition, essential oil components, their dose and delivery to the system as well as synergy among other bioactive compounds in modulation of feed intake and body weight changes in livestock.

The components of essential oils and their metabolites could be present in milk and meat and improve the organoleptic and nutritional properties of animal products. Molnar et al. (1997) reported limonene and carvone, the main essential oil components of caraway seed in goat milk after feeding caraway seed. Various terpenoids have been reported in milk of cows grazed on aromatic plants (Chion et al., 2010). This area of research needs to be strengthened for human health benefit.

6. Conclusion

EOs and their components reduce entric methane and ammonia emission by modulating rumen microbial community structure and could be used as phytogenic feed additive. However, their effects need to be studied by long term *in vivo* experimentation, as adaptation of rumen microbes could lead to ineffectiveness. Optimum dose of bioactive essential oil components and their appropriate combinations needs to be standardized in relation to dietary composition of animals to achieve consistent benefit of their feeding for livestock health and performance before their application in large scale livestock production systems.

6. References

Benchaar, C., Petit, H., Berthiaume, R., Ouellet, D., Chiquette, J., Chouinard, P., 2007.

Effects of essential oils on digestion, ruminal fermentation, rumen microbial populations, milk production, and milk composition in dairy cows fed alfalfa silage or corn silage. Journal of Dairy science 90, 886-897.

Chion, A.R., Tabacco, E., Giaccone, D., Peiretti, P.G., Battelli, G., Borreani, G., 2010. Variation of fatty acid and terpene profiles in mountain milk and "Toma piemontese" cheese as affected by diet composition in different seasons. Food Chemistry 121, 393−399

Dey, A., Dutta, N., Sharma, K., Pattanaik, A., 2008. Effect of dietary inclusion of Ficus infectoria leaves as a protectant of proteins on the performance of lambs. Small Ruminant Research 75, 105−114.

Dey, A., Paul, S.S., Dahiya, S.S., Punia, B.S., 2016. Garlic oil supplementation: effects on in vitro methanogenesis, rumen fermentation and gas production in buffaloes, In: Sreekumar, D., Jacob, N., Mahender, M., Rajanna, N. (Eds.), International Livestock Conference and Expo (INDIGENOUS) and 23rd Annual Convention Indian Society of Animal Production and Management (ISAPM) Prof. Jayashankar Telangana State Agricultural University, Hyderabad, Telangana, p. 41.

FAO, 2010. Greenhouse gas emissions from the dairy sector. A life cycle assessment, FAO Rome.

Fernandez-Lopez, J., Zhi, N., Aleson-Carbonell, L., Perez-Alvarez, J., Kuri, V., 2005. Antioxidant and antibacterial activities of natural extracts: application in beef meatballs. Meat science 69, 371−380.

Gershenzon, J., McConkey, M.E., Croteau, R.B., 2000. Regulation of monoterpene accumulation in leaves of peppermint. Plant Physiology 122, 205−214.

Greathead, H., 2003. Plants and plant extracts for improving animal productivity. Proceedings of the Nutrition Society 62, 279-290.

Hart, K., Yanez-Ruiz, D., Duval, S., McEwan, N., Newbold, C., 2008. Plant extracts to manipulate rumen fermentation. Animal Feed Science and Technology 147, 8−35.

Kumar, K., 2017. Effects of feed additives rich in essential oils on rumen fermentation, methanogenesis and nutrient utilization in buffalo, ICAR- Central Institute for Research on Buffaloes, M.V.Sc. thesis submitted to Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar, India.

Macheboeuf, D., Morgavi, D., Papon, Y., Mousset, J.-L., Arturo-Schaan, M., 2008. Dose–response effects of essential oils on in vitro fermentation activity of the rumen microbial population. Animal Feed Science and Technology 145, 335−350.

Min, B., Pomroy, W., Hart, S., Sahlu, T., 2004. The effect of short-term consumption of a forage containing condensed tannins on gastro-intestinal nematode parasite infections in grazing wether goats. Small Ruminant Research 51, 279−283.

Molnar, A., Lemberkovices, E., Spiller, S., 1997. Detection of caraway and chamomile components in goat milk. Tejqazdasaq 57, 22−27.

Patra, A.K., 2014. Trends and projected estimates of GHG emissions from Indian livestock in comparisons with GHG emissions from world and developing countries. Asian-Australasian journal of animal sciences 27, 592.

Ryzner, M., Takacova, J., Cobanova, K., Placha, I., Venglovská, K., Faix, S., 2013. Effect of dietary Salvia officinalis essential oil and sodium selenite supplementation on antioxidative status and blood phagocytic activity in broiler chickens. Acta Veterinaria Brno 82, 43−48.

Santos, M., Robinson, P., Williams, P., Losa, R., 2010. Effects of addition of an essential oil complex to the diet of lactating dairy cows on whole tract digestion of nutrients and productive performance. Animal feed science and technology 157, 64−71.

Seifzadeh, S., Aghjeh-Gheshlagh, F.M., Abdi-Benemar, H., Seifdavati, J., Navidshad, B., 2016. The Effects of a Medical Plant Mixture and a Probiotic on Performance, Antioxidant Activity and Weaning Age of Newborn Holstein Calves. Iranian Journal of Applied Animal Science 6.

Singh, R.K., 2016. Effect of some plant bio-active compounds on methane production, growth and nutrient utilization in buffaloes, DCN Division, M.V.Sc. thesis submitted to the ICAR- National Dairy Research Institute, Karnal, India.

Singh, R.K., Dey, A., Punia, B.S., Paul, S.S., Singh, M., 2016. Influence of lemongrass (Cymbopogon citratus) essential oils on in vitro methanogenesis, fatty acids composition and fermentation of feeds in buffalo, X Biennial Animal Nutrition Conference of Animal Nutrition Association Sri Venkateswar Veterinary University, Tirupati, p. 194.

Singh, R.K., Dey, A., Punia, B.S., Paul, S.S., Singh, M., 2017. Influence of plant secondary metabolites and their blends on in vitro methanogenesis, fatty acids composition and fermentation of feed in buffalo Animal Feed Science and Technology (Submitted).

Ultee, A., Bennik, M., Moezelaar, R., 2002. The phenolic hydroxyl group of carvacrol is essential for action against the food-borne pathogen Bacillus cereus. Applied and environmental microbiology 68, 1561−1568.

Wallace, R.J., McEwan, N.R., McIntosh, F.M., Teferedegne, B., Newbold, C.J., 2002. Natural products as manipulators of rumen fermentation. Asian Australasian Journal of Animal Sciences 15, 1458−1468.

Wanapat, M., Cherdthong, A., Pakdee, P., Wanapat, S., 2008. Manipulation of rumen ecology by dietary lemongrass (Stapf.) powder supplementation. Journal of animal science 86, 3497−3503.

Yang, W., Benchaar, C., Ametaj, B., Chaves, A., He, M., McAllister, T., 2007. Effects of garlic and juniper berry essential oils on ruminal fermentation and on the site and extent of digestion in lactating cows. Journal of dairy science 90, 5671−5681.

