

## MILK YIELD AND COMPOSITION IN RESPONSE TO YEAST SUPPLEMENTATION TO LACTATING BUFFALOES DIETS.

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### ABSTRACT

Yeast culture additive product (Alpha Yea Mix ®) was offered to 4 multiparous lactating buffaloes through 4x4 Latin Square design. Buffaloes with 591± Kg average weight, ± Kg average milk yield, 70 DIM and 3-4 lactation seasons. The study aimed to investigate the effect of yeast culture supplementation on nutrients digestion, milk yield and constituents of buffaloes. Each experimental period lasted for 30 days. By the end of each period fecal grab samples for successive three days and milk samples for successive five days were collected and analyzed. Results obtained indicated that DM, OM, NFE, ADL and hemicellulose digestibilities were not changed among different supplemented diets. Crude protein (CP) and ether extract (EE) digestibilities tended to be insignificantly higher for buffaloes fed 30 and 40 g/h/d of yeast culture. Crude fiber (CF) and its fractions digestibility, including NDF, ADF and cellulose were significantly increased (10 - 22 %) in buffaloes fed 30 and 40 g/h/d of yeast comparing with those fed the control and 20g yeast. Diet feeding value expressed as TDN and DCP were insignificantly increased with buffaloes fed the higher levels of yeast supplementation. Actual milk yield, Kg/g was insignificantly increased with buffaloes fed 30 and 40 g/d of yeast culture. Milk constituents including fat percent and yield, protein percent and yield, lactose, urea, ash, TS, SNF percentages and somatic cell count (SCC) were not affected as a result of yeast culture supplementation to lactating buffalo's diets. Milk constituents tended to be higher with buffaloes fed 20g/d yeast supplemented diet vs. the other diets. The basic economic measures explicit to cost / benefit analysis indicated that the milk net return over feeding cost was proportionally higher as much as yeast culture supplementation increased in the diet. The implication of the present study is to add yeast culture products which mainly contain *Saccharomyces cerevisiae* ( $2 \times 10^9$ )/g at level of 30 – 40 g/h/d to improve milk yield and consequently the net revenue.

**Keywords:** Yeast, supplementation, buffaloes, fiber digestibility, and milk yield

### INTRODUCTION

Feeding ruminants exogenous enzymes was previously an unacceptable practice because these proteins were thought to be degraded by ruminal proteases (Kopečný *et al.*, 1987). However, Fontes *et al.* (1995) reported that some xylanases were extremely stable in ruminal fluid. Treating feeds with fibrolytic enzymes just before feeding has improved animal performance (Schingoethe *et al.*, 1999; Beauchemin *et al.*, 2000). Rode *et al.* (1999) reported that treatment of a barley-based concentrate with fibrolytic enzymes resulted in marked improvements in organic matter (OM) and fiber digestion of a total mixed ration (TMR) fed to lactating cows. Cows fed the diet supplemented with enzymes produced 10% more milk than cows fed the untreated TMR. Yeast cultures such as *Saccharomyces cerevisiae* are widely used in diets for lactating dairy cows. *Saccharomyces cerevisiae* is usually marketed as a yeast culture and may contain both viable yeast cells and a

dried preparation of the medium in which the cells were grown. Results of numerous studies with yeast supplementation to diets of lactating cows have been shown to be variable and inconsistent (Kilmer, 1993). Various studies indicated effects of yeast on the rumen environment (Henics and Gombos, 1992, Martin and Nisbet, 1992 and Williams *et al.*, 1991), rumen microbial fermentation (Dawson, 1990, Erasmus *et al.*, 1992, Nisbet and Martin, 1991 and Offer, 1990), rumen microbial populations (Dawson 1990, Harrison 1988, Weidmair *et al.*, 1987), ration digestibility specially for NDF and ADF (Kim *et al.*, 1992a, Kim *et al.*, 1992b and Bowman *et al.*, 2002 ), and DMI (Williams 1989 and Wohlt *et al.*, 1991). The effect on milk production parameters were found to be variable (Kilmer, 1993). Improvements in milk production (Williams *et al.*, 1991, Wohlt *et al.*, 1991 and Schingoethe *et al.*, 2004), milk fat percentage (Harri *et al.*, 1992), and milk protein percentage (Harris and Webb, 1990) have been reported in some studies. However, other studies (Erdma and Sharma, 1989, Henics and Gombos, 1992 and Kim *et al.*, 1992) showed no significant response to yeast supplements. The cause of the variability in response is unknown and may be related to the yeast strains and enzyme type (Schingoethe *et al.*, 1999 ), the amounts fed and various dietary factors (Bowman *et al.*, 2002 and Cook *et al.*, 2007).

The objective of this trial was to investigate the effects of daily supplementation of yeast culture preparation (Alpha Yea Mix) on milk production, milk composition, and nutrients digestibility with special reference to fiber of lactating Egyptian buffaloes.

## **MATERIALS AND METHODS**

The present study was carried out through the winter, 2007 at buffaloes farm, Agricultural Research Station, Faculty of Agriculture, Cairo University.

### **Animals, Diets and treatments**

Four multiparous mid lactating buffaloes with average body weight of  $591 \pm 75.0$  Kg, aged 4-6 years old, post-partum 70 days in milk and  $10.2 \pm 0.75$  Kg average milk yield were used in 4x4 Latin Square. Each experimental period was lasted for 30 days. All animals were fed on the regular feed regimen of the farm and requirements were adjusted according to Ghoneem 1967. Dietary treatments were (1) control, consisted of CFM (15% corticated cotton seed meal, 20% wheat bran, 53% yellow corn, 10% soybean meal, 1% limestone, and 1% sodium chloride salt), berseem and rice straw; (2) control + 20g yeast culture (Alpha Yea Mix<sup>®</sup>), consisted of 900 g dry *Saccharomyces cerevisiae* yeast with total number of  $(2 \times 10^9)$ /g and 100 g Alpha anti caking. The product produced and dispatched by Alpha Chemical Egypt and registered at the Ministry of Agriculture, No. 3500; (3) control + 30g yeast culture, and (4) control + 40g yeast culture. The dietary allowance was administered twice daily (7:00 a.m. and 4:00 p.m.). All animals were freely accessed fresh water. The yeast culture of *Saccharomyces cerevisiae* was mixed bi weekly with the concentrate feed mixture (CFM).

### **Sampling and analytical procedures**

By the end of each experimental period, fecal grab samples were withdrawn at 9: 00 a.m. and 4:00 p.m. for three successive days from each animal. A representative sub sample of total collected feces of each individual was taken and preserved with 10% formaldehyde and 10% sulfuric acid. Proximate analysis of feeds and feces samples including DM, ash, CP, CF, EE and NFE was determined according to A.O.A.C. (1995). Acid Insoluble Ash (AIA) as natural internal marker was applied for digestibility determination (Gallup *et al.*, 1945 and Forbes and Garrigus 1948). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and ADL were determined in feeds and feces, according to Goering and Van Soest (1970). Hemicellulose was calculated as the difference between NDF and ADF, while cellulose was calculated as the difference between ADF and ADL. Lactating buffaloes were hand-milked twice daily and data were recorded during the entire experimental period for yield. During the last four days of each period, about 5% representative milk sample was collected from each animal at evening and morning milking, respectively. Composite daily milk sample (relative to the quantity of milk produced) were taken for analysis of total solids (TS), fat, total protein (TP), lactose, solids not fat (SNF) and urea using infrared Milkoscan (B. Foss Electric, France), and ash according to Ling, 1963.

### **Statistical analysis**

The data of milk yield, milk constituents, dry matter intake (DMI), nutrients digestibility and feeding values were analyzed according to MSTATC (Nissen, 1989) using Latin Square design where the model was:  $Y_{ijk} = U + T_i + P_j + A_k + E_{ijk}$

Where, Y: stands for every observation of the K<sup>th</sup> animal in the J<sup>th</sup> period given I<sup>th</sup> treatment, T: treatment effect, P: periods effect, A: animal effect and E: the experimental error. Duncan's multiple range test (Duncan, 1955) was run out for means separation.

### **Economic Study**

A cost / benefit analysis and other simple economic measures were computed to indicate for the feasibility of supplementing yeast cultures as external cellulase and xylanase into lactating buffalo's diet.

## **RESULTS AND DISCUSSION**

### **Chemical composition of ingredients and experimental diets**

Chemical composition of ingredients and dietary treatments are shown in Table 1 and 2. There were no wide variations among the different diets ingredients, amounts, and chemical composition, except the yeast supplement (Table 2). Fiber fractions involving NDF, ADF and cellulose concentrations were proportionally linear with crude fiber content of the concentrate feed mixture, berseem and rice straw (Table 1).

The proximate analysis of the different diets represented a good balanced diets for lactating buffaloes. The acid insoluble ash content was

about 2% and represented up to 20% of the complete diet ash content. The acid insoluble ash is implicit to the silica content.

**Table (1): Chemical analysis of dietary ingredients (DM basis).**

Items	Diet ingredients		
	CFM	B	RS
Dry matter	90.28	19.79	94.30
Organic matter	88.53	86.39	83.48
Ash	11.47	13.61	15.11
Crude protein	16.37	18.13	3.78
Crude fiber	6.96	22.97	38.50
Ether extract	4.20	3.56	2.18
Nitrogen-free-extract	61.00	41.73	39.02
<b>Fiber fraction</b>			
NDF	31.5	56.00	70.35
ADF	9.01	41.80	53.80
ADL	2.5	9.35	5.36
Hemicellulose	22.49	14.20	16.55
Cellulose	6.51	32.45	48.44

CFM: Concentrate feed mixture B: Berseem RS: Rice straw

**Table (2): Dietary treatments and chemical composition (DM basis).**

Ingredients, Kg	Enzyme supplementation, g/h/d			
	Control (0)	20	30	40
<b>Diet Ingredients, Kg/h/d:</b>				
Yellow corn	6.46	6.52	6.52	6.31
SBM, 44%	1.22	1.23	1.23	1.19
CSM	1.83	1.85	1.85	1.79
Wheat bran	2.44	2.46	2.46	2.38
Limestone	0.08	0.08	0.08	0.08
Mineral mix	0.08	0.08	0.08	0.08
Berseem	25	27	25	25
Rice straw	1.4	1.9	1.85	1.4
Enzyme supp., g	0	20	30	40
<b>Diet proximate analysis, % (DM basis):</b>				
DM	36.50	36.40	37.30	36.20
OM	89.51	89.28	89.37	89.51
Ash	10.49	10.72	10.63	10.49
EE	3.78	3.72	3.72	3.78
CP	15.92	15.51	15.49	15.91
NFE	54.31	53.66	54.01	54.32
CF	15.50	16.39	16.15	15.50
AIA	2.10	2.14	2.13	2.10
<b>Fiber fractions:</b>				
NDF	43.73	44.79	44.44	43.72
ADF	24.73	25.92	25.46	24.72
ADL	5.20	5.24	5.15	5.19
Hemicellulose	19.00	18.87	18.98	19.00
Cellulose	19.53	20.68	20.31	19.53

AIA: Acid insoluble ash

**Nutrients digestibilities and feeding values:**

Digestibilities of different diets nutrients including DM,OM,EE,CP,NFE, CF, and Fiber fractions are presented in Table 3. It could be seen that DM, OM, NFE and hemicellulose digestibilities showed insignificant comparable values. Similar results have been reported by (Mwenya *et al.*, 2005). Increasing the yeast supplement slightly increased EE, CP and ADL digestibilities. Adding *Saccharomyces cerevisiae* yeast at levels of 30 and 40 g/h/d significantly increased digestibility coefficients of CF (10-13%) and both NDF, ADF and cellulose (16-22%) compared to the control and 20 g yeast supplement diets, which which is in agreement with Erasmus *et al.*, 1992 when dairy cows fed on yeast culture supplemented diet. Eun *et al.* (2007) reported that exogenous enzymes potentially improving the cell wall degradation of rice straw and significantly increased degradabilities of DM, NDF and ADF at 24 h incubation. Addition of exogenous fibrolytic enzymes (fibrozyme) to growing lambs ration increased crude fiber, hemicellulose and lignin digestibility (Abdel Gawad *et al.*, 2007) and NDF digestibility (Lewis *et al.*, 1996). Beauchemin *et al.* 1999 indicated that the effects of enzyme supplementation were mostly intestinal, showing more total tract digestibility of starch (5%), NDF (8%), and ADF (11%) than rumen digestibility.

**Table (3): Nutrients digestibility and feeding value of the experimental diets.**

Item	Enzyme supplementation, g/h/d				± SE
	Control (0)	20	30	40	
<b>Digestibilities, %:</b>					
DM	52.29	49.80	51.99	52.59	1.10
OM	56.07	53.37	55.80	56.41	1.20
EE	62.02	60.54	61.52	64.14	1.32
CP	60.33	61.36	63.08	63.79	1.37
NFE	67.23	65.05	66.47	66.65	0.80
CF	49.71 <sup>b</sup>	49.43 <sup>b</sup>	54.80 <sup>a</sup>	56.33 <sup>a</sup>	3.05
NDF	44.74 <sup>b</sup>	45.48 <sup>b</sup>	52.06 <sup>a</sup>	54.64 <sup>a</sup>	4.23
ADF	33.55 <sup>b</sup>	34.11 <sup>b</sup>	39.04 <sup>a</sup>	40.98 <sup>a</sup>	3.17
ADL	22.37	22.24	24.66	25.35	1.37
Hemicellulose	57.15	55.30	56.50	56.65	0.68
Cellulose	35.79 <sup>b</sup>	36.38 <sup>b</sup>	41.65 <sup>a</sup>	43.71 <sup>a</sup>	3.38
<b>Feeding Value, %:</b>					
TDN	59.21	57.75	59.84	60.63	1.22
DCP	9.61	9.51	9.77	10.15	0.28

<sup>a, b, ...</sup> Means in the same row with different superscripts are significantly different (P <0.05).

In this connection, the findings of Colombatto *et al.*, 2003 might explain the results through their suggested by which feed enzymes increase the digestion and the utilization of feedstuffs in ruminant diets through three main factors; 1) feeds are structurally very complex, containing a variety of polysaccharides, proteins, lipids, lignin, and phenolic acids, often in intimate association; 2) the enzyme products are mixtures of enzymes containing

many different activities, each of which differs in their optimal conditions and specificities; and 3) ruminal fluid is by nature an extremely complex microbial ecosystem, containing many hundreds of microbial species and their secreted enzymes. Feding values of the experimental diets are represented in Table 3. It is obvious that TDN and DCP were insignificantly increased with buffaleos fed on 30 and 40 g Alpha Yea Mix supplemented diets.

**Intake, Milk yield and milk composition:**

Based on nutrients requirement calculations, data in Table 4 indicated no differences among the different treatments in total dry matter intake or TDN and DCP intakes. Total DM intake represented about 2.4% of the average live body weight for all animals. Concentrate /roughage ratio was 55/45 on average for all the experimental diets. Rice straw intake tended to be higher with buffaloes fed diets supplemented with 20 and 30 g Alpha Yea Mix, compared to those fed the control and 40 g supplemented diets. Mwenya *et al.*, 2005 reported that DM intake was insignificantly changed when yeast was added to Holestin cows diet. Erasmus *et al.*, 1992 indicated that DMI was significantly increased when cows were fed on yeast supplemented diets. Milk yield of Holestin cows was significantly increased in response to yeast supplementation (Williams 1989 and Wohlt *et al.*, 1991).

**Table (4): Feed and nutrients intake, milk yield and constituents of buffaloes fed enzymes supplemented diets.**

Item	Enzyme supplementation, g/h/d				± SE
	Control (0)	20	30	40	
<b>Feed intake, Kg/h/d</b>					
Berseem	5.03	5.29	4.95	4.89	0.45
Rice straw	1.32	1.79	1.75	1.30	0.18
CFM	7.80	7.91	7.91	7.69	0.39
Total DM intake	14.01	14.99	14.60	13.87	0.84
<b>Nutriments Intake:</b>					
TDN, Kg	8.35	8.62	8.70	8.43	0.16
DCP, g	1351	1420	1427	1408	34.5
<b>Milk yield:</b>					
Actual milk, Kg	8.93	9.53	9.61	9.71	0.54
7% FCM, Kg	8.73	9.53	9.24	9.10	0.70
Proportional yield	100	106.7	107.6	108.7	
<b>Milk constituents:</b>					
Fat, %	6.64	7.10	6.92	6.76	0.46
Fat yield, Kg	0.595	0.685	0.651	0.666	0.06
Protein, %	2.55	2.98	2.59	2.43	0.24
Protein yield, g	228	295	244	235	27.0
Lactose, %	5.04	4.88	5.08	5.01	0.09
Urea, %	14.94	16.08	17.03	14.92	1.42
Ash, %	0.90	0.97	0.93	0.95	0.11
T. Solids, %	15.12	16.00	15.51	15.14	0.61
Solids Non Fat, %	8.49	8.92	8.60	8.38	0.24
Somatic cell Count	442	522	611	469	59.0

The data concerning actual milk yield, Kg/d (Table 4) indicated insignificant increase by 6, 7 and 9% in buffaloes consumed 20, 30 and 40 g Alpha Yea Mix supplemented diets respectively vs. the control diet. Fat corrected milk at 7% insignificantly differed among all treatments. Williams *et al.*, 1991, Wohlt *et al.*, 1991 and Schingoethe *et al.*, 2004 reported slight increase up to 15% of cows milk yield with yeast supplemented diets. Milk constituents including fat percent and yield, protein percent and yield, lactose, urea, ash, TS, SNF percentages and SCC are presented in Table 4.

All milk constituents showed insignificant differences among all experimental treatments. In this respect, Milk fat percentage (Harris *et al.*, 1992) and milk protein percentage (Harris and Webb, 1990) were found to be increased in response to yeast supplementation to dairy cows rations. On the other hand, Kung *et al.*, 2002, and Erasmus *et al.*, 1992 reported that milk fat and protein percentages were insignificantly changed when dairy cows consumed alfalfa plus corn silage based diet (45%) and supplemented with fibrolytic enzymes.

**Economic evaluation:**

The economic feasibility of supplementing yeast culture to lactating buffalo's diets are presented in Table 5. Feeding cost, milk return and economic efficiency were estimated as basic economic indicators for the supplement feasibility. The whole-market existed price of feed ingredients, raw milk and Alpha Yea Mix were used. Economic data indicated that milk net return over feeding cost was proportionally increased as yeast culture supplement increased. The absolute return as a result of yeast supplementation was 1.0, 1.5 and 2.2 LE/h/d for 20, 30 and 40 g Alpha Yea Mix, respectively.

**Table (5): Economic evaluation of enzyme supplementation into lactating buffaloes diet.**

Item	Enzyme supplementation, g/h/d			
	Control (0)	20	30	40
<b>Feeding cost, LE/h/d</b>				
Berseem	6.25	6.75	6.25	6.25
Rice straw	0.20	0.30	0.30	0.20
CFM	12.2	12.3	12.3	11.9
Enzyme Supp.	0.0	0.4	0.6	0.8
Total cost	18.65	19.75	19.45	19.15
<b>Milk return, LE/h/d:</b>				
Total return, LE/d	31.30	33.40	33.60	34.00
Return / feeding cost	12.65	13.65	14.15	14.85
<b>Economic efficiency:</b>				
Return increase, %	100	108	111.9	117.4
Absolute return, LE/d	0.0	1.00	1.50	2.20

Ingredients and raw milk market prices are as follows: Rice straw = 150 LE/ton, Berseem = 250LE/ton, CFM = 1400 LE/ton, Enzyme = 20 LE/Kg, Raw milk = 3.50 LE/Kg

**CONCLUSION**

Supplementing the Egyptian lactating buffaloes diets with yeast culture products (Alpha Yea Mix®) at 30 – 40 g per animal daily significantly increased

fiber digestion with special reference to NDF, ADF and cellulose. Milk yield of buffaloes consumed yeast culture was increased however insignificantly, but improved the economic return over the supplement cost. It was obvious that adding yeast cultures and other exogenous fibrolytic enzymes might have different responses with the different fiber structures existed in the diet. So, further investigations are needed to test the most effective fibrous ingredients properly respond to yeast or enzyme supplementation.

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## إستجابة إنتاج وتركيب اللبن لإضافة الخميرة إلى علائق الجاموس الحلاب.

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غذيت أربعة جاموسات حلابة متعددة الولادات علي منتج (ألفا بي مكس) الذي يمثل مزرعة خميرة *Sacchaomyces cerevisiae* في تصميم المربع اللاتيني  $4 \times 4$  بمعدل صفر، ٢٠، ٣٠، ٤٠ جم/رأس / يوم. إستخدمت جاموسات حلابة متوسط وزنها  $591 \pm 75$  كجم ومتوسط إنتاجها من اللبن  $10,2 \pm 0,75$  كجم و ٧٠ يوم حليب وفي موسم الحليب الثالث والرابع . يهدف البحث إلي دراسة تأثير إضافة الخميرة علي هضم المركبات الغذائية وإنتاج وتركيب اللبن. استمرت كل فترة تجريبية إلي ٣٠ يوما. أخذت عينات روث من المستقيم لمدة ثلاثة أيام متتالية، كما أخذت عينات اللبن لمدة خمسة أيام متتالية بنهاية كل فترة تجريبية للتحليل المعمل.

أوضحت النتائج عدم تأثير معام هضم كل من المادة الجافة والعضوية والمستخلص لخالي من الأزوت واللجنين والهيمي سليلوز بين المعاملات المختلفة. كان هناك اتجاهها للزيادة غير المعنوية لكل من معام هضم البروتين الخام ومستخلص الإثير مع الجاموس الذي تناول عليقة بها ٣٠ أو ٤٠ جم خميرة/ رأس / يوم. زادت معنويا معاملات هضم الألياف الخام ومكوناتها المختلفة ADF ، NDF ، السليلوز بنسبة تتراوح بين (١٠-٢٢%) في الجاموس الذي تغذى علي عليقة بها ٣٠ أو ٤٠ جم خميرة/ رأس/ يوم مقارنة بتلك التي تناولت عليقة المقارنة أو ٢٠ جم خميرة. زادت القيمة الغذائية في صورة مركبات كلبية مهضومة وبروتين خام مهضوم غير معنويا في علائق الجاموس الذي تناول المستوى المرتفع من إضافة الخميرة. وبخصوص إنتاج اللبن فقد زاد غير معنويا مع الجاموس الذي غذى علي ٣٠، ٤٠ جم خميرة/ يوم. لم تتأثر كل مكونات اللبن (نسبة وكمية الدهن، نسبة وكمية البروتين ونسبة اللاكتوز واليوريبا والأملاح المعدنية والجوامد الكلية والجوامد اللادهنية وأيضاً عدد الخلايا الجسمية) نتيجة إضافة الخميرة بمستوياتها المختلفة إلي عليقة الجاموس الحلاب. تم حساب بعض المؤشرات الإقتصادية البسيطة الدالة علي تحليل التكلفة والعائد، والتي أوضحت أن صافي عائد إنتاج اللبن بعد خصم تكاليف التغذية كان أعلى بشكل مضطرد مع زيادة كمية الخميرة المضافة إلي العليقة.

يستنتج من هذه الدراسة أنه يمكن إضافة منتجات خميرة *Sacchaomyces cerevisiae* ( $10 \times 2$ ) / جم بمعدل ٣٠ - ٤٠ جم/ رأس/ يومياً إلي الجاموس الحلاب لزيادة إنتاج اللبن وكذلك صافي العائد.

الكلمات المفتاحية: مزارع الخميرة، الإضافات، الجاموس ، معام هضم الألياف، إنتاج اللبن.

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