

## Adding Natural Juice of Vegetables and Fruitage to Ruminant Diets: (A) Lemon, Onion and Garlic Juice Supplement to Diets Fed to Suckling Buffalo Calves and its Effect on Digestibility, Growth Performance and Fungi Count

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**Abstract:** Twenty four suckling buffalo calves average body weight  $38.78 \pm 0.95$ . Calves divided into four groups (6 calves, each). Group one ( $G_1$ ) was control while ( $G_2$ ), ( $G_3$ ) and ( $G_4$ ) were supplemented with 2.5%, 5%, and 7.5% natural juice / kg diet/day. The juice were consisted from lemon, onion and garlic (0.125 : 1 : 1) added for two litter clean water and squeeze in electric machine. Total counts of fungi and yeast were determined and aflatoxins estimation in the diet. The main results indicate that diets contain 5% and 7.50% natural juice showed significantly ( $P < 0.05$ ) better OM, CP, CF, EE and NFE digestibility coefficients than other diets. The nutritive value as TDN and DCP were increase significantly ( $P < 0.05$ ) with supplementetation of the natural juice. While the diet supplemented with 7.50 % natural juice showed increase significantly ( $P < 0.05$ ) for TDN and DCP as g/ kg w<sup>0.75</sup>. Daily weight gain of calves fed diets supplemented with 2.50 and 5.00 %, respectively natural juice were increase significantly ( $P < 0.05$ ) than other diets while the diet supplemented with 7.50 % natural juice insignificantly increase by 10.91% than the control one. Feed conversion expressed as kg DM, TDN and DCP/kg gain indicated that supplementetation of Lemon, Onion and Garlic juice to suckling buffalo calves diets improved DM conversion by 31.05, 36.64 and 21.75 %, respectively ; over the control, the same trend was observed with TDN and DCP conversion. However diet  $D_3$  tended to get better feed conversion with DM, TDN and DCP conversion. Diet supplemented with 7.5 % natural juice showed lower ( $P < 0.05$ ) pH value at 3 hrs. post feeding and higher  $NH_3-N$  and TVFA's concentration than the control diet. The highest number of isolated fungi was obtained from milk  $2 \times 10^{-4}$  cfu / g. Isolated fungi were identified as *Aspergillus niger*, *Uleolodium consortiale*, *Penicillium mautanense* and *Speregillus aculeatus*. The determined total counts of fungi in the water was  $77 \times 10^{-3}$  and  $7 \times 10^{-3}$  from calf starter. The three level of natural additive to milk, water and calf starter inhibited the mycelial growth of various fungi species. On the other hand, yeast count was increased in milk by increasing the concentrations of natural juice, while the high values of yeast was recorded with  $D_4$  ( $230 \times 10^{-4}$ ). The results of economic evaluation of the experimental diets showed decreasing in cost by 11.06 and 17.77 % of  $D_2$  and  $D_3$  than the control, while  $D_4$  showed increasing in cost by rate of 5.06 % than the control one.

**Key words:** Buffalo calves • lemon-onion and garlic juice • growth performance • digestibility • fungi count • aflatoxins

### INTRODUCTION

Dietary natural additive to calve diets have the ability to enhance and stabilize fermentation and improve viscosity texture, moisture and overall yields involved in meat production [1]. Adding different amounts of natural additive to whole milk and rations as described

by Sanz *et al.* [2] led to better utilization of the protein ingested in ruminants. Also, the observations of Quigley *et al.* [3] indicated that young calves given high additive (fat milk 40% on dry matter) improved their weight gain, feed efficiency and improvement of nutrients digestibility. Earlier research had led to the development of an early weaning program involving the use of a

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pre-starter and natural additive to stimulate feed consumption and destroy the harmful bacteria [3].

Before weaning, sources of energy protein and additive are derived mainly from the intestinal absorption of milk, while after weaning is derived primarily from ruminal fermentation and subsequent absorption [4, 5].

Garlic, onion and lemonade juice is highly inhibitory to molds and fungi [6]. Also, Wangenstein *et al.* [7] and Khir *et al.* [8] showed that, addition natural additive to food well increase the antioxidant content and may have potentials a natural antioxidant and thus inhibit unwanted oxidation processes. Gupta *et al.* [9] found that improvement in the digestibility coefficients of different nutrients is probably due to improved gross activity of rumen micro flora.

The objective of the present study was to evaluate of supplemented vegetable and fruitage juice as natural additive to diets of suckling buffalo calves on digestibility of nutrients, growth performance. Total count of fungi, yeast and aflatoxins were determine in milk, water and calf starter.

## MATERIALS AND METHODS

This work has been conducted at Mahillet Mossussa Experimental Station, Animal Production Research Institute (APRI) (Kafer El Shaikh Governorate). Chemical analysis were conducted at Dokki Laboratory of By-Product Utilization Department. Animal Production Research Institute. The fungi, yeast count and aflatoxins estimation were conducted at Regional Center for Food and Feed (RCFF), Agricultural Research Center, Ministry of Agriculture, Egypt.

Twenty-four indigenous suckling buffalo calves average body weight of  $38.78 \pm 0.95$  kg and one week of age. Calves were left with dams for three days nursing colostrum then divided randomly into four experimental groups after approximately similar weight and age. Four experimental diets were used from milk and calf starter. Calf starter consisted of 50 % yellow corn, 15 % barley, 25 % soybean meal, 8 % molasses, 0.50 % minerals mixture, 1.50 % limestone, 0.30 % antibiotic and 0.20 % vit. AD<sub>3</sub>E. The juice were consisted from lemon, onion and garlic (0.125 : 1 : 1) added for two litter clean water and squeeze in electric machine. The mixed juice were filtered and kept in dark glass bottle at 4°C. The 1<sup>st</sup> diet D<sub>1</sub> without supplementetation (control). The 2<sup>nd</sup> (D<sub>2</sub>) was supplemented with 2.5% mixed juice, 3<sup>rd</sup> (D<sub>3</sub>) was supplemented with 5% mixed juice and 4<sup>th</sup> (D<sub>4</sub>) was supplemented with 7.5 % mixed juice.

Feces and diets samples were daily collected and composite samples were prepared for chemical analysis according to conventional procedures of Aiad [10].

Calves were housed in four pens bedded with rice straw for the duration of the experiment. From the start of the experimental each calf suckled buffalo milk on 10 % of birth weight. Feeds were offered in two equal portions at 8.00 a.m and 4.00 p.m. as described by Salama and Mohy El-Deen [11] to over their nutritional requirements [12].

Berseem hay (BH) as a bulky roughage was offered *ad lib*. Fresh water and mineral mixture blocks were freely available to animals. The trial lasted 105 days and calves were biweekly weighed in the morning before offering any feed or water. Live body weight changes and feed intakes were recorded at biweekly intervals.

At the end of 8<sup>th</sup> week of the growth experiment, three calves were taken at random from each group to determine the digestibility and nutritive value of experimental diets. Acid insoluble ash (AIA) method was used as described by Van keulen and Young [13]. During feces grabbing period calves were fed chopped Berseem hay as 80 % from the average daily free choice intake. Feeding and rectum grabbed feces collection was practiced for 5 days. Feces samples were treated with 10 % sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) and kept frozen at -18°C for further chemical analysis.

Rumen fluid samples were collected from three animals from each group using a rubber stomach tube at 0, 3 and 6 hrs after morning feeding. Ruminal pH was directly estimated by pH meter and ammonia-N was determined according to Conway [14]. Total volatile fatty acids (VFA's) were measured according to Warner [15]. Molar proportions of VFA's were determined according to Erwin *et al.* [16].

**Determination of total count of fungi:** Total counts of fungi were determined on potato dextrose agar (PDA) medium [17] plates were incubated at 25°C for 7 days and the counts of fungi (cfu/g) were determined as described in American Public Health, Association [18] and Oxpoid Manual [19].

**Isolation and identification of fungi:** Developed colonies on PDA medium were transferred to PDA slants and purified using the single spore technique [20] and /or hyphol tip technique [21]. Purified isolates were identified according to their morphological and microscopically characters as described by Jenes *et al.* [22] and confirmed by Plant Pathology Institute ARC., Egypt.

**Mycotoxins analysis:** All standards of Mycotoxins were purchased from sigma company, USA. All chemicals and solvents used were of ACS grade. Thin layer chromatography (TLC) was performed using 20x20 cm TLC aluminum plates recoated with 0.25 mm silica gel 60 (Merk). aflatoxins, zearalenon and funonisn were extracted by B.F. method as described in A.O.A.C. [23]. Extracts were dissolved in soul chloroform and vortexed, 20µl aliquot and 10µl of the standards were stopped on TLC plates and developed in dark room with ethyl ether : methanol : water (96 : 3 : 1). After drying the spots were examined with U.V at a wave length of 365 nm [24].

**Yeasts count:** Total counts of yeast were determined on Ruse-bengl chloramphenicol agar according the methods described in Oxpoid Manual [19]. Plates were incubated at 22-25°C for 7 days.

**Diets and water pH values:** The value of pH was measured according to Ling [25]. pH values of samples were estimated by means of an electric pH-metr (Wissens chaftlich tehcnisch werkstatten D 8/20 weitheim pH 40).

An economical study of supplement the natural juice of lemon, onion and garlic the diets of suckling buffalo calves was under taken using the current feed purchase prices to find out the feed cost of producing one kilogram live bodyweight.

The chemical analysis of feeds and feces were carried out according to A.O.A.C. [23]. The collected data were statistically analysis according to Snedecor and Cochran [26]. The differences among means were tested using Duncan's multiple range test [27]. The following model was used:

$$Y_{ij} = \mu + Ti + e_{ij}$$

Where:  $Y_{ij}$  = observed trait,  $\mu$  = overall mean,  
 $Ti$  = effect of treatment,  $e_{ij}$  = random error.

## RESULTS AND DISCUSSION

**Chemical composition:** The chemical composition of feed ingredient and experimental diets are presented in Table 1. Data indicated that the experimental diets were iso- nitrogenous iso-caloric. The chemical composition of the BH and calf starter were within the normal published ranges for CP, CF and DE [28]. The chemical composition of buffalo milk were nearly similar reported by EL-Ashry *et al.* [29].

**DM intake, Digestibility and Nutritive Values:** Nutrient consumed during digestibility evaluation trial were calculated according to feeds consumed and their chemical composition. The composed diets showed comparable nutrients as shown in Table 1.

Nutrient digestibility of these diets are presented in Table 2. The diets contain 5% and 7.50% natural juice of lemon, onion and garlic supplementation showed significantly ( $P < 0.05$ ) better OM, CP, CF, EE and NFE digestibility coefficients than those control and diet contain 2.50 % experimental natural juice. The present results agree with those of Khir and Ibrahim [8], Moawd *et al.* [31] and Zaki *et al.* [32]. The nutritive value as TDN for the four diets showed comparable results and ranged between 65.53 and 71.26 %. the low digestibility of CP (67.67 %) for the control diet tended to significantly ( $P < 0.05$ ) the lowest digestible crude protein (DCP), being 13.80% while those of D2, D3 and D4 were 14.07, 14.51 and 14.55 % respectively. The total DM intake kg /head /day appeared to higher with animals fed diet (D4) containing 7.5% natural juice of lemon, onion and garlic as supplementation than those of the others. Consequently, the TDN intake was higher with animals fed same diet when expressed as kg /100kg BW or g/kg  $W^{0.75}$  being 1.337 and 38.79 respectively. These results were agreement with those obtained by Khir and Ibrahim [8] and El-Ashry *et al.* [29] and Zaki *et al.* [32].

Table1: Chemical composition of the experimental diets, buffalo milk, berseem hay and calf starter

Items	D1	D2	D3	D4	Milk	BH	Calf starter
Moisture	8.86	8.64	8.55	7.75	84.90	9.71	11.07
<b>DM composition %:</b>							
CP	20.39	20.49	20.59	20.51	25.52	12.30	18.60
CF	7.90	7.88	7.82	7.95	-	28.50	3.25
EE	3.51	3.53	4.00	4.60	43.17	2.70	2.93
NFE	59.34	59.61	59.04	58.19	25.41	44.00	68.68
OM	91.14	91.51	91.45	91.25	94.10	87.50	93.46
Ash	8.86	8.49	8.55	8.75	5.90	12.50	6.54
DEkcal/kg*	2.69	2.94	2.69	2.69	2.94	2.03	2.83

\*DE= 4.36-0.049 x NDF

NDF = 28.924 + 0.657 (CF%) according to Cheeke [30].

Table 2 : Effect of experimental diets fed to suckling calves during feces grabbing period on daily dry matter intake, nutrient digestibility and nutritive values

Item	Experimental Diets				SE
	D <sub>1</sub>	D <sub>2</sub>	D <sub>3</sub>	D <sub>4</sub>	
Animal weight, kg	69.92	72.82	74.25	70.75	±1.90
Body weight, kg W <sup>0.75</sup>	24.18	24.93	25.29	24.39	±0.25
DMI intake, kg /head /day :					
Milk	0.515	0.504	0.492	0.510	±0.48
Calf starter	0.612 <sup>b</sup>	0.541 <sup>a</sup>	0.585 <sup>a</sup>	0.621 <sup>b</sup>	±0.39
Brseem hay	0.229	0.224	0.219	0.227	±0.42
Total DMI kg /head /day	1.356	1.269	1.296	1.358	±0.26
Nutrient digestibility %:					
DM	64.22 <sup>b</sup>	66.17 <sup>b</sup>	68.45 <sup>a</sup>	66.85 <sup>a</sup>	±0.61
OM	66.83 <sup>b</sup>	67.80 <sup>b</sup>	71.36 <sup>a</sup>	68.37 <sup>a</sup>	±0.68
CP	67.67 <sup>b</sup>	68.67 <sup>b</sup>	70.50 <sup>a</sup>	70.94 <sup>a</sup>	±0.39
CF	51.82 <sup>b</sup>	52.24 <sup>b</sup>	56.64 <sup>a</sup>	56.46 <sup>a</sup>	±1.42
EE	65.23 <sup>c</sup>	66.27 <sup>c</sup>	75.19 <sup>a</sup>	73.29 <sup>b</sup>	±1.26
NFE	71.60 <sup>c</sup>	72.23 <sup>c</sup>	77.16 <sup>a</sup>	74.06 <sup>b</sup>	±2.21
Nutritive value:					
TDN	65.53 <sup>b</sup>	70.10 <sup>b</sup>	71.26 <sup>b</sup>	69.70 <sup>a</sup>	±0.25
DCP	13.80 <sup>b</sup>	14.07 <sup>b</sup>	14.51 <sup>a</sup>	14.55 <sup>a</sup>	±0.22
TDN intake:					
Kg DM /head / day	0.8886 <sup>b</sup>	0.8896 <sup>b</sup>	0.9235 <sup>a</sup>	0.9465 <sup>a</sup>	±0.03
Kg / 100 kg body weight	1.2709 <sup>b</sup>	1.2190 <sup>b</sup>	1.2430 <sup>b</sup>	1.3370 <sup>a</sup>	±0.30
g / kg W <sup>0.75</sup>	36.7500 <sup>b</sup>	35.8800 <sup>b</sup>	36.5200 <sup>a</sup>	38.8100 <sup>a</sup>	±1.48
DCP intake:					
Kg DM /head / day	0.1871 <sup>b</sup>	0.1785 <sup>b</sup>	0.1881 <sup>b</sup>	0.1976 <sup>a</sup>	±0.02
Kg / 100 kg body weight	0.2676 <sup>b</sup>	0.2452 <sup>b</sup>	0.2533 <sup>b</sup>	0.2793 <sup>a</sup>	±0.42
g / kg W <sup>0.75</sup>	7.3800 <sup>b</sup>	7.1600 <sup>b</sup>	7.4300 <sup>a</sup>	8.1000 <sup>a</sup>	±0.05

<sup>a,b,c</sup> Means in the some row having different superscripts are significantly different at (p< 0.05)

Table 3: Growth parameter of suckling calves fed the experimental diets

Item	Experimental Diets				SE
	D <sub>1</sub>	D <sub>2</sub>	D <sub>3</sub>	D <sub>4</sub>	
Duration period / days	105	105	105	105	
Number of suckling calves	6	6	6	6	
Average body weight (kg):					
Initial body weight, kg	39.67 <sup>a</sup>	38.82 <sup>a</sup>	37.88 <sup>a</sup>	39.30 <sup>a</sup>	±2.11
Final body weight, kg	88.86 <sup>a</sup>	94.90 <sup>b</sup>	98.34 <sup>b</sup>	91.08 <sup>c</sup>	±3.20
Total weight gain, kg	49.19 <sup>a</sup>	56.08 <sup>bc</sup>	60.46 <sup>b</sup>	51.78 <sup>cc</sup>	±0.29
Av. Daily Gain (ADG), kg	0.469 <sup>a</sup>	0.534 <sup>bc</sup>	0.576 <sup>b</sup>	0.493 <sup>cc</sup>	±0.15
DMI intake, kg /head /day :					
Buffalo Milk	0.464 <sup>a</sup>	0.454 <sup>a</sup>	0.443 <sup>a</sup>	0.460 <sup>a</sup>	±0.28
Calf starter	0.651 <sup>a</sup>	0.693 <sup>ab</sup>	0.696 <sup>ab</sup>	0.732 <sup>b</sup>	±0.38
Berseem hay	0.297 <sup>a</sup>	0.332 <sup>ab</sup>	0.332 <sup>ab</sup>	0.347 <sup>b</sup>	±0.18
Total DM intake	1.594 <sup>a</sup>	1.469 <sup>b</sup>	1.461 <sup>b</sup>	1.539 <sup>a</sup>	±0.83
Feed conversion:					
Kg DMI/kg gain	3.990 <sup>a</sup>	2.751 <sup>bc</sup>	2.536 <sup>b</sup>	3.122 <sup>c</sup>	±1.11
Kg TDN/kg gain	2.227 <sup>a</sup>	1.928 <sup>bc</sup>	1.807 <sup>c</sup>	2.176 <sup>bc</sup>	±1.11
Kg DCP/kg gain	0.469 <sup>a</sup>	0.387 <sup>b</sup>	0.368 <sup>b</sup>	0.454 <sup>a</sup>	±0.23

<sup>a,b,c</sup> Means in the some row having different superscripts are significantly different at (p< 0.05)

Table 4: Rumen fluid parameter of suckling calves fed the experimental diets

Item	Time (hrs)	Experimental Diets				SE
		D <sub>1</sub>	D <sub>2</sub>	D <sub>3</sub>	D <sub>4</sub>	
pH	0	6.05 <sup>a</sup>	5.83 <sup>b</sup>	5.65 <sup>b</sup>	5.93 <sup>a</sup>	0.06
	3	5.69 <sup>a</sup>	5.64 <sup>a</sup>	5.58 <sup>a</sup>	5.44 <sup>b</sup>	0.07
	6	5.82 <sup>a</sup>	5.71 <sup>a</sup>	5.69 <sup>a</sup>	5.56 <sup>b</sup>	0.05
NH <sub>3</sub> -N (mg/dl)	0	13.96 <sup>a</sup>	14.12 <sup>a</sup>	13.64 <sup>a</sup>	13.81 <sup>a</sup>	1.10
	3	18.20 <sup>a</sup>	18.62 <sup>a</sup>	18.72 <sup>a</sup>	18.75 <sup>a</sup>	0.20
	6	14.21 <sup>a</sup>	14.35 <sup>a</sup>	14.40 <sup>a</sup>	14.60 <sup>a</sup>	0.49
VFA's (meq/dl)	0	7.45	7.51	7.71	7.85	0.75
	3	9.46	10.58	10.79	10.83	0.65
	6	8.35	5.53	8.82	8.85	0.12
Acetate %	0	48.2	48.00	49.70	49.80	0.43
	3	45.3	45.20	47.10	46.35	0.57
	6	45.3	45.75	46.35	46.85	0.40
Propionate %	0	39.35	41.25	38.90	40.90	0.53
	3	40.45	41.30	39.80	40.85	0.72
	6	40.85	41.55	39.95	39.35	0.57
Butyrate %	0	12.45	10.15	11.40	9.30	0.04
	3	14.25	13.50	13.10	12.80	0.05
	6	13.50	12.10	13.70	13.20	0.04

<sup>a, b, c</sup> Means in the some row having different superscripts are significantly different at (P<0.05)

**Growth performance:** The average daily body weight gain values were 0.469, 0.534, 0.576 and 0.493 kg for D<sub>1</sub>, D<sub>2</sub>, D<sub>3</sub> and D<sub>4</sub>, respectively as presented in Table 3. Daily weight gain of calves fed diets supplemented with 2.50 and 5.00 % natural juice were increase significantly (P<0.05) than other diets while no significant differences were found with group fed diet supplemented with 7.5% natural juice. These results indicated that the use of lemon, onion and garlic juice as feed supplement to suckling calves diets had no adverse effect on average daily gain. Results obtained in this study are in line of the finding of Khir and Ibrahim [8], Salama and Mohy El-Deen [11], El-Ashry *et al.* [29], Mowad [31], Zaki *et al.* [32] and Roy *et al.* [33].

Feed conversion expressed as kg DM, TDN and DCP/kg gain indicated that supplementetation of Lemon, Onion and Garlic juice to suckling buffalo calves diets improved DM conversion by 31.05, 36.64 and 21.75%, respectively; over the control, the same trend was observed with TDN and DCP conversion. However diet D<sub>3</sub> tended to get better feed conversion with DM, TDN and DCP conversion. These results agree with those of Khir and Ibrahim [8] EL-Ashry *et al.* [29], Mowad [31], Zaki *et al.* [32], Roy *et al.* [33], Parasd *et al.* [34], Ali *et al.* [35] and Safaa Nadi [36].

**Ruminal parameters:** Results of pH values; ammonia nitrogen concentrations (NH<sub>3</sub>-N); volatile fatty acids

concentrations are (VFA's) presented in Table 4. Diet supplemented with 7.5% juice D<sub>4</sub> showed lower (P<0.05) pH value at 3 hrs post feeding and higher NH<sub>3</sub>-N and TVFA's concentration than the control diet. Increasing TVFA's with increasing juice level in diet may be related to better fermentation of the soluble carbohydrate within 3.00 hrs post feeding. These results of rumen parameters were within the normal range of suckling calves. The results of molar proportion (%) as acetate; propionate and butyrate were not affected with supplementation of natural juice of lemon, onion and garlic compared with the control diet. These results are in agreement with those found by White *et al.* [37], Williams [38], Dawson *et al.* [39] and Robinson and McQueen [40].

**Bacteriological results:** The occurrence of isolated fungi strains varied from milk, water and calf starter are shown in Tables 5 & 6. The highest number of isolated fungi was obtained from milk 2x10<sup>-4</sup> cfu / g. isolated fungi were identified as *Aspergillus niger*, *Uleolodum consortiale*, *Penicillium mautanense* and *Speregillus aculeatus* from milk. The determined total counts of fungi in the water was 11x10<sup>-3</sup> and 7x10<sup>-3</sup> from calf starter.

From the above general the most common species were *Aspergillus niger*, *Penicillium* spp and *Fusarium* spp. These findings are almost in agreement with those reported by Gherbawy [41] during study on the atmosphere at Hurghada city. In the present study all

Table 5 : Effect of natural juice supplementation to experimental diets on total fungal and yeast counts and pH values of milk, water and calf starter.

Test samples	Supplementation %	fungi Count cfu/g	yeast Count cfu/g	pH
Milk	Without	$2 \times 10^{-4}$	$45 \times 10^{-3}$	7.02
	2.5	$1 \times 10^{-3}$	$120 \times 10^{-3}$	4.44
	5	nil	$120 \times 10^{-3}$	3.96
	7.5	nil	$230 \times 10^{-4}$	3.83
Water	Without	$11 \times 10^{-3}$	$35 \times 10^{-3}$	6.96
	2.5	$8 \times 10^{-2}$	nil	3.33
	5	nil	nil	3.19
	7.5	nil	nil	3.50
Calf starter	Without	$7 \times 10^{-3}$	nil	5.12
	2.5	$4 \times 10^{-2}$	nil	4.52
	5	$1 \times 10^{-2}$	nil	4.20
	7.5	nil	nil	4.18

Table 6: Isolated fungi from milk, water and calf starter

Test samples	Isolated Fungi	Total count before supplementation
Milk	<i>Aspergillus niger</i>	$2 \times 10^{-4}$
	<i>Uicoladium consortiale</i>	
	<i>Penicillium montaneuse</i>	
	<i>Spergillus aculeatus</i>	
Water	<i>Apserigillus spp.</i>	$11 \times 10^{-3}$
	<i>Penicillium spp.</i>	
	<i>Apserigillus niger</i>	
	<i>Apserigillus versicolor</i>	
	<i>Apserigillus carneus</i>	
Calf starter	<i>Apserigillus niger</i>	$7 \times 10^{-3}$
	<i>Aspergillus aculeatus</i>	
	<i>Aspergillus syndowii</i>	
	<i>Aspergillus tamari</i>	
	<i>Fusarium emitectium</i>	
	<i>Fusarium pallidoro serum</i>	
	<i>Aspergillus falvus</i>	

Table 7: Economic evaluation of the experimental diets for growth performance

Item	Experimental diets			
	D1	D2	D3	D4
Feed cost / kg gain , LE				
Milk <sup>1</sup>	13.12	11.26	10.19	12.35
Starter <sup>2</sup>	2.02	1.88	1.75	2.15
BH <sup>3</sup>	0.63	0.62	0.56	0.70
Mixed Juice <sup>4</sup>	0.00	0.44	0.89	1.41
Total cost, LE / kg gain	15.77	14.20	13.39	16.61
Relative economic efficiency	100.00	111.06	117.77	94.94

1 - on the basis of a price of milk 2.00 LE / litter

2 - on the basis of a price of 1.45 LE kg

3 - on the basis of a price of Berseem hay 1.00 LE / kg

4 - on the basis of a price of lemon 50.0, onion 20.0 and garlic 20.0 PT/ kg

5 - Assuming that cost p of kg gain of the control equals 100.

tested fungi were sensitive to natural additive specially garlic and onion extracts. The three level of natural additive to milk, water and calf starter inhibited the mycelial growth of various fungi species. Aflatoxins not extract when the fungi not growth. The mycelial growth of *Aspergillus niger*, *Penicillium mantanese*, *Aspergillus falvus* and *Fusarium semitectium* and *Fusarium pallidoro serum* was significantly ( $P < 0.05$ ) retarded by medium and high doses.

On the other hand, yeast was increased in milk by increasing the concentrations of juice while the high values of yeast was recorded at 7.5 % supplemented juice concentration  $230 \times 10^{-4}$ . The present results are agree with Brufau [42] when use of essential oils in animal nutrition, yeast as on example of the mode of action of probiotics in mono-gastric and ruminant species, nutritional effects of organic acids acidification and other physiological additives, interactions of feed enzymes.

Natural additive cause pH reduction, organic acid profiles and fungi counts were determined. In juice added to calves diets citric acid and acetic acid were produced and the pH dropped to a value 4.0 and fungi products could not be detected in various materials tested.

After supplementation the juice to the water, milk and calf starter the of aflatoxins was nil in spite the fungi found in diets.

The results showed that the produced citric and acetic acid and the pH in the feed are responsible for fungi reduction in the ruminant feed. The present finding was agreement with Parasd *et al.* [34] and Gherbawy [41], they observed that the topical application of the crude extract of garlic at 1:0 concentration in distilled water could compact dermaphytosis produced in rabbits with Microsporin canis without causing any apparent side. Shekhawat and Proada [43] reported that boiled water extracts of onion caused inhibition to the growth of fungi. More recently Zohari *et al.* [44] noticed that onion

oil (200 ppm) completely inhibited the growth of *Asperigullus niger sp.* and *pencillium spp.*

Helen *et al.* [45] showed that the activity of glutathione reductase was decreased significantly ( $P<0.05$ ) in all almost tissues studied from treated rats. With onion oil or garlic oil treatment, glutathione peroxidase activity was increased significantly ( $P<0.05$ ) in almost all the tissues. In the present finding the antioxidant effect of this sulfur rich oils from *Allium* species is reflected by demonstrating an increase correlation between antioxidant enzyme and lipid peroxidation products in the milk, water and diet of suckling buffalo calves. These results suggest that garlic and onion (Natural additive) are useful antioxidants and their supplementation nullifies ill-effects caused by fungi and bacterial by affecting lipid peroxidation and the antioxidant defense system.

**Economic evaluation:** The economic evaluation of the experimental diets for suckling buffalo calves is presented in Table 7. Results showed that daily feed cost (L.E) was reduced in supplementation of garlic, onion and lemon diets with rate of 2.50 % (14.20 L.E) and 5% (13.39 L.E) compared to the control group (15.77 L.E). While the diet supplemented with 7.5 % juice show increase of daily feed cost (16.61 L.E) than the control. the high economic efficiency was recorded for the calves fed diet (D3) supplemented with 5 % juice. The economic efficiency was improved by the diet supplemented with 5 % juice of garlic, onion and lemon compared the control. This is in full agreement with the results obtained by Khir and Ibrahim [8], Salama *et al.* [11] and EL-Ashry *et al.* [29].

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