THE EFFECTS OF SOME ESSENTIAL OILS ON RUMEN TOTAL BACTERIA AND PROTOZOA NUMBERS

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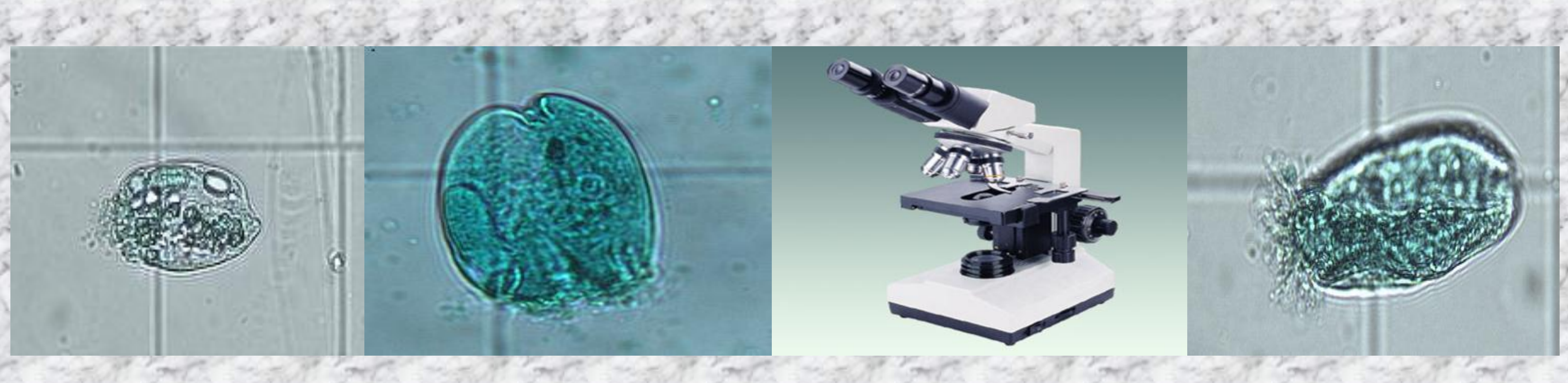
Introduction

Essential oils are plant secondary metabolites responsible for odour and colour of plants and spices. Because many essantiaal oils have strong antimicrobial properties, research to exploit essentaial oils as feed additives in animal nutrition has been accelerated recently due to ban of some antibiotic growth promoters (Patra et al., 2011). These products exhibit antimicrobial activity against gram-positive and gram-negative bacteria (Helander et al., 1998; Castillejos et al., 2004). Many essential oils have dose-dependent effects on bacteria, protozoa, and fungi (Greathead, 2003). In general, gram-positive bacteria appeared to be more susceptible to inhibition by plant essential oil compounds than did gram-negative bacteria (Davidson and Naidu, 2000). This effect has been related to the presence of an outer membrane on gram-negative organisms, which endows them with a hydrophilic surface that acts as a strong impermeability barrier (Nikaido, 1994 ; Wanapat et al., 2008). The aim of this study was to determine the effect of essential oils (EO) of oregano (ORE, *Origanum vulgare*), black seed (BSD, *Nigella sativa*), laurel (LAU, *Laurus nobilis*), cummin (CUM, *Cumminum cyminum*), garlic (GAR, *Allium sativum*), anise (ANI, *Pinpinella anisum*), cinnamon, (CIN, *Cinnamomum verum*), oleaster (OLE, *Eleagnus angustifolia*), mint (MNT, *Mentha longifolia*), rosemary (ROS, *Rosmarinus officinalis*), coriander (COR, *Coriandrum sativum*), grape seed (GRA, *Vitis vinifera*), orange peel (ORA, *Citrus cinensis*) and fennel (FEN, *Foenicum vulgare*) on the number of rumen microorganisms.



Material and Method

In vitro gas productions were determined in *in vitro* gas production technique (IVGPT) by supplying rumen liquor from three infertile Holstein cows. Then, rumen microorganisms were determined by using residual rumen liquor from IVGP. The study was carried out in a completely randomized design in factorial arrangement. Barley, soybean meal and wheat straw were incubated as substrat in *in vitro* gas production technique for 50, 100, 150 ppm doses were tested for all essential oils. Bacteria and protozoa counts were determined according to method of Dehority and Tirabasso (2001) and protozoa Dehority (1984), respectively (Figure 1). The data were analyzed statistically using the General Linear Model procedure of SPSS.

Figure1: Protozoa (a) and bacteria (b) under microscope

Conclusion

It is known that there is a symbiotic relative between protozoa and methanogenic bacteria. In conclusion, the highest total protozoa counts were determined at BSD for barley, wheat straw and soybean meals. For this reason, BSD can be used to reduce methanogenesis in the rumen. Besides, in vivo experiment is required to determine the effects of BSD and other EOs on methanogenesis. The results revealed that essential oils may change microbial population in rumen.

Results

The effects of some essential oils on rumen total bacteria and protozoa numbers were given in Table 1. The bacteria content of BSD treatment and protozoa contents BSD and MNT treatments for barley were found higher compared to other plant extracts. Total bacteria counts at MNT treatment for wheat straw were higher compared to other extracts. Bacteria counts for MNT were higher than those for other extracts. BSD ranked first in terms of protozoa counts. It was concluded that EO had significant effects on bacteria and protozoa counts ($P<0,05$), but EO doses had no significant effect ($P>0,05$).

Table 1. The effects of some essential oils on rumen total bacteria and protozoa numbers

		Barley		Wheat straw		Soy bean meal	
Esential Oils	Doses (ppm)	Bacteria($10x^7$)	Protozoa ($10x^5$)	Bacteria($10x^7$)	Protozoa($10x^5$)	Bacteria($10x^7$)	Protozoa($10x^5$)
Control	0	13.85^{c-l}±3.03	4.16^{c-i}±1.35	12.89^{c-f}±4.11	4.63^{d-j}±1.40	14.93^{b-j}±3.52	3.86^{d-i}±.99
BSD	50	22.91^{a}±1.54	6.88^{ab}±.68	17.12^{a-d}±1.27	7.36^{a-c}±.45	19.66^{a-c}±.38	6.4^{ab}±.00
	100	19.36^{ab}±5.16	6.08^{a-d}±.45	17.56^{a-c}±3.42	9.12^{a}±1.58	18.77^{a-d}±2.51	6.72^{a}±.45
	150	23.02^{a}±.57	6.24^{a-c}±.68	19.86^{ab}±1.52	7.52^{ab}±.23	18.45^{a-e}±.79	6.08^{a-c}±.45
ORE	50	8.59^{m}±1.06	4.8^{b-h}±.45	7.57^{f}±.21	5.92^{b-d}±.23	13.41^{d-j}±.81	4.16^{d-h}±.00
	100	8.91^{lm}±.88	4.16^{c-i}±.00	9.83^{ef}±.81	5.6^{b-f}±.23	13.26^{d-j}±.25	4^{d-h}±.23
	150	13.17^{c-m}±.20	4.00^{d-i}±.23	9.84^{ef}±.11	4.64^{d-j}±.23	13.15^{e-j}±.09	4^{d-h}±.23
MNT	50	13.33^{c-m}±.43	5.92^{a-e}±.23	21.57^{a}±.13	5.76^{b-e}±.00	20.70^{a}±1.18	4.96^{b-e}±1.13
	100	17.36^{bc}±1.65	7.2^{a}±.23	22.11^{a}±.04	5.28^{c-h}±.23	20.10^{ab}±.27	4.96^{b-e}±1.13
	150	12.50^{c-m}±.66	6.72^{ab}±.45	22.47^{a}±.05	5.12^{d-i}±.00	19.95^{ab}±.61	5.44^{a-d}±.00
ROS	50	13.62^{c-l}±.07	4.00^{d-i}±.68	9.75^{ef}±.62	4.8^{d-j}±.00	10.96^{ij}±.48	3.84^{d-i}±.00
	100	13.36^{c-m}±.48	3.52^{f-i}±.45	10.38^{ef}±.34	5.44^{b-g}±.45	12.74^{f-j}±.27	3.84^{d-i}±.45
	150	13.42^{c-m}±.66	4.32^{c-i}±.68	12.02^{c-f}±.66	4.8^{d-j}±.00	13.82^{d-j}±.91	3.2^{e-j}±.91
LAU	50	11.30^{g-m}±.54	4.8^{b-h}±.45	9.35^{ef}±.36	3.68^{d-j}±.23	11.57^{g-j}±1.20	3.68^{d-j}±.68
	100	11.14^{g-m}±.18	4.8^{b-h}±.45	9.76^{ef}±.23	4.16^{d-j}±.91	9.98^{j}±.27	3.68^{d-j}±.23
	150	12.03^{d-m}±.14	5.12^{b-f}±.00	9.91^{ef}±.43	4.16^{d-j}±.45	9.82^{j}±.05	4.32^{c-g}±.68
COR	50	11.58^{f-m}±.23	4.16^{c-i}±.45	8.60^{ef}±.21	5.6^{b-f}±.23	11.84^{f-j}±.00	3.52^{e-j}±.45
	100	11.97^{d-m}±.50	4.48^{c-i}±.91	9.91^{ef}±.25	5.6^{b-f}±.23	12.67^{f-j}±.86	1.92^{j}±2.72
	150	14.34^{c-k}±.23	3.84^{e-i}±.45	9.57^{ef}±.13	4.8^{d-j}±.91	13.71^{d-j}±.11	4.32^{c-g}±.68
CIN	50	15.02^{b-j}±.57	3.04^{f-i}±.68	14.77^{b-e}±.30	3.36^{f-j}±.68	16.58^{a-g}±.14	3.68^{d-j}±.23
	100	16.11^{b-g}±.02	3.52^{f-i}±.00	14.10^{b-e}±.02	3.36^{f-j}±.23	14.94^{b-j}±.05	3.04^{f-j}±.23
	150	15.79^{b-h}±.29	3.68^{f-i}±.23	14.83^{b-e}±.07	3.04^{h-j}±.23	16.54^{a-h}±.18	3.2^{e-j}±.45
GAR	50	11.78^{e-m}±.05	2.72^{hi}±.23	10.93^{d-f}±.16	2.72^{j}±.23	12.19^{f-j}±.68	2.88^{g-j}±.00
	100	10.29^{j-m}±.07	2.88g^{hi}±.00	10.90^{d-f}±.25	3.52^{e-j}±.00	12.06^{f-j}±.14	2.08^{ij}±.23
	150	13.58^{c-m}±.29	2.56^{i}±.00	11.23^{c-f}±.41	3.36^{f-j}±.23	13.17^{e-j}±.43	2.4^{h-j}±.23
OLE	50	16.74^{b-e}±.32	3.84^{e-i}±.45	12.23^{c-f}±.13	3.52^{e-j}±.00	13.57^{d-j}±.23	4^{d-h}±.23
	100	16.75^{b-e}±.38	3.68^{f-i}±.23	13.38^{c-f}±.09	3.84^{d-j}±.00	15.62^{a-i}±.27	3.84^{d-i}±.00
	150	16.85^{b-d}±.02	3.68^{f-i}±.23	13.02^{c-f}±.00	3.52^{e-j}±.45	16.35^{a-i}±.09	3.36^{e-j}±.23
GRA	50	12.32^{d-m}±.36	3.2^{f-i}±.00	10.64^{ef}±.20	2.72^{j}±.23	12.98^{e-j}±.11	3.52^{e-j}±.45
	100	12.61^{c-m}±.18	3.36^{f-i}±.23	11.15^{d-f}±.07	3.2^{g-j}±.45	12.62^{f-j}±.25	3.36^{e-j}±.23
	150	12.85^{c-m}±.16	3.84^{e-h}i±.45	11.68^{c-f}±.59	3.52 ±.00	12.4^{f-j}±.20	3.52^{e-j}±.00
CUM	50	16.34^{b-f}±.48	3.36^{f-i}±.23	13.19^{c-f}±.09	3.52^{e-j}±.00	16.83^{a-g}±.54	3.52^{e-j}±.00
	100	15.30^{b-j}±.41	4^{d-h}i±.23	10.51^{ef}±.11	4.32^{d-j}±.68	17.2^{a-f}±.66	3.84^{d-i}±.00
	150	15.28^{b-j}±.20	3.36^{f-i}±.23	13.43^{c-f}±.02	3.84^{d-j}±.45	16.22^{a-i}±.14	3.68^{d-j}±.23
ANI	50	15.47^{b-i}±.20	4^{d-i}±.23	14.31^{b-e}±.09	2.88^{ij}±.45	16.91^{a-g}±.43	3.52^{e-j}±.00
	100	15.46^{b-i}±.09	3.68^{f-i}±.68	14.3^{b-e}±.00	3.68^{d-j}±.23	16.78^{a-g}±.61	3.84^{d-i}±.00
	150	15.06^{b-j}±.43	3.2^{f-i}±.00	14.76^{b-e}±.09	3.68^{d-j}±.23	15.36^{a-j}±.05	3.52^{e-j}±.45
ORA	50	10.88^{h-m}±.32	3.36^{f-i}±.23	12.36^{c-f}±.09	4.48^{d-j}±.00	15.216^{b-j}±.11	3.2^{e-j}±.00
	100	11.76^{e-m}±1.52	4^{d-i}±.23	11.49^{c-f}±.04	3.68^{d-j}±.23	14.42^{c-j}±.20	3.52^{e-j}±.45
	150	12.03^{d-m}±.36	4.16^{c-i}±.00	11.57^{c-f}±.30	4^{d-j}±.68	13.65^{d-j}±.11	4^{d-h}±.23
FEN	50	10.64^{i-m}±.88	4.48^{c-i}±.45	9.75^{ef}±.84	4.8^{d-j}±.45	10.99^{h-j}±.48	4.8^{b-f}±.45
	100	10.67^{i-m}±.84	4.96^{b-g}±.68	9.89^{ef}±.77	4.8^{d-j}±.45	13.44^{d-j}±.18	4.64^{c-g}±.23
	150	9.92^{k-m}±.68	4.8^{b-h}±.45	10.47^{ef}±.36	4.16^{d-j}±.00	11.78^{f-j}±1.09	3.84^{d-i}±.00

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