



1                   **Comparing chemical composition and phenolic**  
2                   **compounds of some herbals as potential feed additives in**  
3                   **ruminant Nutrition**

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16                  **Abstract.** The chemical composition and phenolic compounds of eight herbals (*Alpinia of-*  
17                  *ficinarum*, *Artemisia absinthium*, *Cuminum cyminum*, *Dittrichia viscosa*, *Mentha rotundifolia*  
18                  L., *Nigella sativa*, *Rosmarinus officinalis* L and *Zingiber officinale*) were evaluated. Feed com-  
19                  ponents were determined by proximate analysis, whereas phenolic and tannin compounds were  
20                  analysed by colorimetric procedures. The crude protein content of the herbal samples varied  
21                  widely, being particularly high for *Nigella sativa* and low for *Alpinia officinarum* and *Rosma-*  
22                  *rinus officinalis*. The highest contents of Total extractable phenols (TEP) and Total extractable  
23                  tannins (TET) were observed in the *Cuminum cyminum* and *Dittrichia viscosa* whereas herbals,  
24                  *Nigella sativa* and *Zingiber officinale* showed lower concentrations. The tannin concentration  
25                  varied considerably between species, but in general the plants investigated in this study had low  
26                  tannin contents (except for *Alpinia officinarum*). Based on the results above, it could be con-  
27                  cluded that a large reserve of herbal species in the local flora is available that could be poten-  
28                  tially used as additives for livestock feeding. These herbs appear to be promising alternatives to  
29                  antibiotics in altering rumen fermentation and reducing methane production in ruminants.

30                  **Keywords:** herbals, plant secondary compounds, rumen fermentation, tannins

31                   **Introduction**

32                  Following the trends in human health care towards herbal medicinal products and  
33                  plant derived dietary supplements also in Veterinary medicine and livestock produc-  
34                  tion an increasing use of herbs, essential oils and plant extracts can be observed.  
35                  Herbs offer a new perspective in the strategy to achieve lower antibiotic use on farm,  
36                  both to contrast antibiotic resistance and to reduce veterinary bills. Animal nutrition-  
37                  ists tested the herbs or their extracts in order to improve the quality of the meat, the  
38                  oxidative stability in particular, (Smeti et al., 2018), to improve fibre digestibility, and  
39                  also to reduce CH<sub>4</sub> emissions and N excretion (Patra et al., 2006).

40                  Plant herbs such as garlic, lemongrass and peppermint are widely used as antibacterial  
41                  agents and extensively used to maintain the microbial ecosystem of the gastrointesti-  
42                  nal tract especially in tropical regions. Despite their potential as feeds, most herbals

43 contain large amounts of tannins, which have most likely been evolved by plants as a  
 44 defense mechanism against being consumed by herbivores. The presence of tannins at  
 45 high level in plants often limits their utilization as feedstuffs (Medjekal et al., 2018).  
 46 The anti-nutritive effects of tannins are associated with their ability to combine with  
 47 dietary proteins, polymers such as cellulose, hemicellulose and pectin, and minerals  
 48 thus retarding their digestion (McSweeney et al., 2001). As there is little information  
 49 regarding these aspects for locally available herbs so the study was conducted to  
 50 compare chemical composition and phenolic compounds of the different species of  
 51 herbs as potential feed additives in ruminants animals.

## 52 **1 Materials and Methods**

53 Eights herbals were used in this study: *Alpinia officinarum* (Rhizom) (*A. officinarum*)  
 54 (*Artemisia absinthium* (aerial part) (*A. absinthium*), *Cuminum cyminum* (seeds)  
 55 (*C. cyminum*), *Dittrichia viscosa* (aerial part) (*D. viscosa* *Mentha rotundifolia* L. (aerial  
 56 part) (*M. rotundifolia* L.), *Nigella sativa* (seeds) (*N. sativa*), *Rosmarinus officinalis*  
 57 L. (*R. officinalis* L.) (leaves) and *Zingiber officinale* (Rhizom) (*Z. officinale*). The  
 58 freeze dried samples were ground in a Willey Mill to pass through 1mm sieve for the  
 59 determination of chemical composition. Feed samples were analysed for Dry matter  
 60 (DM) and following the method of AOAC (2000). Nitrogen was determined using the  
 61 micro-Kjeldahl method (AOAC 2000). Crude Protein (CP) was calculated as N x  
 62 6.25. The Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF) and Acid De-  
 63 tergent Lignin (ADL) were analyzed according to Van Soest et al. (1991) using the  
 64 ANKOM Fiber Analyzer (ANKOM Technology, Fairport, NY). Both fibre fractions  
 65 were expressed including residual ash. Total extractable phenols (TEP) were deter-  
 66 mined according to the method of Julkunen-Tiitto (1985) using the Folin-Ciolateau  
 67 reagent and tannic acid as standard. Total extractable tannins (TET) were estimated  
 68 indirectly after adsorption of TEP to insoluble polyvinylpyrrolidone, and measuring  
 69 the remaining total phenols in the supernatant (Makkar et al., 1993). Concentration of  
 70 TET was calculated through subtraction as follows  $TET = TEP - \text{non-precipitable}$   
 71 phenols. Free condensed tannins were measured in the extract using the butanol-HCl  
 72 assay (Porter et al., 1986), with the modifications of Makkar (2003) and using purified  
 73 quebracho tannin as standard. All chemical analyses were performed in triplicate.

### 74 **Results**

75 The crude protein content of herbals samples varied widely; it ranged between 47 and  
 76 351 g kg<sup>-1</sup> DM, being particularly high for *N. sativa* and low for *A. officinarum* and  
 77 *R. officinalis*. The lowest NDF and ADF content (199 and 131 g kg<sup>-1</sup> DM) was found  
 78 in *D. viscosa* and the highest (517 and 272 g kg<sup>-1</sup> DM) in *C. cyminum*. The TEP con-  
 79 tent varied widely from 40.33 g kg<sup>-1</sup> DM g in *N. sativa* to 124.70 g kg<sup>-1</sup> DM in *D.*  
 80 *viscosa*, whereas the content of TET ranged from 25.06 g kg<sup>-1</sup> DM in *R. officinalis* to  
 81 82.02 g kg<sup>-1</sup> DM in *C. cyminum*. The highest contents of FCT and TCT were recorded  
 82 for *A. officinarum* whereas *Z. officinale* showed lower concentrations. The FCT and  
 83 TCT varied widely from 4.48 to 15.74 g kg<sup>-1</sup> DM to 41.90 and 386.34 g kg<sup>-1</sup> DM,  
 84 respectively.

85 **Table 1.** Chemical composition and Phenolic compounds (g kg<sup>-1</sup> dry matter) of Herbal plants

86

87 CP: Crude protein; NDF: Neutral detergent fibre; ADF: Acid detergent fibre; ADL: Acid deter-  
 88 gent lignin; TEP: Total extractable phenols; TET: Total extractable tannins; FCT: Free con-  
 89 densed tannins; TCT: Total condensed tannins.

Botanical name	CP	NDF	ADF	TEP	TET	FCT	TCT
<i>A. officinarum</i>	47±4.07	440±1.32	252±2.57	60.73±6.08	39.02±5.43	41.90±3.75	386.34±13.28
<i>A. absinthium</i>	228±11.44	266±5.32	189±1.46	80.31±4.68	57.43±5.75	13.22±2.16	87.71±7.78
<i>C. cyminum</i>	209±12.66	517±7.95	272±3.99	107.10±9.47	82.02±10.52	12.89±2.15	24.29±1.66
<i>D. viscosa</i>	147±40.52	199±4.59	131±4.58	124.70±12.86	68.47±14.62	12.02±6.13	52.16±7.75
<i>M. rotundifolia</i> L.	233±20.92	248±8.24	145±2.71	67.10±5.48	41.82±7.17	10.60±1.63	69.87±9.78
<i>N. sativa</i>	351±10.43	293±36.1	121±8.75	40.33±5.48	30.08±0.82	6.93±2.66	17.87±4.45
<i>R. officinalis</i> L.	80±20.62	329±14.79	256±5.84	76.47±1.5	25.06±5.66	5.81±1.33	41.66±3.25
<i>Z. officinale</i>	95±2.55	347±60.52	75±0.89	41.65±2.18	26.79±2.51	4.48±1.75	15.74±1.93

## 90 2 Discussion

91 The significant variations among herbal samples in the cell-wall components may be  
 92 due to some inherent anatomical or morphological differences related to cell-wall  
 93 rigidity (Wilson, 1994) and leaf/twig ratio in the samples used in the chemical analy-  
 94 sis. The majority of the herbal species considered in this study contained below 40%  
 95 NDF on DM basis and this qualifies them as good quality plants (Singh and Oosting,  
 96 1992).

97 The concentration of phenolic compounds in the collected herbals showed considera-  
 98 ble variation among species. The analysis of specific tannins gives an indication of  
 99 the presence of some anti-nutritive factors in the samples. Except for some few spe-  
 100 cies (*A. officinarum* and *A. absinthium*), the plants material investigated in this study  
 101 had low tannin contents, particularly in *Z. officinale* which would be of little signifi-  
 102 cance in their effects on digestion of nutrients by ruminants, consistently with result  
 103 pointed out in the literature (Frutos et al., 2002) with woody leguminous shrubs.

104 Several studies showed strong antimicrobial activity of certain plant extracts against  
 105 Gram- and Gram+ bacteria. Plants readily synthesize substances for their defense  
 106 against insects, herbivores, and microorganisms. The secondary plant metabolites  
 107 such as flavonoids and tannins have been found to reduce methane production and  
 108 meat' lipid oxidation and increase its alphanocopherol content (Yagoubi et al., 2018);  
 109 moreover tannins prevent bloat of the rumen and possess antihelminthic properties  
 110 (Woodward et al. 2001).

### 111 **3 Conclusions**

112 Based on the results above, it could be concluded that a large reserve of herbal species  
 113 in the local flora is available and could be potentially used for livestock feeding. The-  
 114 se herbs appear to be promising alternatives to antibiotics in altering rumen fermenta-  
 115 tion and reducing methane production in ruminants.

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