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Analysis of Vitamin Content in the Milk of Dairy Animals Vs. Humans

KEYWORDS: Water soluble vitamin, Fat soluble vitamin, Antioxidant, cow, goat, camel, human milk

Dr. Sarah Afaq

Assistant Professor, Department of Clinical Biochemistry, College of Medicine, King Khalid University, Abha, Kingdom of Saudi Arabia

ABSTRACT *For centuries milk has been a good source of various nutrients including vitamin content. The present study was done to evaluate and compare the content of eight important vitamins, water and fat soluble vitamins, in the milk of diary animals like cow, goat and camel along with the human. Camel milk has the highest vitamin B3 and C content, while human milk has highest vitamin A and E content. Cow milk has highest vitamin D,B1,B6 and B12 content, while goat milk has highest vitamin B2 and B3.Cow and goat milk have much similar vitamin profile, but human milk is best in overall vitamin content.*

Introduction:
The content of vitamins in milk is important for several reasons, as it is well known that vitamins play prophylactic role in cancer, autoimmune diseases, heart diseases etc .Vitamin content is an important quality aspect of milk for human consumption, since milk is generally viewed as nutritious food and should contain sufficient amounts of naturally occurring vitamins. Vitamins in milk can play an important part in helping to meet daily nutritional requirements. Many of us take vitamin supplements to help round out our diets, but it's well-recognized that vitamins act synergistically, and offer more benefits when obtained through food sources. Different studies have been done on diary animals like goat, cow and camel as well, regarding their nutritional and therapeutic values including mineral and vitamin content(1,2,3).Studies have been carried out as well on human milk containing its composition and nutritional values for the infant(4).Though there are different sources of milk available ,however sufficient information regarding their vitamin content, especially, comparative study between the different types of milk is not available much.

The present study was carried out to compare the vitamin content: water soluble and fat soluble vitamins, in the milk from different sources commonly available like goat, cow, camel (diary animals) and humans as well.

Materials and methods:
Camel (Camelus dromedaries) milk (20 samples) was obtained from farms at Agmark, areas around Aligarh, India. Human milk (20 samples) was collected from healthy volunteer women at Jawaharlal Nehru Medical College, Aligarh, India. Cow (20 samples) were taken from

Local farms at Swastik dairy farm and goat's milk (20 samples) from local dairy farms around Aligarh,India. The samples were kept refrigerated at 4°C and transported to our laboratory within 24 hours. Prior to refrigeration, all the samples for vitamin C determination were stabilized with 10% metaphosphoric acid. Upon arrival, the milk samples were stored at -20°C until analysis.

Analytical methods:
Both retinol and α-tocopherol were determined on the same sample extract on the milk specimen after saponification, using high-performance liquid chromatography (HPLC) as described by Chavez-Servin, et, al.(5) Prior to testing, the samples were briefly immersed in warm water (40°C) to thaw them, then mixed using a vortex to provide a homogenous sample. One ml of the sample was transferred to a centrifuge tube and 3 ml of ethanol were added. The samples were mechanically shaken and 1 ml of hexane was added, and then shaken for another minute. After resting the samples for 5 minutes, 3 ml of saturated NaCl was added to aid separation. The mixture was shaken by inversion. The samples were centrifuged for 5 minutes at 3000 rpm at room temperature. The hexane phase was recovered and directly filtered through a .22 um nylon filter and collected in a 1 ml amber glass vial. 20 ul was injected into the HPLC system.

For vitamin D analysis, the samples were saponified to hydrolyze triacylglycerols into fatty acids and glycerol, extract vitaminD2 and vitaminD3, and collect both vitamin D2 and D3 as a single peak by using preparative-scale, normal phase HPLC, and separate vitamin D2 and D3 by using analytic reversed-phase chromatography with DA detection. Hexane was used for extraction solvent and internal standards used (6).

Vitamin B1, B2, B3, B6 and B12 were analyzed using HPLC as described by Albala-Hurtado, et al. (7). The samples were protected from light by wrapping tubes and flasks with aluminum foil and preparing the samples in a darkened room. 10.5 grams of sample were weighed into a 50 ml centrifuge tube. 1 g of TCA solid and a magnetic stirring bar were added. The mixture was stirred for 10 min over a magnetic stirring plate, then centrifuged for 10 minutes at 1250 g to separate the two phases. 3 ml of 4% TCA was added to the solid residue obtained and mixed for 10 minutes and then centrifuged. The solid-phase was discarded. The two acid extracts were combined in a 10 ml volumetric flask and the volume was filled with 4% TCA. Acid extracts were filtered through a .45um filter and injected into the HPLC system. Vitamin C content was analyzed using HPLC described by Romeu-Nadal, et al. (8). The samples were protected from light by wrapping tubes and flasks with aluminum foil and preparing the samples in a darkened room. 300 ul of milk mixed with 300ul of 0.56% meta-phosphoric acid solution were added to the same centrifuge and filtration tube, which was shaken for 30 seconds and centrifuged at 10°C (10 minutes, 3000 X g). Ascorbic acid was identified by comparing the retention time of the sample peak with that of the ascorbic standard at 254 nm. Quantification was carried out using external standardization.

Results and Discussion:
The Table shows the average values of water soluble vitamins (B1, B2, B3, B6, B12 and C) and fat soluble vitamins (A, D and E) in Diary animals (like cow, goat and camel) along with humans.

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The coefficient of variation (CV) calculated on 20 double determinations for camel milk samples were found to be ±4.45% for retinol (VitA) , ±4.05% for α-tocopherol (Vit E)and±3.51% for cholecalciferol (Vit.D3). For Thiamin (Vit B1),riboflavin (Vit B2),niacin(Vit B3),pyridoxine (Vit B6),cobalamine (Vit B12) and ascorbic acid (Vit C),CV was found to be ±2.56%,± 2.83%, ±1.35%,±2.12%,±2.76%and ± 1.22% (10 double determinations) respectively.

While coefficient of variation (CV) calculated for samples of cow were found to be ±4.03% for retinol (VitA), ±5.05% for α-tocopherol (Vit E) and±1.51% for cholecalciferol (Vit.D3). For Thiamin (Vit B1),riboflavin(Vit B2),niacin(Vit B3),pyridoxine(Vit B6),cobalamine (Vit B12) and ascorbic acid(Vit C),CV was found to be ±1.50%,± 2.80%, ±3.18%,±1.56%,±1.40%and ± 4.22% respectively.

The coefficient of variation for the available samples of goat milk were calculated as be ±2.45% for retinol(VitA) , ±3.05% for α-tocopherol(Vit E)and±2.51% for cholecalciferol(Vit.D3). For Thiamin(Vit B1),riboflavin(Vit B2),niacin(Vit B3),pyridoxine(Vit B6),cobalamine(Vit B12) and ascorbic acid(Vit C),CV was found to be ±3.56%,± 1.33%, ±1.21%,±2.34%,±3.76%and ± 4.22% respectively.

However, the human samples showed a high variability, as would be expected in human milk Therefore ; median values, minimum and maximum are reported. The range for vitamin A values was 948 IU/L in human milk to a high of 2840 IU/L. For vitamin E, the lowest level was 440 mg/L to a high level of 1318.5 mg/L. The range for Vitamin D values was found to be 10.5mg/L in human milk to a high of 34mg/L.The Vitamin C levels ranged from 32 mg/L to a high of 53 mg/L. The range for vitamin B1 was 0.032 mg/L to 0.676 mg/L. The range for vitamin B2 was 0.156 mg/L to 1.015 mg/L. The range for vitamin B3 was 1.25 mg/L to 7.10 mg/L.VitaminB6 showed a minimum of 0.045 mg/L to a maximum of 0.15mg/L. However, VitaminB12 showed a minimum of 0.0006 mg/L to a high of 0.0001 mg/L. The above results correlate with the other findings done separately but not comparatively (1, 2, 3, 4).

Henceforth, it was found that camel milk has the highest vitamin B3 and C content, while human milk has highest vitamin A and E content. Cow milk has highest vitaminD,B1,B6 and B12,while goat milk has highest vitamin B2 and B3.Cow and goat milk have much similar vitamin profile, but human milk is best in overall vitamin content .Camel milk was found to be one of the best source of vitamin C, an important antioxidant .

Table 1
Average vitamin content of Camel, goat, cow and human milk.

Vitamin	Cow	Goat	Human	Camel
Vitamin A	916.57	1205.87	1898.0	316.63
Vitamin D(cholecalciferol)	33	23.7	22.0	0.016
VitaminB1(Thiamine)	0.44	0.40	0.16	0.12
VitaminB2(Riboflavin)	1.75	1.84	0.36	1.24
VitaminB3(Niacin)	0.94	1.87	2.1	5.2
Vitamin B 6(Pyridoxine)	0.64	0.07	0.10	0.54
Vitamin B 12(Cobalamine)	0.0043	0.0006	0.0003	0.0040
VitaminC(Ascorbic Acid)	10.0	11.0	35.0	46.0
VitaminE(Alpha-tocopherol)	7.56	9.0	879	7.45

Vitamin A is expressed in International Units/liter; all other vitamins as mg/liter. Coefficient of Variation (CV) of vitamins for goat, cow and camel was calculated (given in results and discussion). Human samples showed a high variability: median values, minimum and maximum are reported (result and discussion)

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