# VARIABILITY OF VITAMIN E CONCENTRATION IN CAMEL PLASMA

# Bernard Faye<sup>1</sup> and Rabiha Seboussi<sup>2</sup>

<sup>1</sup>Département Environnement et Sociétés, CIRAD, Campus International de Baillarguet, UR 18, TA/C Dir B, 34398 Montpellier Cedex, France <sup>2</sup>United Arab Emirates University, PO Box No. 16641, Al-Ain, United Arab Emirates

### **ABSTRACT**

In the present study, variability of plasma  $\alpha$ -tocopherol was reported in different groups of camels (non-lactating and non-pregnant, pregnant, lactating and their new-born, young, selenium intoxicated young camels). The mean values in the different groups were  $1.13\pm0.61~\mu g/ml$  (non-lactating and non pregnant),  $1.12\pm0.81~\mu g/ml$  (pregnant),  $1.20\pm0.80~\mu g/ml$  (lactating),  $0.82\pm1.06~\mu g/ml$  (new-born),  $0.56\pm0.22~\mu g/ml$  (young) and  $0.68\pm0.36~\mu g/ml$  (intoxicated young). There was no significant change in the time. Those values appeared lower than in plasma of other ruminants, but the effect of vitamin E supplementation was not known in camel.

Key words: Camel, lactating, plasma, pregnancy, selenium intoxication, vitamin E

Vitamin E (α-tocopherol) has long been recognised as a natural biological antioxidant. Vitamin E appears to be the first line of defense against peroxidation which can severely damage the cell and tissues. Vitamin E is also an essential component in the reproduction processes and performance of farm animals and acts in synergy with the selenium (Se), especially in order to prevent white muscle disease (WMD) due to a severe deficiency. In camel, recent studies showed the effect of selenium supplementation to improve the Se status of the animals (Seboussi et al, 2008 and 2009), especially in the young camel calves which are highly susceptible to WMD, a very common disease in the Gulf countries (Al-Qarawi et al, 2001; El-Khouly et al, 2001; Seboussi et al, 2004). However, neither the interaction with vitamin E, nor the variability of vitamin E status according to the age and physiological stage, were studied in camel.

The present paper aims to study vitamin E variability in camel blood according to the different status of the animals (new born, young, non-lactating and non pregnant females, pregnant females, lactating females and Se intoxicated animals).

### Materials and Methods

### Animals and feeding

The study was carried out at Al-Foha experimental farm belonged to the Food and

Agriculture College of the UAE University. It included 4 groups of animals:

- I. Group NLNP: It included 12 non-lactating and non-pregnant 6 to 13 years old female camels of local breed. The approximate mean weights were 430 kg. The animals were monitored for 225 days. For the whole trial, the animals were fed individually with approximatiely 6 kg of Rhodes grass (*Chloris gayana*) hay and 2 kg of concentrates with known vitamin E content. The refusals were daily weighed and the quantity of grass adjusted to the mean intake. There was no concentrate refusal.
- II. Group PFC: It included 12 pregnant females (more than 7 years old) and their new born camel calves after parturition. All the females were pregnant at the beginning of the trial. The trial started approximately 3 months before the parturition and stopped 3 months after the parturition, i.e. 195 days. They received approximately 5.5 kg of Rhodes grass (*Chloris gayana*) hay and 2 kg of concentrates with known vitamin E content.
- III. Group YFC: It included 8 young female camels less than 2 years old. For the whole trial, 70 days length, the animals were fed individually with a similar diet composed of 3 kg DM of Rhodes grass (*Chloris gayana*) hay and 2 kg of concentrates (pellets) containing 10% protein and with known vitamin E content.

SEND REPRINT REQUEST TO BERNARD FAYE email: faye@cirad.fr

IV. Group ISe: It included 12 young 2-yr old males and females, which were Se intoxicated. They received similar diet than YC group, but received 12 mg Se daily under selenite form, which was considered as toxic dose for camel (Faye and Seboussi, 2008).

All, the animals were treated for external and internal parasites using ivermectin (Ivomec N.D.) and were in good health during the whole experiment, or at least at the beginning of the trial for the group ISe. The animals were provided water *ad libitum*.

## Blood and feed sampling

The blood sampling was carried out in the morning before feed distribution. In group NLNP, blood sampling was performed every week (20 samplings). In group PFC, the blood sampling was carried out twice a month in the morning before feed distribution in adult (17 samplings) and young camels (7 samplings). A camel calf's blood sample was taken at the moment of delivery prior to colostrum feeding to evaluate the vitamin E status at birth. For group YFC, a weekly blood sampling was achieved (10 samplings). For group ISe, 7 blood sampling was collected every 2 weeks before food distribution and selenium supplementation.

In all the animals, blood was collected from the jugular vein in both heparinised (H) and nonheparinised tubes (NH). Two tubes of H and NH were transferred to Alqatara veterinary laboratory (agriculture and municipalities department – AbuDhabi) for haematology and biochemistry analysis. From the H tube, 2 ml of whole blood was collected, centrifuged, plasma was removed and stored at -80°C until vitamin E analysis.

The elements of the basal diet were sampled at the beginning, the mid and at the end of the trial; dried, ground and stored for vitamin E analysis.

### Laboratory analysis

Vitamin E quantification in plasma was performed by High-Performance Liquid Chromatography (HPLC) system. For vitamin E determination, 1-ml samples of plasma were extracted using one time the sample volume of ethanol and 2 times the sample volume of hexane. Vitamin E was measured in the extracts as  $\alpha$ -tocopherol by HPLC using a 3.9 x 150-mm silica column and UV detection at 292 nm. The mobile phase was hexane/chloroform (85/15) with isocratic elution (Hatam and Kayden, 1979). Selenium was determined in serum from the group ISe. The Se determination in different samples was done by Inductively Coupled argon Plasma – Atomic

Emission Spectrometer (ICP AES), Varian Vista MPX – CCD Simultaneous.

In plasma, the other mineral parameters studied were trace elements, i.e. copper, zinc and iron. The following biochemical parameters were determined in the serum: glucose, creatinine, total proteins, albumin, bilirubin, creatine phosphokinase (CPK), alkaline phosphatase (AP), alanine aminotransferase (ALT), aspartate aminotransferase (AST), lactate dehydrogenase (LDH) and gamma glutamyl transferase (GGT). The haematological parameters analysed in routine included haematocrit, WBC, haemoglobin, and blood formula (WBC/N, WBC/L, WBC/M, and WBC/E for neutrophil, lymphocyte, monocyte and eosinophil percentages, respectively).

### Statistical Analysis

Descriptive analyses (mean and standard deviation) were used to give raw results. Variance analysis on repeated measures was carried out using the R software. Previously, normality of the distribution was tested by the Skewness and Kurtosis test (test W). Interactions between other elements (minerals and biochemical parameters) and between mother and camel calf parameters were tested by the correlation of Pearson. Significant level at P < 0.05 was retained.

#### Results

The vitamin E content was 5.5  $\mu g/g$  in the Rhodes grass and 0.96  $\mu g/g$  in concentrate, and did not change all along the duration of the trials. The diets provided 35  $\mu g$  per day on average in group NLNP, 32  $\mu g/day$  in group PFC, and 18.4  $\mu g/day$  in groups YCF and ISe. It was the only source of vitamin E for the animals.

Vitamin E concentration in camel plasma varied between 0.002 and 4.86  $\mu$ g/ml with a mean of 0.976  $\pm$  0.672  $\mu$ g/ml. There was a significant difference

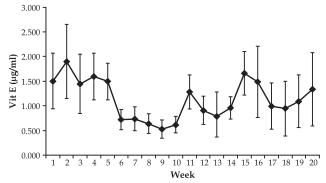


Fig 1. Weekly change in vitamin E content in serum in non-lactating, non-pregnant adult female camel (mean and S.D in  $\mu$ g/ml).

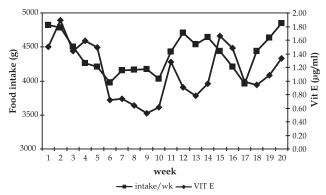


Fig 2. Weekly change in vitamin E concentration in plasma (in µg/ml) (♠) and food intake (in kg DM) (■) all along the trial. Each point for food intake is the mean of the daily intake in the week before blood sampling.

(P <0.001) between the adult animals from the first 2 groups (1.13  $\pm$  0.69  $\mu$ g/ml), the newborn animals (0.75  $\pm$  0.89  $\mu$ g/ml) and the 2-yrs young camels (0.62  $\pm$  0.30  $\mu$ g/ml).

# Change of vitamin E in non-lactating, non pregnant camels

The vitamin E ( $\alpha$ -tocopherol) level in plasma was  $1.13 \pm 0.61 \, \mu g/ml$  with a range of 0.27-3.09. The values from weeks 6 to 14 (except week 11) were significantly lower than other weeks (Fig 1). The maximum mean value observed was at week 2 (1.92  $\mu g/ml$ ). The variability of vitamin E content in plasma is linked to total food intake (Fig 2). The correlation between the total food intake, measured as the mean of the daily food intake during the week before blood sampling, and vitamin E concentration in plasma was significant (r = 0.46; P<0.05).

# Change of vitamin E in pregnant and lactating camel and their newborns

The average vitamin E content was  $1.16 \pm 0.81$  µg/ml for the entire experiment with no difference between the pre-calving ( $1.12 \pm 0.81$  µg/ml) and post-calving period ( $1.20 \pm 0.80$  µg/ml). A slight non-significant decrease was observed at the peripartum time (Fig 3).

In camel calf, vitamin E level was lower than in the mother, and varied from 0.002 to 4.67  $\mu g/ml$  with a mean of  $0.82 \pm 1.06 \, \mu g/ml$ . The plasma vitamin E was significantly higher (P < 0.05) at the second month of lactation. No correlation was observed between vitamin E concentration in she-camel plasma and its newborn plasma.

### Change of vitamin E in young camel

In a 2 year-old, young camel the vitamin E concentration in plasma was quite lower than in

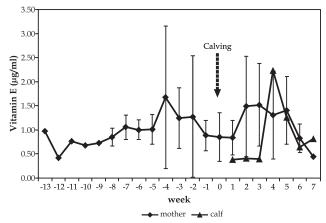


Fig 3. Change in vitamin E plasma concentration in she-camel at the end of gestation and at the beginning of the lactation of she-camel ("mother") and in camel calf (in μg/ml).

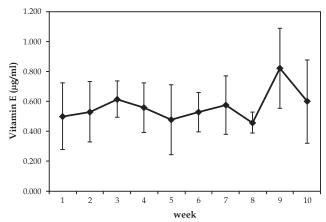


Fig 4. Weekly change in vitamin E concentration ( $\mu g/ml$ ) in young camel.

adult with value of  $0.56 \pm 0.22 \,\mu\text{g/ml}$ . There was no significant variation in the time (Fig 4).

### Change in selenium intoxicated camel

The plasma vitamin E in Se intoxicated camels was on average 0.68  $\pm$  0.36  $\mu g/ml$  and varied between 0.20 and 1.56  $\mu g/$  ml (n=69). There was a significant time difference (Fig 5) with the highest values observed on the second month of Se supplementation and the lowest at the end of the trial. After one month, the first sign of chronic selenosis appeared. A tendency to the decrease of vitamin E in intoxicated animals with clinical signs was observed but no significant correlation was reported with the serum Se concentration.

### Correlation with other blood parameters

By taking in account all the groups of camels, 4 parameters of the blood components were correlated to the vitamin E concentration, i.e. PCV, haemoglobin,

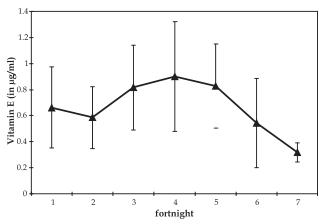


Fig 5. Change in vitamin E ( $\mu$ g/ml) concentration in young camels intoxicated with 12 mg/day Se under sodium selenite.

lymphocytes and eosinophils (Table 1). Among the biochemical parameters, glucose, creatinine, albumin and iron were positively correlated while the lactate dehydrogenase was negatively correlated.

### Discussion

The vitamin E concentration in camel plasma in our results was quite similar to those described in the literature; for example 0.3 to 1.65 µg/ml in young camels from Sudan (Barri and Al-Sultan, 2007). Similar results were reported by Al-Senaidy (1996) and Mousa et al (2006). In cow, the normal vitamin E concentration was reported to be above 2 μg/ml (Maas et al, 1990). In cows, values below 2.5 µg/ml were considered by different authors as a deficient status in vitamin E (Pehrson and Hakkarainen, 1986; Braun et al, 1991; Radostits et al, 1995). According to different diets, the plasma concentration of vitamin E in dairy cow receiving 200 g of vitamin-mineral mixture containing 1300 IU/kg of vitamin E, varied from 3.3 to 5.14 μg/ ml (Calderon et al, 2007a). According to LeBlanc

**Table 1.** Significant correlation coefficients and their p value between vitamin E concentration in blood and some blood parameters.

Parameters	Vit E	p value
PCV	0.157	0.000
Haemoglobin	0.158	0.000
Lymphocyte	-0.137	0.001
Eosinophil	0.161	0.000
Glucose	0.090	0.032
Creatinine	0.125	0.003
Albumin	0.195	< 0.0001
LDH	-0.088	0.036
Iron	0.093	0.026

*et al*, (2004), the optimum plasma concentration of α-tocopherol for mammary cell immune function was 3.5 to  $4.0\,\mu\text{g/ml}$ . Thus, the vitamin E concentration in camel plasma from our study appeared quite lower compared to cow's plasma (Nozière *et al*, 2004).

In camel, the vitamin E was predominating in the plasma and in the hump i.e. according to 50 mg Al-Senaidy (1996). On apparent deficient camels, an average of 9  $\pm$  0.19 to 2.01  $\pm$  0.18  $\mu g/ml$  was reported by Mohamed (2004).

The lower level of vitamin E in plasma of newborn animals compared to the mother was already observed in dairy cow, as well as the decrease of vitamin E concentration in blood at the calving period (Calderon *et al*, 2007b). The amplitude of this decrease was approximately of 50% from 3 weeks before calving up to calving time both for cow (Calderon *et al*, 2007b) which was similar to results in camel.

The absorption of vitamin E ( $\alpha$ -tocopherol), a fat-soluble vitamin, decreases when the diet is rich in polyunsaturated fatty acids. The camel calf was fed with milk containing similar content of vitamin E than in cow's milk (Farah, 1992). The camel milk contains high proportion of polyunsaturated fatty acids as compared to cow (Konuspayeva *et al*, 2008). This peculiarity of camel milk could stress its low efficiency to maintain high level of vitamin E intake for the camel calf.

In most of the studies on camel, there was no correlation between selenium and vitamin E plasma concentration (Seboussi et al, 2008 and 2009). However, a high level of Se seemed to depress the vitamin E level in plasma as it was recently observed in horses affected by selenosis (Crain, 2007). Selenium and vitamin E are both antioxidants, both protecting the membranes from oxidative damage. Due to this shared duty, there is a relationship between the compounds, in which one can substitute for the other in a very small way. For instance, more Se is needed when an animal's vitamin E concentrations are low. The sparing effect is an extension of this idea of substitution. In lambs, sodium selenite administration resulted in decreased liver vitamin E concentration (Tiwary et al, 2006). The vitamin E concentration in serum appeared at a lower level in camel as compared to other ruminants.

### Acknowledgements

The authors are very grateful to Mr Jumaa Said Hareb for allowing to carryout the analysis at the Al Qattara Veterinary Laboratory and Al Salamat Experimental Station belonging to the Agriculture and Animal Resources Department at Al-Ain, and to the Camel Research Centre of H.H. Sheik Khalifa Ben Zayed Al Nehyan which supported us to carrying out the samples preparation and storage under the best conditions. We thank the French Embassy in Abu Dhabi and Mr Ali Gomaa Al Katby, Dr Jawad Almasri, Dr Tarek Mostafa, Dr Hossam Habib, Eng Omer El-nour, Eng Abdelaziz Awaga, Mr Ramadan El Deeb, Mr Youssef Hassan for their technical support in the experiment.

#### References

- Al-Qarawi A, Abbas A, Haroun B, Mahmoud EM and Al-Hawas MA (2001). Clinicopathological investigation of Selenium responsive myopathy in young adult camels. Journal of Camel Practice and Research 8:23-27.
- Al-Senaidy AM (1996). Tocopherols in camel's plasma and tissues. International Journal of Nutrition Research 66: 210-216.
- Barri MES and Al-Sultan SI (2007). Studies on selenium and vitamin E status of young Megaheem dromedary camels at Al-Ahsa province. Journal of Camel Practice and Research 14:51-53.
- Braun U, Forrer R, Furer W and Lutz H (1991). Selenium and vitamin E in blood sera of cows from farms with increased incidence of disease. Veterinary Record 128: 543.
- Calderon F, Chauveau-Duriot B, Pradel P, Martin B, Graulet B, Doreau M and Nozière P (2007a). Variations in Carotenoids, Vitamins A and E, and Color in Cow's Plasma and Milk Following a Shift from Hay Diet to Diets Containing Increasing Levels of Carotenoids and Vitamin E. Journal of Dairy Sciences 90:5651-5664.
- Calderon F, Chauveau-Duriot B, Martin B, Graulet B, Doreau M and Noziere P (2007b). Variations in Carotenoids, Vitamins A and E, and Colour in Cow are Plasma and Milk during Late Pregnancy and the First Three Months of Lactation. Journal of Dairy Sciences 90:2335-2346.
- Crain SR (2007). Drought Related Nutrition Problems Threaten Midwest Horses. http://www.freshare.net/ article/drought\_related\_nutrition\_problems\_threaten\_ midwest\_ horses/
- El Khouly AA, Abbas TA and Moustafa T (2001). Myocardial dystrophy in camel calves in the United Arab Emirates (field cases). Emirates Journal of Agricultural Sciences 13:11-17.
- Farah Z (1992). Vitamin contents of camel milk. International Journal of Nutrition and Vitamin Research 62:30-33
- Faye B and Seboussi R (2008). Experimental selenium intoxication in camel. Veterinariya 3:18-29.
- Hatam LJ and Kayden HJ (1979). A performance liquid chromatographic method for the determination of tocopherol in plasma and cellular elements of blood. Journal of Lipid Research 20:639-645.
- Konuspayeva G, Lemarie E, Faye B, Loiseau G and Montet

- D (2008). Fatty acid and cholesterol composition of camel's (*Camelus bactrianus, Camelus dromedarius* and hybrids) milk in Kazakhstan. Dairy Sciences and Technology 88:327-340.
- LeBlanc SJ, Herdt TH, Seymour WM, Duffield TF and Leslie KE (2004). Peripartum Serum Vitamin E, Retinol, and Beta-Carotene in Dairy Cattle and Their Associations with Disease. Journal of Dairy Sciences 87:609-619.
- Maas J, Parish SM and Hodgson DR (1990). Nutritional myodegeneration. In. Large animal internal medicine, BP Smith Ed., St Louis, CV Mosby Co. pp 1352-1357.
- Mohamed HE (2004). A note on plasma antioxidant status in sudanese camels (*Camelus dromedarius*) affected by musculoskeletal disorders. Journal of Camel Practice and Research 11:65-66.
- Mousa HM, Omer OH, Ali BH, Al-Wabel N and Ahmed SM (2006). Antioxidant levels in tissues of young and adult camels (*Camelus dromedarius*). Journal of Physiology and Biochemistry 62:213-218.
- Nozière P, Martin B, Grolier P, Durand D, Ferlay A, Gruffat D, Pradel P, Prache S, Rock E, Chilliard Y and Petit M (2004). Evolution des teneurs du plasma et du lait de vache en â-carotène et autres composés d'intérêt nutritionnel lors d'un changement de régime (ensilage d'herbe puis foin). Rencontre Recherches Ruminants 11:63-66.
- Pehrson B and Hakkarainen J (1986). Vitamin E status oh healthy Swedish cattle. Acta Veterinary Scandinavica 27:351-360.
- Radostits OM, Blood DC, Handerson JA, Aundel JH and Gray CC (1995). Veterinary Medicine, A text book of the diseases of cattle, sheep, pigs, goats, and horses, 8th Ed. Bailliere Tindal, London.
- Seboussi R, Faye B, Alhadrami G, Askar M, Ibrahim W, Hassan K and Mahjoub B (2008). Effect of different selenium supplementation levels on selenium status in camel. Biological Trace Element Research 123:124-138.
- Seboussi R, Faye B, Askar M, Hassan K and Alhadrami G (2009). Effect of selenium supplementation on blood status and milk, urine and faecal excretion in pregnant and lactating camel. Biological Trace Element Research 128:45-57.
- Seboussi R, Faye B and Alhadrami G (2004). Facteurs de variation de quelques éléments trace (sélénium, cuivre, zinc) et d'enzymes témoins de la souffrance musculaire (CPK, ALT et AST) dans le sérum du dromadaire (Camelus dromedarius) aux Emirats Arabes Unis. Revue d'Elevage et Médecine Vétérinaire des Pays Tropicaux 57:87-94.
- Tiwary AK, Stegelmeier BM, Panter KE, James LF and Hall JO (2006). Comparative toxicosis of selenium selenite and sélénométhionine in lambs. Journal of Veterinary Diagnostic and Investigation 18:61-70.
- Vatassery GT, Nelson MJ and Kuskowski MA (1991). Vitamin E (tocopherols) in human cerebrospinal. American Journal of Clinical Nutrition Fluide 53:95-99.