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The Effects of Use Medicinal Plants on Rumen Fermentation Parameters in Ruminants

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ABSTRACT

Rumen is a persistent and specific ecosystem consists of bacteria, protozoa and fungus where feed fermentation takes place in it. Produced Hydrogen in rumen can be used in the synthesis of the volatile fatty acids and the microbial protein and its excess would be eliminated through the production of Methane by methanogenesis. Nutritionists have tried to find ways to decrease loss and energy and protein, increase synthesis of microbial protein and the ability of fiber digestion in the rumen through manipulating the rumen's fermentation processes. Hence, changing microbial ecosystem of the rumen by the use of proper feed additives could lead to the reduction of Methane and Nitrogen production and excretion and improve the performance of Ruminants. According to accomplished studies Ionophores increase the efficiency of the use of energy and the protein of feed effectively in rumen but with regard to antibiotic usage ban by European Union in January 2006 and public acceptability reduction of using these compounds in animal nutrition, animal nutritionists decided to use other alternative compounds, spending rumen microbial population control and creating proper changes in rumen fermentation. In recent years the use of herbs as growth stimulus is increasingly considered. The results of this study demonstrate that using of medicine plants effects positively rumen performance and pH, methane production reduction and ammoniac nitrogen concentration, protein metabolism and rumen microbial population improvement and volatile fatty acids production increase.

Key words: Rumen, Herbs, Ruminants, Performance, Ammonia nitrogen

INTRODUCTION

According to the fact that some antibiotics used in ruminants nutrition caused creating resisting microbial strains and transferring this resistance to other species specially in common strains between animal and human, persisting of drug residues in productions using by human and disarranging digestive tract population balance, using this alternative in ruminants ration has been doubted (Marashi sarai, 2007). Therefore it is necessary to limit the usage of antibiotics in ration and find proper alternatives for them. In recent years herbal compounds have attracted many researchers' attentions to themselves for their

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potential role in alternating growth stimulus antibiotics in animal nutrition. This group consists of material derived from medicine plants or species which have positive effect on animal health and production (Hoson et al, 1999). Nowadays one of the crises of human society is nutrition and food shortage which increases in population growing adds to the crisis. While one of the health principles of human society is good nutrition, then to achieve this goal it is essential to increase in producing agricultural productions specially protein resources which are considered as one of the most important nutrition resources. Because protein resources are mostly provided from domestic animals, one of the ways to increase the production among these animals is to manipulate ruminant's rumen ecosystem and apply new scientific and practical techniques with connection to animal breeding and nutrition which could operate better with animal health preservation and lower costs. There are plenty of researches done in association with various techniques of manipulating rumen microbial ecosystem for the purpose of improvement in ruminant's production efficiency (Taghizadeh et al, 2010). One of the most important ways to achieve the goal and create economic optimal condition is to use herbal essential oils in animal ration for the purpose of manipulating rumen. Essential oils are aromatic volatile compounds extracted from plants which their secondary metabolites are usually consisting of terpenoids and phenylpropanoids. Herbal essential oils contain antimicrobial characteristics which could effect on inappropriate microbes of rumen. Table 1 demonstrates antimicrobial activities of some essential oils and their main active compounds. Accordingly essential oils are taking into consideration among lots of nutritionists and rumen microbiologists for the purpose of extracting oils as natural feed additives for rumen fermentation improvement such as producing rumen volatile fatty acids, preventing methanogenesis, improving protein metabolism and feed efficiency and increasing conjugated linoleum acids (CLA) in ruminants that is derived from their feed (Alman, 2011). Protein degradation in rumen by microbes causes producing ammonia and peptides and amino acids are as intermediates in this process (Reynal et al, 2007). Chalupa (1975) stated that the most important metabolic characteristics of true protein in rumen are to provide peptide nitrogen. Peptides are intermediate compounds of protein metabolisms in rumen and their concentration may temporarily be high in rumen (Chen et al, 1992). Rumen microbes need 3 nitrogen sources included ammoniac nitrogen sources, amino acid nitrogen and peptide nitrogen to synthesis of microbial protein (Russell et al, 1983; Sannes et al, 2002). It is important to adjust ammonia concentration in rumen to digest fiber and synthesis of microbial protein, but extra produced ammonia in rumen may cause reduction in efficiency of the protein use in lactating cow's ration (Chen et al, 1992). Hart et al. (2007) reported in their study that essential oils apply their main effects on rumen through reduction of protein and starch the degradation and prevention of amino acids the degradation which their effects depend on dose and chemical structure of essential oils, ration combinations, animal physiology and pH. Results of some studies showed that garlic oils, cinnamaldehyde (its main active compound is cinnamon), eugenol (its main active compound is from chili peppers) and anise oils could increase the production of propionate and decrease the production of acetate or methane, protein the degradation, peptides and amino acids (Busquet et al, 2006). It seems that evaluation of Iran medicine plants which antibiotic effects are apparent is necessary for using them in ruminant's nutrition. Thus the goal of this study is to investigate effects of medicine plants usage on rumen fermentation parameters.

Table 1. Essential oils with antimicrobial activity, their main active components, and susceptible microorganisms

Essential oil	Name	Active components	Susceptible microorganisms
Allium sativum	Garlic	Allicin, diallyl sulfit	Enteropathogenic bacteria
Anethum graveolens	Dill	Limonene, carvone	Gram-positive and gram-negative bacteria
Capsicum annum	Paprika	Capsaicin	Gram-positive and gram-negative bacteria
Cinnamomum cassia	Cassia	Cinnamaldehyde	*Escherichia coli, Staphylococcus aureus, Listeria monocylogenes, Salmonella enteritidis*
Juniperus oxycedrus	Juniper	Cadinene, pinene	*Aeromonas sobria, Enterococcus fecalis, Staph aureus*
Melaleuca alternifolia	Tea tree	Terpinen-4-ol	*Staph. aureus, E.coli, gram-positive and gram-negative bacteria*
Origanum vulgare	Oregano	Carvacrol, thymol	Gram-positive and gram-negative bacteria
Pimpinella anisum	Anise	Anethol	*Aeromonas hydrophila, Brevibacterium linens, Brochothrics thermosphacta*
Rosmarinus officinalis	Rosemary	1,8-Cineole	*Staph. aureus, L.monocytogenes, Campylobacter jejuni*
Syzygium aromaticum	Clove	Eugenol	*E.coli, Staph. Aureus, L. monocytogenes, S.enteritidis, C.jejuni*
Thymus vulgaris	Thyme	Thymol, carvacrol	*Salmonella typhimurium, Staph. aureus, Aspergillus flavus*

ORIGIN AND CLASSIFICATION OF ESSENTIAL OILS

Essential oils are blends of secondary metabolites obtained from the plant volatile fraction by steam distillation (Gershenzon et al, 1991). The term “essential” derives from “essence,” which means smell or taste, and relates to the property of these substances of providing specific flavors and odors to many plants. They are characterized as having a very diverse composition, nature, and activities. The most important active compounds are included in 2 chemical groups: terpenoids (monoterpenoids and sesquiterpenoids) and phenylpropanoids.

TERPENOIDS

Without doubt one of the biggest secondary compounds groups is terpens and terpenoids. More than 22 thousand types of these compounds are reported up to now (Connolly et al, 1991). Every year researchers recognize about 1000 types of these compounds with new structures in organic herbs. Most researchers pay special attention to synthesis and production of terpens and terpenoids in herbs due to medical and industrial importance and value of these compounds. Generally terpenoids (C_5-C_{20}) take substantial responsibilities in plants such as growth, evolvement, reproduction and defending against natural enemies. The characteristic of terpenoids compounds is that the base structure of them is derived from 5 carbons which are usually called unit isoperne. Classification of terpenoids depends on the number of the units in their structures. There are important compounds of essential oils inside terpenoids which most of plants are belong to terpenoids and Sesquiterpenoids family (Castillejos, 2007).

PHENYLPROPANOIDS

Phenylpropanoids are not the common compounds of essential oils but some plants have them in considerable amounts. The term phenylpropanoid refers to compounds with chain of 3 carbons which are adjoined to an aromatic ring of 6 carbons (Sangwan et al, 2001). The results of investigations show that these compounds play an important role in plants compatibility and the increase of their competition in limited resources like light, water, nutrition, space and … with other plants species.

EFFECTS OF ESSENTIAL OILS ON RUMEN FERMENTATION

Ruminant's performance depends directly on rumen ecosystem and microbial population activity. Rumen is a complicated ecosystem where numerous microorganisms in digestive tract of ruminants are there. These microorganisms are only with rumen microbial population and there is a real symbiotic relationship between them and the host animal. Microbes, including bacteria, protozoa and anaerobic funguses are mainly dependent to the ruminant host that provides necessary physiological condition for open survival (Jafari Khorshidi et al, 2009). Rumen acts like a fermentation vat. Carbohydrates fermentation takes place in rumen which produces ATP, provides required energy for rumen bacteria (Nocek et al, 1980). Using of essential oils in ruminants ration was firstly reported in 60s (Borchers, 1965). Whereas the use of antibiotics in animal feed started from 70s. After those using antibiotics as feed additives was banned in European Union. In most cases the positive effects of essential oils such as increase in propionates and decrease in acetate , methane and production of ammonia nitrogen have been reported without decreasing the total production of volatile fatty acids. Busquet et al. (2006) reported that in vitro condition carvacrol (2.2 mg/l) decreases increase in ammonia nitrogen concentraion 2 h after feeding. They suggested that carvacrol prevents protein degradation or stimulation of lipid breakdown. Brochers (1965) at first reported that thymol prevents amination. Cardozo (2005) reported that the lowest concentrations of eugenol, 0.3 and 3 mg/L tended to increase the ammonia concentration whereas the intermediate level of 30 mg/L had no effect, and the higher levels of 300, 3000 and 5000 mg/L significantly reduced ammonia in an in vitro batch culture system. Castiilejoes et al. (2007) found that total volatile fatty acid production was unaffected on post feeding of essential oils, but it increased 3 h post feeding in sheep. In another study by Newbold et al. (2004) found that 110 mg/d essential oils tended to stimulate total volatile fatty acid production 6 h after feeding sheep. This researcher showed in the results that mixture of essential oils decreases the speed of soybean meal degradation in sheep rumen. Study results of Castiilejoes et al. (2006) showed that increase of thymol in ration increases acetate to propionate ratio in ration consist of 60% forage and 40 % concentrate and leads to increase in rumen pH to 6.4. Laboratory studies have shown that garlic oils decreases production of methane and rumen volatile fatty acids from 0.2 to 0.05 (Van Nevel and Demeyer, 1988). Cardozo et al. (2004) suggested that garlic oils prevent deamination while reported other variable effects. Ferme et al. (2004) also tested the effects of garlic oils using rumen fluid and a high concentrate diet typically found in feedlot beef at different pH (7.0 vs 5.5). While at pH 7.0 garlic oils resulted in lower ammonia-N and total volatile fatty acids concentrations, at pH 5.5 ammonia-N concentrations was also reduced. But total volatile fatty acids concentration and propionate proportion increased, and acetate proportion and the acetate to propionate ratio decreased compacted with a control group.

EFFECTS OF VOLATILE ESSENTIAL ON RUMEN MICROBIAL POPULATION

An important part of digestive process in matured ruminants is established on rumens microorganisms (Tabatabai, 2003). Microbial population of rumen is adjusted by an especial ecological balance ruling there. Number of factors impact on rumen fermentation which can be named as amount and composition of feed which are external changes and effect on digestion amount and passage rate and rumen contents heterologous (Abbas Pour et al, 2008). There are usually 10^{10} to 10^{11} bacteria, $10^{5}$to 10^{6} protozoa and 10^{3} to 10^{5} funguses per milliliter of ruminant's rumen fluid (Hopson, 1963). There are certain differences in conspicuity and amount of rumen microbes between ruminant's species and even for similar host species in different places which activities lead to the production of volatile fatty acids and methane due to anaerobic conditions (Dehority et al, 1997).

EFFECTS ON RUMEN PROTOZOA

Rumen ciliated protozoa are a part of natural microorganisms of ruminant's digestive tract which their biodiversity can be affected by factors such as nutrition, physiological condition, and age and sustenance condition. Nutritional stresses like applying feed restrictions or acidosis could lead to animal deprotozoa. Yet protozoa are found in animal normal condition in rumen and numbers of them are in the range of 10^{5} to 10^{6} in each milliliter of rumen fluid. The importance of protozoa presence in rumen ecosystem can be due to effects on digestive ability of feed nutrition, rumen environmental conditions and rumen microbial population. Yet protozoa unlike bacteria are not necessary for survival and development of ruminants host digestive tract and application of some feed rations for deprotozoa could leads to increase in animal growth and production (Gocmen and Dehority, 2001). Existing protozoa in ruminant's rumen belong to two groups of flagellates and ciliates which are the most significant and important rumen protozoa (Williams and Coleman, 1998). Many reports have been reported on the effect of essential oils on rumen protozoa population. Benchaar et al. (2003) reported that feed 750 mg/d from composition of essential oils in dairy cattle had no effect on the number of determined protozoa in vivo condition. Cardozo et al. (2006) reported in their studies that adding a combination of cinnamaldehyde (180 mg/d) and eugenol (90 mg/d) to feedlot cattle increases the number of Holotrishes and has no effect on the number of Antodinomorphs. Also no effect observed on the number of protozoa species when a combination of essential oils included higher concentration than cinnamaldehyde (600 mg/d) and eugenol (300 mg/d) used in feedlot cattle nutrition. Ando et al. (2003) showed that feeding 200 mg dried mint to cannula attached cows meaningfully decreases the number of protozoa to 50%. Fraser et al. (2007) in investigation on cinnamon leaf oils effect on rumen microbial fermentation in 2 continuous culture systems observed significant reduction in the number of rumen protozoa in the group which received cinnamon leaf oils in compare with the control group. In another study by Yong et al. (2010) feeding 2 g/d anise extract to heifers including 100 g/kg anitol decreases the number of Holitrish and Anthodinomorph protozoa. Patra et al. (2010) concluded in their experiments that anis extract has no effect on the number of rumen protozoa.

EFFECTS ON RUMEN BACTERIAL NUMBERS

Essential oils can inhibit extra ammonia producer bacteria which results in reduction of amino acids deamination (Wallace, 2004). McIntosh et al. (2003) observed that essential oils of compounds can inhibit the growth of certain extra ammonia producer bacteria (Clostridium Acetic Lundi and Peptostreptococcus Anaerobius) but other extra ammonia producer bacteria are less sensitive to essential oils (Clostridium Aminophilus). Nagy et al. (1964) reported that extracted essential oils from certain plant species can significantly reduce rumen bacterial activities in sheep and deer through accumulation of gas production. Evans and Martin (2000) reported that thymol inhibit ruminants selenomonas growth selectively.

EFFFCTS OF ESSENTIAL OILS ON RUMEN VOLATILE FATTY ACIDS

Rumen acts like a fermentation vat. Carbohydrates fermentation takes place in rumen and provides required energy for bacteria (Williams et al, 1998). Anaerobic nature of rumen fermentation produced volatile fatty acids. In ruminants most of the energy used for maintenance and production performance is provided by volatile fatty acids (Allen, 1997). The most important volatile fatty acids have been formed in rumen during the fermentation of acetate, propionate and butyrate (Van Houtert, 1993). Rumen estate like pH and the type of substrate is affected by produced volatile fatty acids and through this way the activity of certain rumen microorganisms groups' changes. During the carbohydrates fermentation, intermediate monomers are formed and also they catabolized to Pyruvate (Van Houtert, 1993). After digesting rapid fermentation feed, microbial activities increased quickly which results in volatile fatty acids concentration increase and rumen pH decrease. Different reports show that volatile fatty acids concentration becomes more than double from the starting point of feeding to their peak. According to these reports the lowest percentage of change among volatile fatty acids have been orderly for butyrate, acetate, propionate according to various investigations acetate, propionate and butyrate mean of molar percentage is estimated 64.3, 19.8 and 11 percent (Chuerch, 1979). Total concentration of volatile fatty acids in rumen usually varies decrease or less especially when essential oils concentration is higher (Chaves et al, 2008). Cardozo et al. (2006) in investigation on the effect of anise oils in ration of growing heifers fed by ration consisted of 10% forage and 90% concentrate, concluded that total concentration of volatile fatty acids didn’t change but the amount of acetate molar decreased and the amount of propionate molar increased and acetate to propionate of ration is decreased. Busquet et al. (2006) showed in their studies that garlic oil decreases acetate amount and increases butyrate and propionate amount. Some studies showed that ration supplementation with cinnamaldehyde by 0.2 g/kg DMI or with extracted essential oils from oregano by 0.25 g/kg DMI increases total volatile fatty acids in rumen (Kozelov et al, 2001). On the other hand the response of essential oils to volatile fatty acids total concentration could be in associated with the type of substrate fed to ruminants. In another study by Benchaar et al. (2007) no changes were observed in total concentration of volatile fatty acids in dairy cows fed by rations based on alfalfa silage but in rations based on corn silage, addition of 0.75 g essential oils combination per day decreased fatty acids concentration. Castillejoes et al. (2006) reported that low doses of thymol (50 mg/l) had no effects on in in vitro rumen microbial fermentation, but at higher doses (500 mg/l) total volatile fatty acids and ammonia-N concentrations decreased, and the acetate to propionate ratio increased. Also this researcher reported that thymol increases acetate to propionate ratio in rations consist of 60 % forage and 40 % concentrate. Giannenas et al. (2011) in the investigation of essential oils effects on production and milk combinations and rumen microbial population in lactating ewes concluded that the amount of volatile fatty acids showed that acetate, butyrate, valerate, fatty acids with branched chains and total concentration of volatile fatty acids did not change among the experimental groups. Their observations showed that propionate production in ewes fed by amounts of 100 and 150 mg essential oils linearly increased compare to other experimental groups. Also acetate to propionate ratio in highest and lowest control group is observed in ewes fed by 150 mg/kg essential oils. As well as lowest amount of acetate was related to ewes fed by ration contains 150 mg essential oils. These changes in volatile fatty acids concentration could be caused by change in rumen microbial population.

CONCLUSION

The results obtained from this study showed that herb oils has dissuasive activities against number of rumen's micro-organisms (involved in organic materials fermentation and then gas production) and addition of these herbs could improve the ration fermentation and volatile fatty acids production increase. It is recommended to investigate the effects of these herb oils in various levels and in combination with other oils and herbal compounds for microbial protein production and volatile fatty acids production.

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