



## Feeding pattern, metabolic status and milk composition of Chilika buffaloes in their natural habitat

S MOHANTY<sup>1</sup>, N PANDA<sup>2</sup>, B PANIGRAHI<sup>3</sup>, R K SWAIN<sup>4</sup>, S K DASH<sup>5</sup>, A MISHRA<sup>6</sup> and S S GIRI<sup>7</sup>

Orissa University of Agriculture and Technology, Bhubaneswar, Odisha 751 003 India

Received: 10 June 2016; Accepted: 1 November 2016

### ABSTRACT

Chilika buffaloes, recognized as a breed since 2009 vide accession no. India\_Buffalo\_1500-Chilika\_01012, are found in and around Chilika lake of Odisha. These buffaloes are reared under purely extensive system with no feed and medicine supplementation. The present study was undertaken to know the feeding pattern, metabolic status and milk composition of Chilika buffaloes in their natural habitat as very few literature is available about these buffaloes which are reared with zero external input system. The major portion of their feed is composed of different brackishwater weeds growing in and around the lake and grasses growing in the shallow water beds. The study was undertaken taking 60 buffaloes in their first to fourth lactation from 3 villages. The proximate composition of weeds and grasses were comparable with a good quality fodder but having higher acid insoluble ash. From the serum metabolic status it was seen that the protein level was adequate in the animals but the glucose level below normal range. These buffaloes have optimum serum level of major and minor minerals except Na, K and Fe, which is higher than the normal values. The milk fat content was higher than other buffaloes. It was found that among the PUFA the ratio of n3 to n6 was quite good which was higher than cattle and comparable with the fish. Further research is required to explore the availability of the biomass and exploit the potentiality of these buffaloes for the rich n:3 fatty acid content in the milk to be utilized by the today's health conscious society.

**Key words:** Chilika buffaloes, Feeding pattern, Metabolic status, Milk fatty acid

Chilika buffaloes which are recognized as a breed since 2009 vide accession no. India\_Buffalo\_1500-Chilika\_01012 is found in and around Chilika lake of Odisha, the largest brackish water lake in Asia (area of 1165 km<sup>2</sup>). Mishra *et al.* (2009) investigated these buffaloes cytogenetically that possess a somatic chromosome count of 50, identical to that of typical riverine buffaloes comparable with Indian riverine buffalo breeds like Nagpuri, Murrah and Toda. These buffaloes are reared under purely extensive system with no feed supplementation, not even straw or grass which come a population of 29,000 covering in and around Chilika Lake (Sethi *et al.* 2007). These animals have been contributing the livelihood support of more than 5,000 landless and marginal farmers of these areas. These animals not only provide handful amount by selling the milk but also draught in agricultural operations. The animals graze throughout the night in the water bodies

spending more than 12 h in the lake and only come to the resting place near the villages in the morning time, where they are milked, provided sufficient fresh drinking water and allowed to take rest till afternoon when they are again escorted back into the water bodies.

The major portion of their feed is composed of different brackishwater weeds growing either free floating or submerged and also some of the grasses growing in the shallow water beds or near the banks. The local names of 2 such grasses, which are very much relished by these buffaloes are 'Chhera' and 'Pitta', which are mainly of Poaceae family and *aeluopus* genus. The buffaloes have the ability to immerse into the salty water and pick up the submerged grasses which is unique quality for these buffaloes. It is often noticed that the Murrah upgraded progenies of these buffaloes do not show this type of feeding behavior of picking up the submerged grasses. As very few literature is available on these buffaloes, it is an urgent need to conserve this native germplasm, the present study is planned to know the feeding pattern, serum status and milk composition of these buffaloes.

### MATERIALS AND METHODS

*Selection of experimental animals:* The study was undertaken in 3 villages namely Panasapadar, Gambhari and Satapada on 60 buffalo cows selected randomly

Present address: <sup>1</sup>Addl. VAS (dr.symphony@gmail.com), VD, Kendrapada. <sup>2</sup>Associate Professor (npandaouat@gmail.com), Department of Animal Nutrition, College of Veterinary Science and Animal Husbandry. <sup>3</sup>Associate Professor (bpanigrahi65@yahoo.co.in), Department of LPM. <sup>4</sup>Professor and Head (rkswainovc@yahoo.com), Department of Animal Nutrition. <sup>5</sup>Professor (susantdash46@gmail.com), ABG. <sup>6</sup>PhD Scholar (drakasmishra88@gmail.com), NDRI. <sup>7</sup>Principal Scientist and Head (ssgiri123@yahoo.co.in), CIFA.

amongst the lactating ones (within 1 to 5 months of lactation) from 9 farmers. The buffaloes were within their first to 4<sup>th</sup> lactation period. The average body weight of the animals was around 300 kg and the selected animals have no external sign of physiological disorder.

*Collection of samples and parameters studied:* These buffaloes are neither fed any concentrate mixture nor any feed ingredient. They thrive on only grazing the sea weeds and grasses grown nearby Chilika lake. Three different samples were collected, one of them being a type of saline water weed growing purely in water. The other two are grasses growing in shallow water beds and on the banks respectively. Just after collection, the samples were washed properly with freshwater to remove extra dust and dirt. Proximate composition (AOAC 2005), macro mineral contents like Ca, P, Na, K, Mg and micro mineral contents like Zn, Cu, Mn and Fe were analyzed by atomic absorption spectrophotometer (ELICO).

For serum mineral and metabolic status study, blood samples were taken from 30 animals in the morning hour just after completion of milking. One day prior the owners were informed about it, which helped for the preparedness. The serum samples were carried to the laboratory as quick as possible in ice box. The serum samples were stored in deep freeze at a temperature of (-) 20 °C in properly labeled for analysis.

To study the milk parameters and fatty acid profile, milk samples were collected from 30 buffaloes in batches in the morning hour, during their usual time of milking. 100 ml of milk sample from each animal was collected in sterile container. The sample containers were brought to the laboratory as soon as possible in vaccine carrier. The fatty acid profile was analyzed at Central Institute of Freshwater Aquaculture (CIFA), Bhubaneswar by gas chromatography method (Shimadzu GC-2010, Kyoto, Japan). The column used in gas chromatography for separation of fatty acids was DB 225, which reads the fatty acids from C-8 to C-24 of both saturated and unsaturated type. Lipid extraction from the milk was done by using dichloromethane method (Stefanov *et al.* 2010). Milk fat, SNF, total solid, crude protein, lactose were as per the standard procedure.

*Statistical analysis:* The statistical analysis of the data was done according to Snedecor and Cochran (1994). The data were analyzed for analysis of variance (ANOVA) and DMR test (Duncan 1955) was used to test the difference treatment means wherever necessary.

## RESULTS AND DISCUSSION

*Proximate values of fodder:* From the proximate analysis of the collected naturally grown saline water weeds from Chilika water beds and the grasses of submerged and dry land (Table 1) it was found that the moisture content was highest in the brackishwater weed followed by the submerged grass and dry land grass at a level of 91.39, 77.22 and 71.11% respectively. The crude protein content was 19.68, 18.08 and 18.06% respectively for the weed, sub merged grass and dry land grass, which was comparable

Table 1. Proximate values of the fodders taken by the buffaloes (DM basis)

| Parameters (%) | Brackish water weed | Submerged grass | Grass on dry land |
|----------------|---------------------|-----------------|-------------------|
| Moisture       | 91.39±0.45          | 77.22±0.35      | 71.11±1.73        |
| Crude protein  | 19.68±0.43          | 18.08±0.57      | 18.06±0.20        |
| Ether extract  | 0.23±0.03           | 0.18±0.03       | 0.36±0.12         |
| Crude fibre    | 17.75±0.40          | 28.35±0.73      | 31.45±1.20        |
| Total ash      | 26.21±2.44          | 11.17±0.08      | 11.33±.33         |
| AIA            | 9.34±1.14           | 2.03±0.102      | 2.97±0.13         |
| NFE            | 36.13±0.81          | 42.22±0.63      | 38.8±1.03         |

to any good quality fodder. This quality of the fodders may be responsible for maintaining the serum protein and albumin levels of the buffaloes (Table 3) and also fulfilling the other vital protein needs of the body without any extraneous supplementation of concentrates.

The ether extract of the fodders was 0.23, 0.18 and 0.36%, respectively, which is very low. The crude fibre of the 3 samples ranged from 17.35 to 31.45%, which maintain the bulkiness and rumen motility. The crude fibre from the grasses was at par with the other grasses, but that of weeds was much lesser, comparable with water hyacinth (Sen *et al.* 1978). Both total ash and acid insoluble ash content of the brackishwater weed is much higher in comparison to two other grasses (Table 1). Krishnaiah *et al.* (2008), found that the mineral content of sea weed species like *Caulerpa*, *Ulva*, *Sargassum*, *Gracilaria*, *Eucheuma*, *Gelidiell a* and *Kappaphycus*, commonly available in Sabah South China (Malaysia) had high proportions of ash content (20.56–40.5%). The green and brown seaweed ash content (37.27–40.5%) was higher than the red seaweeds (20.56–22.41%). The higher acid insoluble ash in these 2 water submerged fodders may be due to higher silt and silica deposition. The higher percentage of total ash reflects the higher content of total minerals including sodium and potassium. The higher acid insoluble ash in these 2 water submerged fodders may be due to higher silt and silica deposition. The NFE were 36.13, 42.22 and 38.8%, respectively, for weed, submerged grass and dry land grass. Banerjee *et al.* (2009) reported that the red seaweed collected from Gangetic delta, North east coast of India, had crude protein which varied from 4.01 to 16.03% and carbohydrate from 21.52 to 35.74% and the lipid from 0.17 to 0.24%, depending upon the seasons.

*Mineral status of the fodder:* The calcium and phosphorus of the weed, submerged grass and grass of dry land and fodder samples are given in Table 2. While the calcium in these 3 samples was comparable with any good quality fodder and even to some leguminous fodders, but the phosphorus level was too below the critical level. Though the dry land grass showed a little lower concentration for these elements, but still it contained more than normal concentration when compared with the results from other workers of different area (Udar *et al.* 2003). The concentration of magnesium in the fodders was below

Table 2. Mineral status of the fodders

| Name of minerals      | Brackishwater weed | Submerged grass | Grass on dry land |
|-----------------------|--------------------|-----------------|-------------------|
| <i>Macro minerals</i> |                    |                 |                   |
| Ca (g/100g)           | 1.58±0.08          | 2.03±0.13       | 2.65±0.10         |
| P (g/100g)            | 0.04±0.01          | 0.008±0.001     | 0.007±0.001       |
| Na (g/100g)           | 1.82±0.085         | 1.69±0.082      | 1.51±0.030        |
| K (g/100g)            | 1.82±0.14          | 1.73±0.14       | 0.98±0.07         |
| Mg (g/100g)           | 0.0185±0.0001      | 0.0175±0.007    | 0.0178±0.0003     |
| <i>Micro minerals</i> |                    |                 |                   |
| Zn (ppm)              | 48.74±2.09         | 49.11±1.73      | 57.20±0.27        |
| Fe (ppm)              | 1003.22±44.65      | 924.02±15.68    | 731.62±30.43      |
| Mn(ppm)               | 151.8±2.87         | 131.43±14.96    | 101.56±5.26       |
| Cu (ppm)              | 33.55±0.86         | 28.84±5.64      | 20.55±2.10        |

normal concentration when compared with the values of other fodders. The higher concentration of sodium and potassium in the fodder sample collected may be attributed to the salinity of the water of the Chilika Lake, which differs from 8 to 12 points depending upon the seasons. Through generations the Chilika buffaloes are well habituated of consuming these fodders with higher concentrations of sodium and potassium without showing any adverse physiological symptoms. The concentration of magnesium in the fodders below normal concentration when compared with the values of other fodders as estimated by different workers (Garg *et al.* 2002, Sukla *et al.* 2010). All the 3 samples showed almost nonsignificant difference in the magnesium content.

Among the micro minerals content of these fodders, zinc showed normal concentration, being lowest in the brackishwater weeds and highest in the grass of dry land. Perhaps this optimum concentration of zinc in these fodders is reflected in the serum concentration of the Chilika buffaloes. Iron concentration in the 3 fodder samples was exceptionally high. It was noticed that the salt water weeds have maximum iron concentration followed by the submerged grasses. Krishnaiah *et al.* (2008) reported that the iron content of the seaweeds found in Malaysia was in the range of 6.6–10.94 mg/100 g on dry weight basis.

Likewise the concentration of manganese in the collected fodder samples was also much high for the weed, submerged grass and dry land grass. The copper content of these fodders was within the normal range and comparable to other fodders of good copper source (Gowda *et al.* 2001, Ramana *et al.* 2001, Das *et al.* 2002, Sukla *et al.* 2010).

It was noticed that the grass of dry land was with higher concentrations of calcium among the macro minerals and zinc among the micro minerals. But in all other mineral status, the salt water weed was in upper hand followed by the submerged grass.

*Serum metabolic profile:* The total protein, albumin and globulin levels of serum of Chilika buffaloes are given in Table 3. The total protein and albumin levels of these buffaloes were at par with other buffaloes as reported by various workers (Hayashi *et al.* 2005, Singh *et al.* 2008,

Table 3. Serum minerals and metabolic profiles of Chilika buffaloes

| Minerals    | Mean values  | Metabolic profile    | Mean values |
|-------------|--------------|----------------------|-------------|
| Ca (mg/dl)  | 9.73± 0.17   | Total protein (g/dl) | 8.02±0.11   |
| P (mg/dl)   | 5.01±0.22    | Albumin (g/dl)       | 3.10±0.06   |
| Mg (mg/dl)  | 3.02±0.07    | Globulin (g/dl)      | 4.93±0.13   |
| Na (mmol/l) | 162.05± 1.80 | Glucose (mg/dl)      | 51.94±1.03  |
| K (mmol/l)) | 6.02± 0.22   | Cholesterol (mg/dl)  | 25.27±1.26  |
| Zn (ppm)    | 3.49± 0.26   | Triglyceride (mg/dl) | 158.93±2.45 |
| Fe (ppm)    | 86.12±3.93   |                      |             |
| Mn(ppm)     | 1.17±0.14    |                      |             |
| Cu (ppm)    | 1.10± 0.09   |                      |             |

Hagawane *et al.* 2012). The globulin content was higher than the Murrah buffaloes (Wadhwa and Bakshi 2006) and buffaloes of Terai region of Nepal (Hayashi *et al.* 2005). This higher content of globulin in these buffaloes may be providing them better immune status as the morbidity and mortality rates are much lower as compared to other bovines, without even any special care and health management. Through generations, these buffaloes have adopted well with the harsh situation and climatic condition of Chilika Lake. The cause of facing minimal health hazards by these animals even after continual grazing in the brackishwater, being maintained without any extra care and management, without any supplementary feeding and even many a times without vaccination and medication also, may have some relation with this higher globulin concentration. Through our frequent visit to these villages and interaction with the farmers, it was reported that the crossbreds (Murrah × Chilika) are often prone to various diseases leading to higher mortality. Glucose level of this buffaloes was at the lower end of normal range may be credited to their extra burden of exercises for search of feed in the water beds and even not supplemented with extraneous feed. Also it may be possible due to that as all the samples were collected from majority of early lactating animals, where the glucose level generally remains lower in early part of lactation. The serum cholesterol level lower and again the cause of lactation may be given attribution to. The triglyceride level of these animals was higher than normal, which may be due to the utilization of reserve fat in the body during early part of lactation.

*Serum mineral status of the animals:* The serum calcium, phosphorus and magnesium status of these buffaloes is shown in Table 3.

The calcium status of these buffaloes was seen to be at par with the other buffaloes reported by various workers (Singh *et al.* 1979, Baruah *et al.* 2000, Gowda *et al.* 2001). In another study at NDRI, Karnal by Panda and Kaur (2005), the calcium level of Murrah buffaloes during periparturient period was within 8.28 to 9.66 mg/dl. Singh *et al.* (2011), reported the average serum Ca, P content of cows in mid-central table land zone of Odisha to be 6.91±0.13 mg/dl,

3.25±0.08 mg/dl respectively. Panda *et al.* (2015), reported the serum calcium and phosphorus levels of bovines in North Eastern Ghat region to be 6.69 and 4.35 mg/dl. Considering the particulars of serum calcium level of animals in Odisha, where maximum animals suffer from a low level of calcium from normal (9.7 to 12.4 mg/dl), the optimum concentration of calcium in the serum of Chilika buffaloes even without extraneous supplementation, may be attributed to the calcium in fodders along with the debris of Chilika bed.

Again the maintenance of optimum phosphorus level in the serum of Chilika buffaloes, even without supplementation and with a very low content in the fodders may be due to the intake of debris containing fish and shrimp remnants or even shells along with the submerged weed and grasses. There is a good number of shrimp farming in and around the lake, which is an alternate livelihood to the people of this region. There is also possibility that the water of Chilika Lake is rich in these mineral contents, to be further explored.

The serum magnesium level of Chilika buffaloes was at par with the other buffaloes as described earlier (Singh *et al.* 1979, Sharma *et al.* 2002, Pasha *et al.* 2012), even through the content of this mineral in the fodder was very low. The serum sodium and potassium level was comparatively higher than the other buffaloes of different regions as estimated by various works (Sharma *et al.* 2002, Pasha *et al.* 2012). This more than normal level in their serum may be credited to the higher sodium and potassium content in the grazed fodders as well as the intake of saline water at the time of grazing upon the submerged weed and grasses.

*Composition of the milk:* The average milk production of these buffaloes is 1.8 l/day with a range of 1.2 to 3.5 l/day with an average lactation yield of 440 to 514 l. The milk was collected from the selected Chilika buffaloes at the morning hour, at their usual time of milking. The fat, solid not fat (SNF) and total solid (TS) content of the milk was 8.92, 8.89 and 17.86% respectively. The crude protein was found at 5.49%, ash 0.89% and lactose 2.51% as recorded from the milk. Dash *et al.* (2010) also reported that the average fat % of the milk of these buffaloes was 8.5% which is higher than most of the Indian buffaloes. The SNF and TS content of the milk from these buffaloes were at par with the Murrah buffaloes (Peeva 2001). But the fat content of the Chilika buffaloes is higher than the Murrah buffaloes (Mallick 1991, Panda and Kaur 2007), hill buffaloes (Meena *et al.* 2007), Bhadwari buffaloes (Sachen *et al.* 2010). In some Chilika buffaloes the butter fat content was exceptionally high at a level of more than 10%.

The crude protein concentration of these buffaloes also does not vary with other buffaloes, but the lactose content was very low as compared with other buffaloes (BeiZhong *et al.* 2007, Khan *et al.* 2007). This lower lactose content of the milk may be attributed to higher serum sodium status of the animals.

Table 4. Fatty acid profile of fat from Chilika buffalo and crossbred cow milk

| Fatty acid   | Chilika buffalo milk |       | Cow milk           |       |
|--------------|----------------------|-------|--------------------|-------|
|              | Mean                 | SE    | Mean               | SE    |
| C8           | 0.69                 | 0.037 | –                  | –     |
| C10          | 0.05                 | 0.002 | 0.15               | 0.017 |
| C11          | –                    | –     | 0.06               | 0.004 |
| C12          | –                    | –     | 5.21               | 0.333 |
| C13:0        | 0.10                 | 0.004 | 1.02               | 0.119 |
| C14:0        | 15.87                | 0.730 | 6.05               | 0.626 |
| C15:0        | 2.88                 | 0.115 | 14.51              | 2.234 |
| C16:0        | 15.89                | 0.265 | 18.75              | 2.571 |
| C17:0        | 1.56                 | 0.057 | 0.69               | 0.083 |
| C18:0        | 15.46                | 1.176 | 20.74              | 1.351 |
| C20:0        | 0.30                 | 0.01  | 0.45               | 0.056 |
| C21:0        | 0.03                 | 0.003 | –                  | –     |
| C22:0        | 0.11                 | 0.007 | 0.10               | 0.01  |
| C23:0        | 0.01                 | 0.002 | 0.13               | 0.012 |
| C24:0        | 0.04                 | 0.005 | –                  | –     |
| “SFA         | 52.98 <sup>b</sup>   | 1.183 | 67.85 <sup>a</sup> | 1.307 |
| C14:1        | 1.13                 | 0.055 | 0.99               | 0.101 |
| C15:1        | 0.38                 | 0.016 | 0.65               | 0.075 |
| C16:1        | 1.18                 | 0.073 | 1.44               | 0.078 |
| C17:1        | 0.33                 | 0.018 | 5.78               | 0.799 |
| C18:1n9t     | 4.97                 | 0.435 | –                  | –     |
| C18:1n9c     | 32.10                | 1.24  | 18.22              | 1.430 |
| C20:1        | 0.08                 | 0.01  | 0.24               | 0.05  |
| C24:1        | 0.06                 | 0.01  | –                  | –     |
| “MUFA        | 40.21 <sup>a</sup>   | 1.09  | 27.31 <sup>b</sup> | 1.22  |
| C18:2n6t     | 0.52                 | 0.02  | 0.41               | 0.03  |
| C18:2n6c     | 1.96                 | 0.06  | 1.59               | 0.17  |
| C18:3n6      | 0.20                 | 0.01  | 0.34               | 0.05  |
| C20:2        | –                    | –     | 0.14               | 0.01  |
| C20:3n6      | 0.05                 | 0.004 | 0.03               | 0.002 |
| C20:4n6      | 0.01                 | 0.00  | 0.06               | 0.00  |
| “PUFA-n6     | 2.74                 | 0.07  | 2.56               | 0.10  |
| C18:3n3      | 2.98                 | 0.07  | 1.54               | 0.05  |
| C20:3n3      | 0.03                 | 0.003 | 0.10               | 0.01  |
| C20:5n3      | 0.10                 | 0.001 | 0.04               | 0.004 |
| C22:6n3      | 0.14                 | 0.01  | –                  | –     |
| “PUFA-n3     | 3.25 <sup>a</sup>    | 0.08  | 1.68 <sup>b</sup>  | 0.05  |
| “PUFA        | 5.98 <sup>a</sup>    | 0.12  | 4.23 <sup>b</sup>  | 0.15  |
| “SCFA & MCFA | 38.12 <sup>b</sup>   | 2.25  | 48.82 <sup>a</sup> | 1.24  |
| SFA:PUFA     | 9.05 <sup>a</sup>    | 0.38  | 16.23 <sup>b</sup> | 0.704 |
| n 3:n6       | 1.20 <sup>a</sup>    | 0.04  | 0.66 <sup>b</sup>  | 0.014 |

Means with different superscripts within a row differ significantly (P<0.01).

**Sample Information**  
**Analysis Date & Time** : 7/10/2012 12:25:44 PM  
**User Name** : Admin  
**Sample Type** : Standard  
**Sample Amount** : 1  
**Data Name** : G:\GC Database\FAME Standard 10.07.2012.gcd  
**Method Name** : G:\GC Database\DB-225 COLUMN offline.gcm  
**Chromatogram of G:\GC Database\FAME Standard 10.07.2012.gcd**

| Quantitative Results - Channel 1 |          |          |       | Method                            |                |
|----------------------------------|----------|----------|-------|-----------------------------------|----------------|
| ID#                              | Name     | Ret.Time | Area% |                                   |                |
| 1                                | C4       | 1.594    | 3.073 | <Analytical Line 1>               |                |
| 2                                | C6       | 2.731    | 4.040 | [Injection Port SPL1]             |                |
| 3                                | C8       | 3.862    | 4.768 | Injection Mode                    | : Split        |
| 4                                | C10      | 4.885    | 5.125 | Temperature                       | : 240.0 C      |
| 5                                | C11      | 5.353    | 2.450 | Carrier Gas                       | : H2           |
| 6                                | C12      | 5.804    | 5.110 | Pressure                          | : 394.9 kPa    |
| 7                                | C13:0    | 6.227    | 2.492 | Total Flow                        | : 31.2 mL/min  |
| 8                                | C14:0    | 6.635    | 5.033 | Column Flow                       | : 0.91 mL/min  |
| 9                                | C14:1    | 6.748    | 2.389 | Linear Velocity                   | : 59.3 cm/sec  |
| 10                               | C15:0    | 7.022    | 2.365 | Purge Flow                        | : 3.0 mL/min   |
| 11                               | C15:1    | 7.145    | 2.316 | Split Ratio                       | : 30.0         |
| 12                               | C16:0    | 7.457    | 7.305 | [Column Oven]                     |                |
| 13                               | C16:1    | 7.550    | 2.035 | Initial Temperature               | : 35.0 C       |
| 14                               | C17:0    | 7.956    | 2.149 | Equilibration Time                | : 3.0 min      |
| 15                               | C17:1    | 8.081    | 2.130 | =Column Oven Temperature Program= |                |
| 16                               | C18:0    | 8.587    | 4.263 | Total Program Time                | : 16.36 min    |
| 17                               | C18:1n9t | 0.000    | 0.000 | Rate(C/min)                       | Temperature(C) |
| 18                               | C18:1n9c | 8.672    | 6.422 | -----                             | 35.0           |
| 19                               | C18:2n6t | 8.873    | 1.867 | 1                                 | 25.0           |
| 20                               | C18:2n6c | 8.945    | 1.901 | 2                                 | 3.0            |
| 21                               | C18:3n6  | 9.079    | 1.698 | 3                                 | 8.0            |
| 22                               | C18:3n3  | 9.303    | 1.688 |                                   | 230.0          |
| 23                               | C20:0    | 10.231   | 3.910 | [Column Information]              |                |
| 24                               | C20:1    | 10.375   | 1.899 | Column Name                       | : DB-225       |
| 25                               | C20:2    | 10.736   | 1.701 | Film Thickness                    | : 0.10 um      |
| 26                               | C20:3n6  | 10.904   | 1.473 | Column Length                     | : 20.0 m       |
| 27                               | C20:3n3  | 10.985   | 1.484 | Inner Diameter                    | : 0.10 mm ID   |
| 28                               | C21:0    | 0.000    | 0.000 | Column Max Temp                   | : 240 C        |
| 29                               | C20:4n6  | 11.173   | 3.389 | [Detector Channel 1 FID1]         |                |
| 30                               | C20:5n3  | 11.438   | 1.370 | Temperature                       | : 240.0 C      |
| 31                               | C22:0    | 12.142   | 3.612 | Sampling Rate                     | : 40 msec      |
| 32                               | C22:1n9  | 12.287   | 1.770 | Stop Time                         | : 16.36 min    |
| 33                               | C22:2    | 12.643   | 1.582 | Makeup Gas                        | : He           |
| 34                               | C23:0    | 13.039   | 1.647 | Makeup Flow                       | : 30.0 mL/min  |
| 35                               | C22:6n3  | 13.446   | 0.999 | H2 Flow                           | : 40.0 mL/min  |
| 36                               | C24:0    | 13.983   | 3.038 | Air Flow                          | : 400.0 mL/min |
| 37                               | C24:1    | 14.169   | 1.505 |                                   |                |
|                                  |          |          |       |                                   | Hold Time(min) |
|                                  |          |          |       |                                   | 0.50           |
|                                  |          |          |       |                                   | 0.00           |
|                                  |          |          |       |                                   | 0.00           |
|                                  |          |          |       |                                   | 3.00           |

Fig. 1. Chromatogram showing the fatty acid profile of the standard.

**Fatty acid profile of milk:** The total saturated fatty acid content of the milk of this buffalo is given in Table 4. From the total saturated fatty acid (SFA) and mono unsaturated fatty acid (MUFA) content, the concentration of the short chain fatty acid (SCFA) and medium chain fatty acids (MCFA) in total was derived to be 38.12%. The poly unsaturated fatty acid (C18:2 to C22:6) content of the milk was 5.98%. Out of this total content of poly unsaturated fatty acids (PUFA), the concentrations of n-6 PUFA and n-3 PUFA were 2.74 and 3.25 g/100 g of milk fat respectively. The ratio of n-3 to n-6 PUFA was calculated to be 1.19 and the ratio of SFA and PUFA was 9.05. The fatty acids with highest concentration in total milk fat were C16:0, C18:0 and C14:0 amongst the saturated fatty acids, C18:1n9c among the MUFA and C18:3 n3 followed by C18:2 n6

among the PUFA. At the same time from the fatty acid analysis of local cow's milk fat, it was found that, the content of SFA, MUFA and PUFA were 67.85g, 27.31g and 4.23g per 100g of milk fat respectively. The concentration of SCFA and MCFA in total was 48.82% of the milk fat. The fatty acids of highest concentration in the cow milk fat were C18:0 followed by C16:0 and C15:0 among the SFA, C18:1 n9 among the MUFA and C18:2 n6 followed by C18:3 n3 among the PUFA. The ratio of n-3 to n-6 PUFA was calculated to be 0.66 and the ratio of SFA and PUFA was 16.23. Achaya and Banerjee (1946) reported the content of Palmitic acid among SFA and Oleic acid among UFA to be 31.9% and 16.8% respectively in Indian buffaloes' milk fat. Talpur *et al.* (2007), evaluated the fatty acid composition of two Pakistani dairy water buffalo breeds named Kundi

**Sample Information**

**Analysis Date & Time** : 8/10/2012 8:56:35 PM  
**User Name** : Admin  
**Sample Type** : Unknown  
**Sample Amount** : 1  
**Data Name** : G:\GC Database\CB MILK 21, 10.8.12.gcd  
**Method Name** : G:\GC Database\DB-225 COLUMN offline.gcm  
**Report Name** : E:\GCsolution\System\DEFAULT.gcr  
**Chromatogram of G:\GC Database\CB MILK 21, 10.8.12.gcd**

**Quantitative Results - Channel 1**

| ID# | Name     | Ret.Time | Area%  |
|-----|----------|----------|--------|
| 1   | C4       | 0.000    | 0.000  |
| 2   | C6       | 0.000    | 0.000  |
| 3   | C8       | 3.856    | 0.907  |
| 4   | C10      | 0.000    | 0.000  |
| 5   | C11      | 0.000    | 0.000  |
| 6   | C12      | 0.000    | 0.000  |
| 7   | C13:0    | 6.205    | 0.139  |
| 8   | C14:0    | 6.626    | 20.011 |
| 9   | C14:1    | 6.739    | 1.394  |
| 10  | C15:0    | 7.012    | 3.463  |
| 11  | C15:1    | 7.227    | 0.461  |
| 12  | C16:0    | 0.000    | 0.000  |
| 13  | C16:1    | 7.538    | 1.337  |
| 14  | C17:0    | 7.941    | 1.867  |
| 15  | C17:1    | 8.063    | 0.299  |
| 16  | C18:0    | 8.580    | 28.691 |
| 17  | C18:1n9t | 0.000    | 0.000  |
| 18  | C18:1n9c | 8.674    | 34.446 |
| 19  | C18:2n6t | 8.818    | 0.648  |
| 20  | C18:2n6c | 8.925    | 1.897  |
| 21  | C18:3n6  | 9.014    | 0.195  |
| 22  | C18:3n3  | 9.281    | 2.931  |
| 23  | C20:0    | 10.137   | 0.395  |
| 24  | C20:1    | 10.348   | 0.171  |
| 25  | C20:2    | 0.000    | 0.000  |
| 26  | C20:3n6  | 0.000    | 0.000  |
| 27  | C20:3n3  | 0.000    | 0.000  |
| 28  | C21:0    | 0.000    | 0.000  |
| 29  | C20:4n6  | 0.000    | 0.000  |
| 30  | C20:5n3  | 11.311   | 0.152  |
| 31  | C22:0    | 12.049   | 0.193  |
| 32  | C22:1n9  | 0.000    | 0.000  |
| 33  | C22:2    | 0.000    | 0.000  |
| 34  | C23:0    | 0.000    | 0.000  |
| 35  | C22:6n3  | 13.309   | 0.161  |
| 36  | C24:0    | 0.000    | 0.000  |
| 37  | C24:1    | 0.000    | 0.000  |

Fig. 2. Chromatogram showing the fatty acid profile of Chilika buffaloes.

and Nilli Ravi and found the amount of SFA, MUFA and PUFA per 100g of milk fat to be 66.96g, 69.09g; 27.62g, 25.20 g and 2.76g, 2.77g respectively.

Thus as per the investigation of Varricchio *et al.* (2007), on fatty acid composition of milk fat from 4 different Mediterranean buffalo herds, it was revealed that, SFA, MUFA and PUFA contents were 65.5%, 27% and 4.5% respectively. The content of linolenic acid (C18:3) varied from 0.96% to 1.38%. Another study on fatty acid profile

of Brazilian Murrah buffaloes (Fernandes *et al.* 2007) showed that the total SFA MUFA and PUFA of the milk varied from 55.6% to 62.6%, 27.3% to 36.5% and 3.1% to 3.9% of the total milk fat. Mihaylova and Peeva (2007) reported that SFA, MUFA and PUFA in Bulgarian Murrah buffalo milk were 72.15%, 24.7% and 3.15% respectively.

From the above literature it was found that SFA, MUFA and PUFA varied from 55.6% to 72.15%, 24.7% to 36.5% and 2.76% to 4.5% respectively in the milk fat of different

**Sample Information**

**Analysis Date & Time** : 7/10/2012 5:05:07 PM  
**User Name** : Admin  
**Sample Type** : Unknown  
**Sample Amount** : 1  
**Data Name** : G:\GC Database\Cattle milk 10.07.12.gcd  
**Method Name** : G:\GC Database\DB-225 COLUMN offline.gcm  
**Report Name** : E:\GCsolution\System\DEFAULT.gcr  
**Chromatogram of G:\GC Database\Cattle milk 10.07.12.gcd**

**Quantitative Results - Channel 1**

| ID# | Name     | Ret.Time | Area%  |
|-----|----------|----------|--------|
| 1   | C4       | 0.000    | 0.000  |
| 2   | C6       | 0.000    | 0.000  |
| 3   | C8       | 0.000    | 0.000  |
| 4   | C10      | 5.036    | 0.107  |
| 5   | C11      | 5.194    | 0.062  |
| 6   | C12      | 5.796    | 6.763  |
| 7   | C13:0    | 6.219    | 1.366  |
| 8   | C14:0    | 6.626    | 4.738  |
| 9   | C14:1    | 6.739    | 0.821  |
| 10  | C15:0    | 7.012    | 20.710 |
| 11  | C15:1    | 7.135    | 0.856  |
| 12  | C16:0    | 7.445    | 14.178 |
| 13  | C16:1    | 7.538    | 1.227  |
| 14  | C17:0    | 7.941    | 0.496  |
| 15  | C17:1    | 0.000    | 0.000  |
| 16  | C18:0    | 8.568    | 30.106 |
| 17  | C18:1n9t | 0.000    | 0.000  |
| 18  | C18:1n9c | 8.711    | 13.801 |
| 19  | C18:2n6t | 8.854    | 0.289  |
| 20  | C18:2n6c | 8.941    | 1.312  |
| 21  | C18:3n6  | 9.060    | 0.300  |
| 22  | C18:3n3  | 9.281    | 1.541  |
| 23  | C20:0    | 10.201   | 0.419  |
| 24  | C20:1    | 10.347   | 0.177  |
| 25  | C20:2    | 10.725   | 0.042  |
| 26  | C20:3n6  | 10.878   | 0.140  |
| 27  | C20:3n3  | 10.964   | 0.070  |
| 28  | C21:0    | 0.000    | 0.000  |
| 29  | C20:4n6  | 11.254   | 0.084  |
| 30  | C20:5n3  | 11.359   | 0.040  |
| 31  | C22:0    | 12.114   | 0.153  |
| 32  | C22:1n9  | 0.000    | 0.000  |
| 33  | C22:2    | 0.000    | 0.000  |
| 34  | C23:0    | 13.012   | 0.142  |
| 35  | C22:6n3  | 0.000    | 0.000  |
| 36  | C24:0    | 0.000    | 0.000  |
| 37  | C24:1    | 0.000    | 0.000  |

Fig. 3. Chromatogram showing the fatty acid profile of a cow.

buffaloes throughout the world. As far as the Chilika buffalo is concerned, the fatty acid profile of the milk fat is quite beneficial for human beings, as it contains higher percentage of PUFA and lower percentage of SFA. Also among the PUFA, when the omega 3 or n:3 fatty acid content of the milk fat varies between 0.33% and 1.38% in other buffaloes of world, there the n 3 fatty acid content from Chilika buffalo milk fat was 3.25%, which is much higher than any other buffalo milk fat in the world and also from the cow milk fat as found during the experiment. Though from the

experiment it was revealed that the SCFA and MCFA content of milk fat is higher in the fat derived from the cow's milk, but in other aspects the fatty acid profile of Chilika buffalo milk fat seems to be better (Figs 1–3). The n:3 fatty acid content of this milk is even comparable to that of some freshwater fish and hence much better as a milk for today's health cautious world. The ratio of n:3 to n:6 in this milk was 1.19, which is also a good index to judge the quality of milk fat content of the milk.

There is evidence that fish oil may prevent CHD at

relatively low intakes due to presence of more n:3 fatty acids. Now a days the n:3 fatty acid content of egg had been tried to increase by many workers by inducing fish oil in feed of layers for preventing chronic heart disease (CHD) in human beings. Similarly the milk fatty acid profile could be altered substantially by manipulating the diet of the animal (Kennell 1996, Kennelly and Khorasani 1992). But these Chilika buffaloes, which only thrive on the brackish water weeds, algae and grasses grown on the banks of the lake, showed a considerable increase in PUFA content of the milk along with n:3 fatty acid content, lessening the total concentration of SFA as compared to other bovine milk. The natural habitat and feeding habit might be influencing the content and quality of milk fat, which need thorough exploration.

From the result of the present study it is concluded that the Chilika buffalo, which is unique to its habitat and feeding pattern has very high quality milk fatty acid profiles among the buffaloes of other regions of India and abroad, being rich in n:3 fatty acids. Even without supplementation of concentrate and mineral mixture, these buffaloes have optimum serum level of major and minor minerals except Na, K and Fe which is higher than the normal values. Further research is required to explore the availability of the biomass and exploit the potentiality of these buffaloes for its rich n:3 fatty acid content in the milk to be utilized by the today's health conscious society.

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