## INFLUENCE OF LIVE YEAST CULTURE ON MILK PRODUCTION, COMPOSITION AND SOME BLOOD METABOLITES OF OSSIMI EWES DURING THE MILKING PERIOD

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## ABSTRACT

A feeding trial was conducted to evaluate the influence of live yeast culture (Saccharomyces cerevisiae) on milk production, composition, and some blood metabolites of Ossimi ewes during the milking period. The control group(G1) was fed a concentrate mixture (CFM) and hay (H) and grazed twice daily, while the second group (G2) and third group (G3) were fed the same diet supplemented with 3 or 6g of live yeast culture (Yea Sacc1026), respectively. The treated groups had significantly higher values (p<0.05) for fat corrected milk (FCM) (740, 605, 571 g/day, for G3, G2 and G1 vs, respectively), while the values for milk yield, fat yield and lactose yield were higher (p<0.05) only in G3 compared with G1. Milk yield values were constantly higher in G3 than in G1 while the values for the G3 were more variable during milking. Milk composition was not significantly affected by yeast supplementation with the exception of urea values which were significantly (p < 0.05) lower in G3. Yeast administration influenced β-hydroxy-butyrate (BHB) values, which were significantly (p<0.05) higher in the treated groups; and non-esterified fatty acids (NEFA) values, which were significantly (p<0.05) higher only in the G3 compared with the G1. Other blood metabolites values were not influenced by the treatments. It was concluded that supplementation with live yeast culture, under the conditions of this experiment, had a significant effect on the performance and metabolism of Ossimi ewes during the milking period. Based on more constant results, it is recommend to include live yeast culture (Yea Sacc1026) at 6g/animal/day as appropriate level for field conditions. Keywords : live yeast culture, metabolic profile, ewes, milking.

## INTRODUCTION

Sheep milk is uniquely different from cow or goat milk. Sheep milk has about twice the fat of cow milk and 40% more protein than cow milk. In the last twenty years, some probiotics, such as Aspergillus or A. Niger (Pioneer, 1989), yeast culture (*Saccharomyces cerevisiae*) (Wallace,1994) and some microbial growth promoters e. g.thiamine ,niacin (Shields,1981) were used as feed additives in order to improve rumen conditions and cellulose digestion in the rumen and milk yield of dairy cows. Inactive dry yeast is only used to improve the yield and composition of milk in sheep and also as a source of protein and vitamins of B-complex, when added to rations (Dilanyan *et al.* 1974 and Peppler 1979). Products containing *Saccharomyces cerevisiae* vary widely in efficiency, primarily because of differences in strain and the viability of yeast cells . Numerous models have been designed to explain the effects of yeast in the rumen. Data indicate that supplementation of yeast in the ruminant diet may improve feed intake (Williams *et al.*1991 and Robinson and Garrett 1999), milk production (Wang

*et al.* 2001 and Abd El-Ghani 2004), weight gain (Salama *et al.* 2002), digestion (Wohlt *et al.*1991 and Jouany *et al.*1998), numbers of anaerobic and cellulolytic bacteria (Newbold *et al.*1995), ruminal pH value (Doreau and Jouany 1998;Jouany *et al.*1998), and alter the patterns of volatile fatty acids (Arcos-Garcia *et al.*2000) or even supply the animal with unknown growth factors (Girard and Dawson 1995). Nevertheless, the results of these studies have been variable and strongly influenced by ration composition (Dawson 1992 and Newbold 1996). The influence of yeast supplementation on grazing animals has mainly been investigated in grazing steers (Olson *et al.*1994a,1994b and Arakaki *et al.* 2000). Much less is known about the effects of yeast supplementation on grazing dairy ewes, nevertheless in vitro trials (El Hassan *et al.*1994) and trials on grazing steers may give justification for more investigation.

## MATERIALS AND METHODS

The live yeast culture supplements: (Saccharomyces cerevisiae)

(Yea Sacc 1026 ; Alltech, Inc., Nicholasville, Kentucky, USA ) Feeding and management:

The present study was carried out at the Experimental farm of Animal Production Department, Faculty of Agriculture, South Valley University, Qena during the period from February to July 2010. A feeding trial was conducted to evaluate the influence of live yeast culture (Saccharomyces cerevisiae) on milk production, composition, and blood metabolites of Ossimi ewes during the milking period. Sixty Ossimi ewes (aged 3-3.5 years, average body weight 48.3  $\pm$  3.13 kg) were used in the lactation trial from the 42 <sup>th</sup> to the 182 th day of lactation, which is the usual period of milking in the region. All ewes were in second lactation. The sheep were divided into three groups on the 42 th day of lactation after peak to carry out the experiment. The sixty Ossimi ewes at peak of lactation (42day) were divided into three groups (20animals/each): Control group(G1): received only100% of NRC (2001) nutrient allowances of dairy sheep without live yeast culture for 6 weeks after parturition, G2: received 100% of NRC nutrient allowances of dairy sheep (2001) with 3 g/day/sheep of live yeast culture for 6 weeks after parturition and G3: received 100% of NRC nutrient allowances of dairy sheep (2001) with 6g/day/sheep of live yeast culture for 6 weeks after parturition.

Animals were kept in open yards belonging to Animal Production Experimental Farm, Faculty of Agriculture, South Valley University. During the experimental period the animals received 1 kg/ewe/day of concentrate mixture, 0.3 kg/ewe/day rice straw and were allowed to graze (mixed grass pasture and alfalfa hay) from 7.00a.m and 3.00p.m. Animals were fed(at 7.00a.m and 5.00p.m) ration consisted of a concentrate mixture according to their live body weight and level of milk production. Beside the concentrate mixture, animals were fed mixed grass pasture(natural pasture, cereal stubble, crop residue, vegetable by-products )and alfalfa hay. Water was available all day and minerals were supplied in salt licking blocks. Animals were adopted the double daily milking at 6.0a.m. and 5.00 p.m. The daily control ration consisted of concentrate mixture: 36.5%yellow maize,

16% wheat bran, 16% sunflower meal, 8% soybean meal, 20% barley meal, 2% calcium carbonate and 1% sodium chloride, and 0.5% mineral and vitamins additives. The rations were fed to ewes as Total Mixed Rations, based on NRC (2001). The Total Mixed Rations comprised of 65% forage and 35% of a concentrate mix to formulate diets to meet NRC (2001). Approximate and analysis of the concentrate mixture, mixed grass pasture and alfalfa hay are provided in Tables (1 and 2) according to AOAC(1995). Body weight of animals was recorded at the beginning and at the end of the experiment.

| Ingredients             | Concentrate mixture |
|-------------------------|---------------------|
| Soybean meal            | 8%                  |
| Yellow maize            | 36.5%               |
| sunflower meal          | 16%                 |
| Barley meal             | 20%                 |
| Wheat bran              | 16%                 |
| Calcium carbonate       | 2%                  |
| Sodium chloride         | 1%                  |
| Vitamin-mineral mixture | 0.5%                |
| Total                   | 100.0               |

Table (1): Formulation of the concentrate mixture diet .

| Table (2): Composition and chemical | analysis of experimental basal diet |
|-------------------------------------|-------------------------------------|
| of rations.                         |                                     |

| Chemical composition    | Concentrate | Mixed grass | Alfalfa hay |
|-------------------------|-------------|-------------|-------------|
| (%):                    | mixture     | pasture     |             |
| Dry matter              | 88          | 21          | 89          |
| Ash                     | 7.5         | 8.1         | 7.2         |
| Crude fat               | 3.0         | 3.8         | 2.3         |
| Crude protein           | 17.3        | 11.2        | 15.9        |
| Crude fibre             | 9.7         | 33.5        | 33.2        |
| Neutral detergent Fibre | 16.9        | 59.5        | 52.3        |
| Acid detergent fibre    | 7.1         | 36.4        | 38.6        |
| Calcium                 | 0.66        | 0.38        | 1.28        |
| Phosphorus              | 0.78        | 0.30        | 0.29        |
| Magnesium               | 0.24        | 0.14        | 0.30        |
| Sodium                  | 0.23        | 0.19        | 0.12        |
| Sulphur                 | 0.05        | 0.15        | 0.24        |
| Potassium               | 0.81        | 1.68        | 1.9         |
| Chloride                | 0.08        | 0.57        | 0.37        |
| Zinc (mg⋅kg-1)          | 96          | 28 22       | 22          |
| Manganese (mg⋅kg-1)     | 75          | 89 66       | 66          |

# 3–Samples collection and measurement :

Feed samples:

Samples of the concentrate, hay and pasture were collected throughout the experimental period for chemical composition analyses. The samples were ground and analyses were made according to the AOAC (1999). Neutral Detergent Fibre (NDF) and Acid Detergent Fibre (ADF) were

determined by detergent procedure of Robertson and Van Soest (1981) and Van Soest *et al.* (1991), with alpha amylase (SIGMA-ALDRICH, Inc., USA) being added during NDF extraction. Sodium sulphite was not added. Feed samples were dried to ashed at 600°C/6 hrs. then after Calcium, Sodium, Potassium, Chloride , Magnesium, Sulphur , Zinc and Manganese concentrations were measured by using PV9100 atomic absorption spectrophotometer and they were analyzed for Phosphorus by using Varian DMS 1005 UV Visible Spectrophotometer (A.O.A.C, 1990).

## Milk and Blood samples:

Individual milk samples, consisted of proportional volumes of morning and evening milk, were collected in order to evaluate milk composition (5 ml/kg of produced milk). A composed milk sample of each ewe was analyzed weekly. Fat percentage was determined by the standard Gerber method according to the British Standard Institute (1962). Protein percentage of milk was evaluated by Micro Kjeldahl technique (A.O.A.C, 1999). Total solids (TS) percentage of milk was determined gravimetrically using the method by Oser (1965). Solid not fat (SNF) was calculated by the difference (T.S%-fat%). Milk yield was corrected to 7%fat (Raafat and Saleh 1962). 7% FCM=0.265×milk yield (kg) +10.5 × fat yield (kg). The urea values were determined by an enzymatic colorimetric method using commercial kits of reagents (Patton and Grouch 1977). Somatic cell counts (SCC) was determined with by the citoflow method, using the instrument Foosomatic 90. pH : It was determined by using a pH meter combined with a glass electrode (Model SS-3, Beckman, Fullerton, CA, USA

Blood samples were collected on the  $42^{th}$ ,  $112^{th}$  and  $182^{th}$  day of lactation by puncture of the jugular vein, with the addition of heparin as an anticoagulant, prior to morning feeding. Blood was allowed to coagulate at room temperature. The blood plasma was separated by centrifugation and stored at -20°C for a maximum of 60 days until assayed. Obtained blood serum were subjected to determine blood plasma constituents as described by Wiebe and Bernert (1984), Kaplan and Szalbo (1983) , Doumas(1971) and Trinder (1969).

#### 4- Statistical analysis of the data :

Data were statistically analyzed according to the General Linear Model G.L.M) by using SAS (1998) and the differences among means were detected by Duncan's Multiple Range Test (Duncan, 1955).

## RESULTS

The values for milk yield and composition are presented in Table 3. Supplementation with live yeast culture only significantly (p < 0.05) increased the total milk yield during the experimental period in G3 although G2 also had a higher values, but non-significantly, milk yield than the control group. Values for FCM were higher in the treated groups and the differences were significant(p<0.05) than in G1(control).Fat yield and lactose yield significantly (p<0.05) increased only in G3 compared to G1. The chemical composition of the milk was not influenced by the treatments with the exception of milk urea nitrogen which was significantly (p<0.05) lower in the G3, the differences were significant. Values of milk yield and chemical composition did not differences

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from the respective values recorded in other animals in the herd (not included in the experiment) kept on the experimental farm. The values of blood biochemistry are summarised in Table 4. Values for  $\beta$ -hydroxy-butyrate (BHB) were significantly (p<0.05) higher in the treated groups than in the G1 group and non-esterified fatty acids (NEFA) values were significantly (p<0.05) higher in G3 than in the control group. All other values concerning blood components were not significantly different among other groups.

|                       | G1               | G2               | G3               |
|-----------------------|------------------|------------------|------------------|
| Composition of milk   | 0g of live yeast | 3g of live yeast | 6g of live yeast |
|                       | culture per day  | culture per day  | culture per day  |
| NO                    | 20               | 20               | 20               |
| Milk yield (g/day)    | 527± 55b         | 564±53ab         | 670 ± 76a        |
| Fat (%)               | 7.8 ± 0.3        | 7.7 ± 0.3        | 8.0 ± 0.1        |
| Protein (%)           | 5.8 ± 0.47       | 5.7 ± 0.35       | 5.7 ± 0.51       |
| Lactose (%)           | $4.4 \pm 0.1$    | $4.4 \pm 0.2$    | 4.3 ± 0.2        |
| Total solids (%)      | 19.0 ± 0.8       | 18.8 ± 0.5       | 19.5 ± 0.8       |
| Non-fat solids (%)    | 11.2 ± 0.4       | 11.0 ± 0.3       | 11.1 ± 0.6       |
| FCM7(g/day)*          | 571.62 ±63b      | 605.48 ± 73a     | 740.35 ± 81a     |
| Fat yield (g/day)     | 73± 8b           | 76 ± 8b          | 85 ± 15a         |
| Protein yield (g/day) | 54 ± 5           | 56 ± 6           | 60 ± 8           |
| Lactose yield (g/day) | 38 ± 5b          | 43 ± 3ab         | 47 ± 8a          |
| SCC (X 103/ml)        | 405 ± 169        | 380 ± 75         | 490 ± 15.2       |
| Urea N (mg/100ml)     | 27.4 ± 2.46a     | 27.16 ± 1.95a    | 25.01 ± 2.55b    |
| Ph                    | 6.76±0.44 a      | 6.72±0.36 a      | 6.74±0.43 a      |

Table (3): Means  $\pm$ SE for milk yield and composition of Ossimi ewes as affected by yeast supplementation in the ration .

<sup>a and b</sup> means in the same row followed by different letters are significantly different (p<0.05 ). \* FCM7% = fat corrected milk (7% milk fat);

| Table (4): Means ±SE for blood plasma constitunits of sheep groups fed |
|--|
| on different levels of live yeast culture.                             |

|  | G1               | G2               | G3               |
|--|------------------|------------------|------------------|
| <b>Biochemical indicators</b>                          | 0g of live yeast | 3g of live yeast | 6g of live yeast |
|  | culture per day  | culture per day  | culture per day  |
| NO   | 20               | 20               | 20               |
| NEFA (mmol <sup>-</sup> l <sup>-</sup> 1)*             | 0.30 ± 0.10b     | 0.37 ± 0.20ab    | 0.38 ± 0.12a     |
| BHB (mmol <sup>-</sup> l <sup>-</sup> 1) <sup>**</sup> | 0.51 ± 0.08b     | 0.61 ± 0.14a     | 0.60 ± 0.12a     |
| Urea (mmol <sup>-</sup> l <sup>-</sup> 1)              | 7.40 ±1.39       | 7.31 ± 0.86      | 6.94 ±1.17       |
| Triglycerides(mmol <sup>-1-</sup> 1)                   | 0.25 ± 0.06      | $0.24 \pm 0.06$  | 0.28 ± 0.07      |
| Cholesterol (mmol <sup>-</sup> l <sup>-</sup> 1)       | 1.92 ± 0.23      | 1.91 ± 0.25      | 1.96 ± 0.16      |
| VLDL (%)   | 6.3 ± 4.1        | 6.1 ± 4.0        | 7.7 ± 4.0        |
| HDL (%)  | 44.5 ± 12.1      | 46.6 ± 8.7       | 48.5 ± 8.8       |
| LDL (%)  | 49.1 ± 12.6      | 47.2 ± 7.9       | 43.7 ± 10.0      |
| AST (µkat⊺1)   | 2.55 ± 1.01      | 2.78 ± 1.30      | 2.45 ± 0.95      |
| ALT (µkatT1)   | 0.31 ± 0.15      | 0.23 ± 0.12      | 0.26 ± 0.13      |
| GGT (µkatT1)   | 0.82 ± 0.07      | 1.07 ± 0.20      | $0.93 \pm 0.33$  |
| ALP (µkatī]1)  | $3.82 \pm 0.98$  | 3.75 ± 1.43      | 3.28 ± 1.30      |

a and b means in the same row followed by different letters are significantly different (p<0.05).

\* β-hydroxy-butyrate=BHB

\*\* Non-esterified fatty acids= NEFA

## DISCUSSION

In this study, the live yeast culture included the ewes' diet showed a positive effect on milk yield during lactation. This has also been reported in dairy cows (Wohlt *et al.*1991 and Robinson and Garrett 1999 and Wang *et al.* 2001), also the same trend in dairy goats was observed by (Reklewska *et al.* 2000 and Abd El-Ghani 2004 and Stella *et al.* 2007). In contrast, other authors found no improvement of milk yield in dairy cows (Arambel and Kent 1990 ; Swartz *et al.* 1994 and Soder and Holden 1999), dairy goats (Hadjipanayiotou *et al.* 1997; Giger-Reverdin *et al.* 1996) or in dairy ewes (Hadjipanayiotou *et al.* 1997). These results reflect that the effects of live yeast culture.

Administration were strongly influenced by diet composition. Although many authors stated that live yeast cultures are most efficient when animals are fed diets poor in nutrient supply (Plata et al. 1994 and Jouany et al. 1998 ) or high concentrate diets overloaded with energy (Williams et al. 1991 and Zelenak et al. 1994), in some cases it is difficult to find a correlation between diet composition and the results of yeast supplementation. The animals in the present were fed relatively high levels of concentrate (1 kg/animal/day) which could lead to improved buffering capacity in the rumen. the results were also dose-dependent because 3 g of live yeast cultures per day was not efficient enough to maintain a constantly higher milk yield than in the control group. Similar results were obtained by Abd El-Ghani (2004) with 3 and 6 g of live yeast cultures per day fed to dairy goats. Due to the higher amount of total solids in sheep milk, compared to cows and goats, it is expected that the supplementation of yeast may be more efficient in changing milk composition. However, the milk fat content was not significantly higher in the treated groups than in the control group, which is in agreement with Piva et al. (1993) who stated that the common result of yeast supplementation to dairy cows is only a slight (nonsignificant) increase in the milk fat content. Hadjipanayiotou et al. (1997) and Stella et al. (2007) also reported no increase in milk fat content in dairy goats. In Damascus dairy ewes Hadjipanayiotou et al. (1997) found no influence of live yeast administration on milk composition, although in their study the yeast was steam-pelleted with no report on cell viability. On the contrary, Giger-Reverdin et al. (1996), Abd El-Ghani (2004) and Masek et al. (2008) found increased milk fat values in dairy goats and ewes. Milk protein and lactose values did not differ between the treatments, which was also noticed by the majority of authors (Stella et al. 2007; Giger-Reverdin et al. 1996). Milk urea values were significantly (p>0.05) lower in the group fed 6 g per day. Harrison et al. (1988) reported a much lower concentration of rumen ammonia N after yeast supplementation, which is in agreement with the results of Erasmus et al. (1992), who found that the mean concentration of rumen ammonia decreased by 10% after live yeast culture supplementation. Erasmus et al. (1992) explained these reduced concentrations of ammonia in the rumen as the result of increased incorporation of ammonia into microbial protein stimulated microbial activity which could explain lower blood and milk urea values the present experiment. Results significantly subsequent showed (p<0.05) higher non-

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esterified fatty acids (NEFA) and β-hydroxy-butyrate (BHB) values were presently recorded in the treated groups, which is in agreement with Giger-Reverdin et al. (1996) and Quigley et al. (1992). Increase in non-esterified fatty acids (NEFA) values could be explained by increased mobilisation of fat tissue caused by live yeast supplementation, which was also noted in dairy goats (Giger-Reverdin et al. 1996). According to Quigley et al. (1992), the increased ruminal butyrate was at least partially responsible for increased BHB values. Triglycerides and cholesterol values tended to be higher in the treated groups, which was also noted by Pysera and Opalka (2001). The same authors also found, in contrast to our results. All metabolites values were within the normal reference range for lactating dairy ewes (Dubreuil et al. 2005; Roubies et al. 2006; Yokus and Cakir 2006 and Masek et al. 2007). Literature dealing with yeast supplementation in grazing animals is scarce and to our knowledge, involves mainly steers. Various authors found an increased number of protozoa increased the live body weight gain (Arakaki et al. 2000), (Combellas et al. 2002) and increased degradation and digestibility (Olson et al. 1994a, 1994b). (Dawson 1992; Wallace and Newbold 1992; Newbold et al. 1995), showed that the micro-population plays a key role in the mode of action of yeast in the rumen El Hassan et al. (1994) dound that the Yea Sacc1026 stimulated the total bacterial number in a rumensimulating fermentor when the basal diet was grass and increased, the number of cellulolytic bacteria. Subsequent increased the degradalility and digestion then fore the better performance and best daily weight gain. I concluded that the supplementation of live yeast culture (Yea Sacc1026) had a significant beneficial effect on the milk yield of Ossimi sheep, fed pasture and concentrate mixture during the milking period. The significant results were probably a result of the interactions between yeast culture supplementation and diet composition. Since the influence was dosedependent, we could recommend 6 g per day for inclusion in dairy sheep diets. Additional studies under different feeding conditions and in earlier stages of lactation should clarify the influence of live yeast supplementation in the diets of Ossimi ewes and define the dietary situations in which it may be beneficial.

It could be concluded that the supplementation of live yeast culture (Yea Sacc1026) had a significant beneficial effect on the milk yield of Ossimi sheep, fed pasture and concentrate mixture during the milking period. The significant results were probably a result of the interactions between yeast culture supplementation and diet composition. Since the influence was dosedependent, 6 g per day for inclusion in dairy sheep diets, is recommended . Additional studies under different feeding conditions and in earlier stages of lactation should clarify the influence of live yeast supplementation in the diets of Ossimi ewes and define the dietary situations in which it may be beneficial.

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تأثير إضافة الخميرة الحية بالعليقة على أنتاج وتركيب اللبن وبعض مكونات الدم البيوكميانية في النعاج الاوسيمى بصعيد مصر. أحمد عبد الجليل بيومي خليل قسم الإنتاج الحيواني- كلية الزراعة - جامعة جنوب الوادي بقنا.

أجريت هذه الدراسة بمزرعة الإنتاج الحيواني- كلية الزراعة- جامعة جنوب الوادي بقنا من أول فبراير ٢٠١٠ إلى آخر يوليو ٢٠١٠. لمعرفة تأثير استخدام الخميرة (Yea Sacc1026) على الصفات الإنتاجية في النعاج الاوسيمى ( إنتاج وتركيب اللبن ومكونات الدم البيوكميائية ) واستخدم في هذه الدراسة عدد (٢٠ نعجة اوسيمى حلابة) بعد وصولها لقمة الإنتاج من موسم حليبها عند اليوم ٤٢ . وقسمت النعاج إلى ثلاث مجموعات متساوية- الأولى العليقة الأساسية - عيث غذيت فيها الحيوانات على العليقة الأساسية. الثانية: غذيت الحيوانات على العليقة الأساسية + ٣جرام من مستحضر الخميرة- الثالثة: غذيت الحيوانات على العليقة الأساسية + ٣جرام من مستحضر الخميرة - الثالثة: غذيت الحيوانات على العليقة وزن الحيوانات عند بداية ونهاية التجربة . وتم تحليل اللبن وقدرت نسب الدهن والبروتين والجوامد اللاماسية + ٣جرام من مستحضر الخميرة - وكانت المدة الكلية للتجربة ١٨٠ يوم وقد تم وزن الحيوانات عند بداية ونهاية التجربة . وتم تحليل اللبن وقدرت نسب الدهن والبروتين والجوامد اللادهنية والجوامد الكلية ونسبة الرماد وكذلك قدرت بعض قياسات الدم ،ويمكن تلخيص النتائج وزن الحيوانات عند بداية ونهاية التجربة . وتم تحليل اللبن وقدرت نسب الدهن والبروتين والجوامد اللادهنية والجوامد الكلية ونسبة الرماد وكذلك قدرت بعض قياسات الدم ،ويمكن تلخيص النتائج والجوامد الكلية ولا توجد فروق معنوية في اليوريا . كما كان هناك فروق معنوية بين استخدام والجوامد الكلية ولا توجد فروق معنوية في اليوريا . كما كان هناك فروق معنوية بين استخدام والجوامد الكلية ولا توجد فروق معنوية في النتائج المتحصل عليها . كما أظهرت النتائج زيادة في قياسات الدم . وعلى ذلك فانه يمكن استخدام مستوى ٦ جرام/رأس في اليوم للنعاج الاوسيمى الحلاب أثناء موسم الحلب .

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