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Effect of Ginger Powder (*Zingiber officinale*) on Acid-Base Balance, Rumen and Blood Constituents in Healthy Egyptian Sheep

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ABSTRACT

Medicinal herbs have been used for several thousand years in traditional medicine and are known to be inexpensive, effective, readily available, and safe to use, with almost no side effects. As no previous research focused on the effect of ginger powder (*Zingiber officinale*) on acid-base balance in sheep, the goal of this study was studying the effects of ginger (*Zingiber officinale*) powder on acid-base balance, rumen, and blood constituents. Ten Egyptian ewes were given ginger powder 500mg/kg bwt orally in the morning before feeding for 5 days. Blood and rumen juice samples were collected in the morning on 0 (control), 3rd, and 5th day before feeding. Results generally showed a significant increase in rumen fluid pH, WBCS, lymphocytes, MCH, and MCHC. Significant decrease in total volatile fatty acids, serum total protein, and globulin were recorded. Ginger maintained acid-base balance, rumen protozoa activity, total protozoa count, rumen ammonia concentration, RBCs, PCV, Hemoglobin, neutrophils, albumin, BUN, Creatinine, GGT, and AST within normal range. Depending on changes in blood and rumen constituents we may suggest a recommendation for using ginger supplementation as 500mg/kg bwt orally for 3-5days as an immune stimulant and in the treatment of rumen acidosis and respiratory affections in sheep.

Key words: Sheep, Zingiber officinale, Acid-base balance, Rumen and Blood constituents.

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INTRODUCTION

Sheep are valuable livestock species due to their ability to turn forages into meat and milk that are essential sources of human dietary protein. Sheep are the most common animal (other than avian species) raised in Egypt for meat (FAOSTAT 2018). So, to benefit the human population, it is necessary to enhance and sustain the health and productivity of these animals. The history of medicinal herbs used to cure diseases and improve sheep's overall health with a focus on GIT diseases in small ruminants has been reviewed by (Engel 2007). Herbs and spices are considered to have health benefits such as stimulants for appetite and digestion, anti-microbial action, antiinflammatory action, anti-oxidative action, and immune stimulant activity on animals (Al-Azazi et al. 2018).

Ginger (*Zingiber officinale*) is one of most famous and useful herbals. Ginger belongs to *Zingiberaceae* family and *Zingiber* genus (Bhatt et al. 2013; Zadeh and Kor 2014; Bakr et al. 2020). Ginger contains more than 60 active constituents, including gingerols, shogaols, paradols, and even zingerone, and is a good source of essential micronutrients such as potassium, magnesium, copper, manganese, silicone, and small quantities of vitamins such as A, E and some quantities of B and vitamin C are also present in ginger rhizome (Bhatt et al. 2013). In recent years, several studies have focused on ginger's potential in rumen fermentation modification (El Samarany 2015; Soroor and Moeini 2015; Al-Azazi et al. 2018), but previous studies have not concentrated on ginger's impact on acid-base balance. This study was applied to investigate the effect of ginger powder (*Zingiber officinale*) 500mg/kg bwt orally supplement for 5 days on (acid-base balance), rumen (physical, cellular, and biochemical), and blood constituents in apparently healthy Egyptian sheep.

MATERIALS AND METHODS

Ethical Approval

The current study was approved by Veterinary Medicine Cairo University Institutional Animal Care and Use Committee.

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Animals and Experimental Design

A total of 10 apparently healthy non-pregnant Egyptian ewes, belonging to the Department of Medicine and Infectious Diseases, Faculty of Veterinary Medicine, Cairo University, have been used in this study. Their age ranged between 2-4 years and their body weight ranged between 30-40kg. They administered ginger powder as 500mg/kg bwt orally, dissolved in a sufficient quantity of water; in the morning before feeding the traditionally offered ration for 5 days. The dose 500mg/kg bwt was determined by Matthews et al. (2016) and Al-Azazi et al. (2018).

Samples

Blood and rumen juice samples were collected in the morning before feeding on 0(control), 3rd, and 5th day of experiment. Blood samples for acid base balance were collected by puncture of jugular vein on heparin plastic syringe. EDTA vacutainers used for the CBC analysis and plain vacutainers used for serum separation for biochemical analysis. Rumen juice samples (40ml) were collected using a rubber stomach tube, in dry clean cups. Samples were taken to the laboratory immediately after sampling.

Laboratory Examination

Blood samples taken by heparin plastic syringes for acid-base balance (pH, PCO₂, PO₂, HCO₃, and base excess) were analyzed by using the blood gas analyzer. Samples taken with EDTA vacutainers for CBC estimation were examined manually by using the method described by Wintrobe (1976), and samples taken with plain vacutainers were centrifuged at 4000rpm/10 minutes then blood serum separated into a clean dried plastic vial and stored at-18°C till chemical analysis. Serum samples examination includes estimation of serum total protein, albumin, Gamma-glutamyltransferase (GGT), Aspartate aminotransferase (AST), blood urea nitrogen (BUN) and serum creatinine, using APEL spectrophotometer - Japan and specific kits produced by Spectrum Company- Egypt. Globulin and (A/G) ratio was calculated mathematically. Physical examination of rumen samples includes (pH, odor, color, and consistency) according to Radostits et al. (2007) and microscopic examination includes protozoal activity according to Alonso (1979) were immediately examined. Then samples were sieved through 4 folds of sterile gauze, 2ml fixed with strong acids used to determine concentrations of volatile fatty acids by Macro Kgeldahl steam distillation method described by Eadie et al. (1967), 2ml used to determine ammonia N2 concentration by using APEL spectrophotometer- Japan and specific kits produced by Biodiagnostics Company-Egypt, and 2ml fixed and stained with methylene green formal saline for total protozoal count according to the method of Ito et al. (1994).

Statistical Analysis

Statistical analysis was carried out by SPSS program version 24, using one-way ANOVA. Results were expressed as mean \pm SD at P \leq 0.05.

RESULTS AND DISCUSSION

Acid base-balance parameters (Table 1) showed no significant difference in pH, PCO₂, PO₂, HCO_{3⁻} and base excess (P>0.05) on the 3^{rd} and 5^{th} day of the experiment. Findings of day zero were in agreement with what was recorded by Onmaz et al. (2009) and Hussein and Aamer (2013). This result indicates that ginger didn't affect acid-base balance in the body and maintain it within normal ranges.

Rumen physical, chemical, and microscopical constituents (Table 2) showed Yellowish-brown color, aromatic odor, slimy to slightly viscous consistency in agreement with the findings of Anderson and Rings (2008), Karapinar et al. (2008), and Constable et al. (2017). Non-significant (P>0.05) differences were noted along the experiment in color, odor and consistency in agreement with the findings of Al-Azazi et al. (2018). Ginger maintained protozoal activity and TPC in agreement with the findings of Al-Azazi et al. (2018) and Abo Bakr (2019). Regarding rumen pH value, there was a significant increase in pH (P<0.05) and the highest value was after the 3rd day, this in agreement with the findings of Taylor (2017) who reported that ginger is one of most alkaline herbs, and contrary to the findings of Zhang et al. (2011), El Samarany (2015) and Al-Azazi et al. (2018) which reported that ginger didn't change rumen pH, and (Al-Khayat 2011) which reported a decrease in rumen pH. The results of rumen biochemical constituents showed significant decrease in TVFAs (P<0.05) concentration on the 3rd day and increased again on the 5th day of treatment similar to the findings of Zhang et al. (2011). Thus, suggesting that rumen microbial fermentation was inhibited due to antimicrobial activity of ginger. Soroor and Moeini (2015) recorded no effect on TVFAS, while Al-Azazi et al. (2018) recorded slight increases in TVFAs concentration on the 2nd and 4th day following treatment. There was no significant difference in rumen ammonia N2 concentration (P>0.05) and Al-Azazi et al. (2018), while El Samarany 2015) and Soroor and Moeini (2015) reported a significant decrease in ammonia N2 concentration (P<0.05).

Complete blood count (Table 3) showed nonsignificant difference (P>0.05) occurred between different days of sampling in RBCs count, hemoglobin, PCV, neutrophils, and MCV along with the findings of Al-Dain and Jarjeis (2015), AL-Jubori (2017), and Al-Azazi (2019), while Hendawy et al. (2019) reported an increase in RBCs count. WBCs and Lymphocytes increased significantly (P<0.05) in agreement with the findings of Al-Dain and Jarjeis (2015) and Hendawy et al. (2019) and contrary to the finding by Al-Azazi (2019) who found non-significant changes occurred in WBCS. The significant increase in the total WBC may because ginger plays a good role in stimulating inflammatory cells and because ginger containing active ingredients such as gingerdiol and gingerols which have antioxidant activity (Zancan et al. 2002; Lin et al. 2003). The significant increase in the percentage of lymphocytes may be due to active compounds such as gingerols, it increases the body's defense mechanism, particularly the percentage of B-Lymphocytes cells (Al-Dain and Jarjeis 2015). MCHC and MCH showed a significant increase (P<0.05), while Hendawy et al. (2019) reported no-significant (P>0.05) differences in MCHC percentages.

Table 1: Effect of ginger supplementation on Acid-base balance parameters on different days.

0 day (control)	3 rd day	5 th day
	J	7.46±0.02a
41.60±1.56a	41.40±1.00a	42.00±1.55a
53.80±9.48a	47.00±1.52a	42.20±0.87a
29.00±1.17a	29.80±1.58a	29.00±0.75a
4.76±1.39a	5.22±1.94a	4.70±0.92a
-	53.80±9.48a 29.00±1.17a	$\begin{array}{ccccc} 7.46 \pm 0.02a & 7.46 \pm 0.03a \\ 41.60 \pm 1.56a & 41.40 \pm 1.00a \\ 53.80 \pm 9.48a & 47.00 \pm 1.52a \\ 29.00 \pm 1.17a & 29.80 \pm 1.58a \end{array}$

Values (mean±SD) within the same row having common letter are not significantly (P<0.05) different.

Table 2: Effect of ginger supplementation on physical, cellular and biochemical constituents of rumen fluid on different days.			
Variables	0 day (control)	3 rd day	5 th day
Protozoal activity	++ to +++	++ to +++	+++
TPC (×10 ⁴ /ml)	41.80±6.17a	54.20±8.01a	66.20±13.28a
pH	7.20±0.11b	7.82±0.09a	7.54±0.07a
TVFAS (mmol/L)	60.30±3.36a	37.04±2.58b	50.16±2.26a
Ammonia N2 (mmol/L)	7.70±0.56a	5.70±0.86a	7.13±1.01a

Values (mean±SD) within the same row having common letter are not significantly (P<0.05) different.

Table 3: Effect of ginger supplementation on complete blood count on different days.

Variables	0 day (control)	After (3) days	After (5) days
RBCs (×10 ⁶ /cmm)	11.65±0.57a	11.95±0.44a	12.05±0.43a
Hemoglobin (mg/dl)	10.62±0.37a	10.98±0.40a	11.56±0.22a
PCV (%)	36.20±1.43a	36.40±1.22a	37.20±1.45a
WBCs ($\times 10^{3}$ /cmm)	7.40±0.22b	9.26±0.33a	8.72±0.18a
Neutrophils (%)	23.80±0.33a	24.00±0.28a	23.20±1.34a
Lymphocytes (%)	18.00±1.13c	51.00±2.56a	39.40±2.07b
MCV (fl)	30.52±0.69a	28.58±0.17a	28.98±0.63a
MCH (pg)	8.98±0.41b	11.10±0.29a	11.32±0.15a
MCHC (mg/dl)	29.44±1.20b	33.18±0.23a	33.42±0.20a

Values (mean±SD) within the same row having common letter are not significantly (P<0.05) different.

Table 4: Effect of ginger supplementation on Serum biochemical constituents on different d	ays.
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Variables	0 day (control)	After (3) days	After (5) days
AST (units/ml)	66.20±1.28a	84.20±12.63a	104.40±14.61a
GGT (Units/ml)	27.20±2.69a	29.60±5.22a	41.60±7.39a
Total protein (g/dl)	7.10±0.13a	5.92±0.22b	5.84±0.37b
Albumin (g/dl)	2.87±0.07a	2.88±0.07a	2.90±0.04a
Globulin (g/dl)	4.23±0.07a	3.04±0.23b	2.94±0.39b
A/G	0.68±0.01a	0.98±0.10a	1.15±0.25a
BUN (mg/dl)	19.60±1.46a	17.20±0.95a	20.80±1.48a
Creatinine (mg/dl)	1.47±0.15a	1.64±0.07a	1.41±0.13a

Values (mean±SD) within the same row having common letter are not significantly (P<0.05) different.

Serum constituents (Table 4) showed significant decrease (P<0.05) in total serum protein and globulin on 3rd and 5th days compared to day zero of the experiment, the peak of decrease occurred after the 5th day, in agreement with the findings of Al-Homidan (2005), but Al-dain and Jarjeis (2015), Al-Azazi et al. (2018) and Abo Bakr (2019) reported significant increases (P<0.05) in total proteins and globulin after ginger powder supplementation. Serum albumin showed no significant difference (P>0.05) between experiment days, in agreement with the findings of Al-Azazi et al. (2018) and contrary to the findings reported by EL-Gohary et al. (2012) which reported an increase in serum albumin. Regarding serum A/G ratio, no significant difference (P>0.05) was noted in the A/G ratio compared to day zero, while Al-Azazi et al. (2018) reported significant decrease (P<0.05) on the 5th day in A/G ratio compared to day zero. Blood urea nitrogen had no significant difference between different sampling days because ruminal concentration of NH3-N was not affected by the addition of ginger in our study, so no changes in plasma urea-N concentration were expected (Petit and Flipot 1992; Davidson et al. 2003). These findings in agreement with the findings of Al-Azazi et al. (2018) and contrary to the findings of EL Gohary et al. (2012) which reported a significant increase (P<0.05) in BUN. Serum creatinine, AST and GGT showed no significant difference between the different sampling days. In this connection EL-Gohary et al. (2012), Al-dain and Jarjeis (2015) and Al-Azazi et al. (2018), while Abo Bakr (2019) found substantial increases in creatinine, AST and GGT. These results indicated that ginger didn't affect the organs function. The observed variation in all these parameters may be attributed to variations in species, doses, preparation, and factors affecting absorption from the gut.

Conclusion

Ginger powder (Zingiber officinale) 500mg/kg bwt orally giving in sheep for 5 days, maintained acid-base balance, increased rumen pH, WBCs, and lymphocytes. Concerning changes in blood and rumen constituents, we can recommend using ginger supplementation as 500mg/kg bwt orally for 3-5 days as an immune stimulant and in the treatment of rumen acidosis and respiratory affections in sheep. Further investigation should be applied on diseased cases to confirm the effect of ginger as a therapeutic agent in such cases.

Author's Contribution

Zaki MG, performed animal treatments, blood samples and rumen fluid collection, and data analysis. Baraka TA, designed the experiments and also wrote the manuscript. Tayeb FA, reviewed the manuscript. All authors reviewed and approved the final version.

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