



Vitamin and water requirements of dairy sheep

Federico Infascelli¹, Giuseppe Moniello²,
Monica Isabella Cutrignelli¹, Fulvia Bovera¹

¹Dipartimento di Scienze Zootecniche e Ispezione degli Alimenti. Università di Napoli "Federico II", Italy

²Dipartimento di Biologia Animale. Università di Sassari, Italy

Corresponding author: Prof. Federico Infascelli. Dipartimento di Scienze Zootecniche e Ispezione degli alimenti, Sezione Beniamino Ferrara. Facoltà di Medicina Veterinaria, Università di Napoli Federico II. Via F. Delpino 1, 80137 Napoli, Italy - Tel. +39 081 4421931 - Fax: +39 081 292981 - Email: infascel@unina.it

Paper received March 30, 2004; accepted June 7, 2004

ABSTRACT

The authors review the physiological role and the daily requirement of fat-soluble vitamins (A, D, E and K), vitamin C and water in dairy sheep. Regarding the vitamins, classical clinical symptoms and/or non-specific parameters, such as lowered production and reproduction rates are associated with their deficiencies or excesses. Until the last decade, these compounds were considered important only for the prevention of such alterations; currently, there is more emphasis on their function as the vitamins can play a key role in optimising animal health. In this respect, of particular interest is the action of the antioxidant vitamins (especially vitamin C, vitamin E and beta-carotene) in improving the efficiency of the immune system.

Key words: Vitamin requirements, Water, Dairy sheep.

RIASSUNTO

FABBISOGNI VITAMINICI E IDRICI DELLA PECORA IN LATTAZIONE

Gli autori passano in rassegna il ruolo fisiologico e i fabbisogni giornalieri delle vitamine liposolubili (A, D, E e K), della vitamina C e dell'acqua nella pecora in lattazione. Viene inoltre enfatizzato il ruolo delle vitamine nell'ottimizzazione della salute degli animali (in particolare l'aumento dell'efficienza del sistema immunitario ad opera delle vitamine C ed E e del beta-carotene).

Parole chiave: Fabbisogni vitaminici, Acqua, Pecore in lattazione

Introduction

Although today it is known that only some of them contain aminic nitrogen, the term vitamins (vital amines), coined in the 1912, is commonly used to indicate all those organic compounds which in small doses are essential to the vital functions of the animals, their growth and their

health. Vitamins are classified on the basis of their solubility as fat-soluble and water-soluble. In the first group are the vitamins A, D, E and K, and in the second those of the complex B and vitamin C. For ruminants it is necessary to introduce only fat-soluble vitamins with the foodstuffs, as vitamins or in the form of precursors, because in the rumen the vitamins of group B are synthesised, while

vitamin C is produced in the same cells of the organism. However, under specific conditions relating to stress and high productivity, ruminants have recently been shown to have a particular need for the B vitamins, thiamin and niacin. Likewise, vitamin B12 cannot be synthesised in the rumen if the essential building block, cobalt, is lacking in the diet.

Classical clinical symptoms and/or non-specific parameters, such as reduced production and reproduction rates are associated with vitamin deficiencies or excesses. Until the last decade, these compounds were considered important only for the prevention of such alterations; currently, there is more emphasis on their role as the vitamins can play a key role in optimising animal health. In this respect, of particular interest is the action of the antioxidant vitamins (especially vitamin C, vitamin E and beta-carotene) in improving the efficiency of the immune system. These vitamins, in fact, contribute to the maintenance of the structural and functional integrity of the cells, opposing the action of free radicals (Chew, 1995). The latter, whose production is stimulated by granulocytes to destroy the intracellular pathogens, have to be in turn eliminated because their accumulation can cause damage to cells (Padh, 1991). Furthermore, corticosteroids are producers of free radicals. The action of the antioxidant vitamins is strengthened by the presence of numerous metalloenzymes, such as glutathione peroxidase (selenium), catalysis (iron) and superoxide dismutase (copper, zinc and manganese).

Vitamin C is the most important antioxidant among extracellular fluids and can protect biomembranes against lipid peroxidation damage by eliminating free radicals before the latter can initiate peroxidation (Frei *et al.*, 1989). Vitamin C and E diet supplementation resulted in a 78% decrease in the susceptibility of lipoproteins to oxidation (Rifici and Kachadurian, 1993).

In light of the above, if the antioxidant vitamins act whenever free radicals are formed, it is obvious that any infectious process and/or stress factor can determine a depletion of the body's reserves, especially those of vitamin E. For this reason it appears necessary to correct the suitable vitamin requirements for individual species and

for different physiological phases. In other words, to the normal quantities administered for covering the normal needs of maintenance and production, a surplus should be provided to either support the antioxidant activity of such vitamins, or as a preventive measure, taking into account the action of stimulus (for instance that provided by vitamin E, in particular, in the synthesis of immunoglobulines IgG) (Tengerdy, 1980). Smith and Conrad (1987) recorded a 42.2% lower incidence of mastitis in dairy cows whose diet had been supplemented with levels of vitamin E and selenium superior to those recommended by National Research Council (NRC, 1989). These authors attributed such results to the action of stimulus provided by the vitamin E-selenium combination on the activity of the polymorphonuclear neutrophils (PMN), which are the major line of defence against bacteria in the mammary gland. An increase in the bactericidal activity of the PMNs has been reported by Daniel *et al.* (1991) subsequent to increasing the levels of beta-carotene in the diet.

Water is an essential element of equal importance to foodstuffs in the life and health of animals. If it is lacking or is of poor quality, it can represent one of the greatest limiting factors in breeding. Indeed, water represents 65% of the live weight in adults and around 77% in newborns.

Vitamins

Vitamin A or retinol

Vitamin A preserves the structural integrity of the epithelial tissue, plays a very important role in reproductive activity, and it particularly stimulates the growth of skeletal tissue. Vitamin A is also involved in maintaining the normal function of sight (Moore, 1957; Weber, 1983). This vitamin is stored in the liver and in grazing lambs its reserve is completely consumed after approximately 200 days, if it is not renewed.

Ovines do not receive vitamin A per se from vegetable foodstuffs. Rather it is obtained in the form of precursors (of which beta-carotene possesses the greatest biological activity) that are converted into retinol in the intestine (Moore, 1957). Green forage is rich in carotenes which means that grazing ovines rarely manifest defi-

ciency syndromes; this is also due to the aforementioned high quantities of vitamin A stored into the liver.

The activity of vitamin A is measured in International Units (U) which is equal to that expressed by 0.300 micrograms of crystalline trans-retinol, 0.344 micrograms of acetate trans-retinol or 0.550 micrograms of palmitate trans-retinol. Although the equivalent in vitamin A of beta-carotene is equal to 681 U of vitamin A/mg of beta-carotene, this value should only be attributed to the precursor as able to satisfy the minimum requirement of vitamin A. In fact, the potential biological activity of beta-carotene depends on various factors, such as the supply level and the body condition of the animals (Myers et al, 1959; Faruque and Walker, 1970). Also, the content in fat and in proteins of the diet, their digestibility, the mixture with other isomers of the carotenoid, as well as the presence of antioxidants, seem to develop a role which is not indifferent as respects the biological availability of beta-carotene in the foodstuffs (Ullrey, 1972).

The requirement of vitamin A for ovines is expressed as the quantity of retinol or its precursor, beta-carotene, which can prevent crepuscular blindness and ensure a normal reproduction rate in the animals (Guilbert et al, 1937). Subsequent

studies showed, nevertheless, that the best indicator of a vitamin A deficiency in ovines is represented by an increase in the pressure of the cerebrospinal liquid (Eveleht *et al.*, 1949; May, 1982); this condition was avoided by assuring a minimum supply of 8 - 16 micrograms of retinol/kg of live weight in growing lambs. Faruque and Walker (1970) indicated 14 micrograms of retinol or 69 micrograms of beta-carotene/kg of live weight as the quantity to be administered in order to guarantee a small liver reserve of vitamin A in lambs. In absence of additional information, the minimum vitamin A requirement for ovines of any category is recognized as being equal to 69 micrograms of beta-carotene/kg of live weight/day or to 47 U of retinol/kg of live weight/day. Nevertheless, in the end of pregnancy phase and during lactation the requirements are equal to 125 micrograms/kg of live weight/day of beta-carotene and 85 U/kg of live weight/day of vitamin A. Additionally, in the first 6 - 8 weeks of lactation sheep that nurse twins have requirements equal to 147 micrograms/kg of live weight/day of beta-carotene and 100 U/kg of live weight/day of vitamin A (the milk of sheep contains around 1,500 U of vitamin A/litre).

Tables 1-4 report the vitamin A requirements of ovines.

Table 1. Daily requirements (U) of vitamin A and vitamin E in sheep at maintenance level, during the breeding period and in the first 15 weeks of gestation.

Live weight (kg)	Daily weight gain (g)	Vit. A	Vit. E
Maintenance			
50	10	2350	15
60	10	2820	16
70	10	3290	18
Flushing			
50	100	2350	24
60	100	2820	26
70	100	3290	27
First 15 weeks of gestation			
50	30	2350	18
60	30	2820	20
70	30	3290	21

Table 2. Daily requirements (U) of vitamin A and vitamin E in sheep in the first 6-8 weeks of lactation suckling single lambs or twins.

	Live weight (kg)	Daily weight gain (g)	Vit. A	Vit. E
Single lamb	50	-25	4250	32
	60	-25	5100	34
	70	-25	5950	38
Twins	50	-60	5000	36
	60	-60	6000	39
	70	-60	7000	42

Table 3. Daily requirements (U) of vitamin A and vitamin E in ewe lambs.

	Live weight (kg)	Daily weight gain (g)	Vit. A	Vit. E
First 15 weeks of pregnancy - non lactating	40	160	1880	21
	50	135	2350	22
	60	135	2820	24
Last 4 weeks of pregnancy (100-120% lambing rate prevision)	40	180	3400	22
	50	160	4250	24
	60	160	5100	26
Last 4 weeks of pregnancy (130-175% lambing rate prevision)	40	225	3400	22
	50	225	4250	24
	60	225	5100	26
First 6-8 weeks of lactation suckling single lamb	40	-50	3400	26
	50	-50	4250	32
	60	-50	5100	34
First 6-8 weeks of lactation suckling twins	40	-100	4000	32
	50	-100	5000	34
	60	-100	6000	38

Table 4. Daily requirements (U) of vitamin A and vitamin E in growing male and female replacement lambs.

	Live weight (kg)	Daily weight gain (g)	Vit. A	Vit. E
Female replacement lambs				
	30	227	1410	18
	40	182	1880	21
	50	120	2350	22
Male replacement lambs				
	40	330	1880	24
	60	320	2820	26
	80	290	3760	28

Vitamin D

The most important known forms of this vitamin are ergocalciferol (D2) and cholecalciferol (D3) whose precursors (ergosterol and 7-dehydrosterol) are converted into vitamins at the subcutaneous level by the ultraviolet rays of the sun. Sheep use the two active forms indifferently (Church and Pond, 1974). According to De Luca (1974) cholecalciferol is activated in the liver and in the kidney and it influences calcium absorption and regulates calcium deposits and mobilization from the skeletal tissue. Therefore, a lack of vitamin D is associated with such phenomena as rickets and osteomalacia, in young animals and adults, respectively. These conditions, however, are not frequent in ovines thanks to widely adopted grazing systems that guarantee sufficient exposure to sunlight. The benefits of the latter, nevertheless, are less evident in breeds with a dark mantle or with abundant wool. Some authors suggest supplementing the diet with vitamin D in cases in which atmospheric conditions are characterised by cloudy skies for prolonged periods (Crowley, 1961) or when the animals are raised in stalls (Hidioglou *et al.*, 1979). In any case, it is important to note that hay obtained through traditional means represents a good source of vitamin D.

The activity of vitamin D is measured in International Units (U). According to Windholz *et al.* (1983) an U is equal to the anti-rickets activity of 0.025 micrograms of D3 in crystalline form. The vitamin D requirements for all categories of ovines

are equal to 555 I.U./100 kg of live weight/day except for the lambs just weaned for which 666 I.U./100 kg of live weight/day are recommended.

Vitamin E

There are eight compounds in nature known as tocopherols, which share an analogous chemical structure and demonstrate different levels of biological activity. The most active and also the most diffused is alpha-tocopherol, which is widely found in green forages. Its concentration in hays can actually be reduced by as much as 90%, depending on the season of mowing, the period of storage, as well as the time between mowing and the drying process. An important role is attributed to vitamin E in strengthening cellular defences against reactive oxygen and free radicals (Rammel, 1983) and therefore in preserving, together with selenium, the integrity of cellular membranes by avoiding the peroxidation of the phospholipids. Among the different pathologies imputable to vitamin E deficiency is muscular degeneration, which is the most frequent and most important in ovines, particularly in lambs. In addition to the skeletal muscles, the degeneration can also sometimes affect the heart, in which case the animal can die without premonitory symptoms. In the less serious forms it can cause heart or respiratory failure during minimal physical exertion.

The International Units (I.U) for vitamin E is defined as being equal to 1 mg of dl-acetate-alpha-tocopherol.

The vitamin E requirements for ovines are indi-

cated as 20 U/kg of dry matter intake and 15 U/kg of dry matter intake, respectively, for lambs weighing less than 20 kg and for those weighing more, as well as for adult subjects.

Tables 1-4 report the vitamin E requirements of ovines.

Vitamin K

The most widely diffused forms are phyloquinone (vitamin K1) and menaquinone (vitamin K2). Vitamin K is also defined as a coagulation factor or antihæmorrhagic factor, because it is necessary for the synthesis of prothrombin in the liver. Prothrombin is the inactive precursor of thrombin, an enzyme that transforms fibrinogen into fibrin, which is responsible for the formation of clotting. The leaves of either green forage or hay are good sources of vitamin K (Church and Pond, 1974). Vitamin K2 is also widely synthesised in the rumen, which means that under normal conditions its addition to the diet is not useful (McElroy and Gross, 1940; Matshiner, 1970).

Water

Water is an essential element of equal importance to foodstuffs in the life and health of animals. If it is lacking or is of poor quality, it can represent one of the greatest limiting factors in breeding. This is confirmed by the fact that among the components of the animal body, water represents 65% of the live weight in adults and around 77% in newborns.

The water functions in the animal organism, all of notable physiological importance, can be summarized as follows:

- it is the dispersive medium and solvent of substances at the endocell level, in the blood, in the urine and in glandular secretions. Thanks to its elevated dielectric power it determines the ionisation of the polar molecules; it also regulates the temperature of the body by storing and then distributing thermal energy from the biochemical reactions of the metabolism;
- it is the vehicle used by absorbed nourishing substances as well as by the catabolites excreted by the urines;
- it is a fundamental element in a host of bio-

chemical reactions (hydrolysis, hydrations, oxidation-reductions) and, together with the CO₂, it represents the principal and final product of the metabolism. Finally, its lubricating action (through the synovial, peritoneal and pleural liquids) on the articular surfaces and on the viscera cannot be overlooked.

In light of what has been demonstrated and based on numerous studies, while animals can live losing up to 40% of their live weight, a loss between 10 and the 15% of the normal content in water of the organism would result in death. It is therefore evident that it is necessary to maintain a dynamic equilibrium between the quota of water taken in and that eliminated through the urine, faeces, milk, perspiration, aqueous vapours in expired air, and cutaneous transpiration.

An animal's water supply is composed of three main factors: drinking water; water content of foodstuffs; and metabolic water, which results from oxidation-reductions in the loading of carbohydrates, lipids and amino acids. The turnover of water is always very high and differs as a function of the climate and, above all, of the animal species. In sheep half of the water in the organism renews itself in about 5 days.

Based on the above, it appears evident that the drinking water requirement for animals depends, first of all, on the quantity eliminated daily, but also on the water content in foodstuffs, as well as on the quota produced in metabolising nourishing substances. Therefore, conditions exist in which the animal could also live with modest quantities of drinking water: for instance, when it consumes exclusively fresh forages (80-85% of water) or when endermic, faecal and urinary elimination is reduced such that the water produced by the metabolism results almost enough to meet the requirements. It should be noted that while the quota of water produced in the metabolism of fats results remarkably high, it should be considered average and low, respectively, for carbohydrates and proteins. This means that, other conditions being equal, a protein diet determines an increase in the water requirement and that, in conditions in which water is lacking, the animals resort primarily to the catabolism of their own adipose reserves. Finally, it should be noted that the min-

eral composition of the diet plays a relevant role in the modification of the water requirement in animals. The increase in water consumption under conditions of inadequate supply of sodium chloride is a prime example.

For ovines, Forbes (1968) suggests estimating the total intake of water (TI) beginning from the amount of dry matter intake (DMI), by means of the following equation:

$$TI = 3.86 \text{ DMI} - 0.99$$

In light of the significant correlation between water supply and environmental temperature, the Author proposes calculating the water assumed per unit of dry matter intake (kg/kg) under conditions of temperature higher than 1 °C, using the following equation:

$$TI/DMI = 0.18 T + 1.25$$

where T is the average weekly temperature expressed in centigrade.

During pregnancy, the consumption of water increases beginning in the third month and doubles in the fifth, and results greater for the sheep that will give birth to twins in comparison to those with a single birth. A water deficiency during pregnancy, together with the diminution of dry matter intake, represents a predisposing cause of pregnancy toxæmia. According to Forbes (1968), the sheep's water

consumption during the first phase of lactation results superior to the sum of that ingested by a non pregnant subject and the water contained in the milk because of the acceleration of the metabolism and the consequent increase in the water excretion during this phase. On average, the water requirement of a sheep during lactation would be double that of a dry subject (Forbes, 1968).

The temperature of the ingested water is also of a certain importance, at least when it reaches levels close to the 0 °C. Brod *et al.* (1982), in fact, noticed that, in sheep, the consumption of water at such temperature inhibits the activity of the microorganisms in the rumen thereby determining an increase in the pH and reducing the concentration of volatile fatty acids and the digestibility of the foodstuffs.

Finally, the quality of the drinking water is very important. Ovines, like the other ruminants, can consume water with a bacterial content superior to that considered potable for humans without showing problems. They are mostly sensitive to the presence of nitrates that are transformed by the bacteria in the rumen into nitrites with a greater toxic potential. Furthermore, high concentrations of chlorine in the water seem to negatively influence fermentation in the rumen.

Table 5 reports the water requirements of ovines (kg/kg of dry matter intake) by age, productive phase, and environmental temperature.

Table 6 shows the evaluation of water with

Table 5. Water requirements (kg/kg of dry matter intake).

	Environmental temperature (°C)		
	<16	16 - 20	>20
Lambs up to 4 weeks	4.0	5.0	6.0
Growing subjects or non pregnant adults	2.0	2.5	3.0
Central phase of the pregnancy:			
1 lamb	3.0	3.8	4.6
2 - 3 lambs	3.3	4.1	4.9
Final phase of the pregnancy:			
1 lamb	4.1	5.2	6.3
2 - 3 lambs	4.4	5.5	6.6
Lactation:			
First month	4.0	5.0	6.0
Second and following months	3.0	3.7	4.5

respect to chlorine, nitrites and nitrates content (Savoini *et al.*, 2001).

Table 6. Evaluation of water with respect to chlorine (Cl), nitrites (NO₂) and nitrates (N-NO₃) content.

Cl (mg/L):	
500 - 3000	No problem
> 3000	Decrease of rumen fermentation activity
NO ₂ (mg/L):	
traces	Good
> traces	Dangerous for the health
N-NO ₃ (mg/L):	
< 100	No problem
100 - 300	Dangerous for the health, with contemporary intake of nitrate rich feeds
> 300	Poisoning with contemporary intake of nitrate rich feeds

Conclusions

Vitamin nutrition should be considered important not only for preventing signs of deficiency, but also for optimising animal health, productivity and product quality. Thus, a great deal of research is required to further understand the metabolism and functions of all vitamins. Specific studies are required to investigate the interrelationships among vitamins and other nutrients, as well as among vitamin carriers and tissue receptors. From a practical viewpoint, what are the vitamin requirements under diverse conditions and how do requirements differ on the basis of functions (e.g. growth, reproduction, optimum immune response)?

As regards water, in light of what has been demonstrated, it is evident that there is a need not only to maintain a dynamic equilibrium between the quota taken in and that eliminated, but also to guarantee its good quality, mainly by taking into account the toxic potential of nitrites which derive from the transformation of nitrates, and the negative influence of chlorine on fermentation in the rumen.

REFERENCES

- BROD, D.L., BOLSEN, K.K., BRENT, B.E., 1982. Effect of water temperatures on rumen temperatures, digestion and rumen fermentation in sheep. *J. Anim. Sci.* 54:179-185.
- CHEW, B.P., 1995. Antioxidant vitamins affect food animal immunity and health. *J. Nutr.* 125:180-187.
- CHURCH, D.C., POUND, W.G., 1974. Basic animal nutrition and feeding. Albany Printing, Albany, NY, USA.
- CROWLEY, J.P., 1961. Rickets in November-born lambs. *Vet. Rec.* 73:295-297.
- DANIEL, L.R., CHEW B.P., TANAKA, T.S., TJOELKER, L.W., 1991. Beta-carotene and vitamin A effects bovine on phagocyte function *in vitro* during the peripartum period. *J. Dairy Sci.* 74:124-132.
- DE LUCA, H.F., 1974. Vitamin D: the vitamin and the hormone. *Fed. Proc.* 33:2211-2219.
- EVELETH, D.F., BOLIN, D.W., GOLDSBY, A.I., 1949. Experimental avitaminosis A in sheep. *Am. J. Vet. Res.* 10:250-261.
- FARUQUE, O., WALKER, D.M., 1970. Relative the biological potencies of retinyl palmitate and beta-carotene for the milk-fed lamb. *Brit. J. Nutr.* 24:23-28.
- FORBES, J.M., 1968. The water intake of ewes. *Brit. J. Nutr.* 22:33-42.
- FREI B., ENGLAND, L., AMES, B.N., 1989. Ascorbate is an outstanding antioxidant in human blood plasma. *Proc. Natl. Acad. Sci.* 86:6377-6384.
- GUILBERT, H.R., MILLER, R.F., HUGHES, E.H., 1937. The minimum vitamin A and carotene requirement of cattle, sheep and swine. *J. Nutr.* 13:543-550.
- HIDROGLOU, M., WILLIAMS, C.J., IVAN, M., 1979. Pharmacokinetics and amounts of 25 hydroxycholecalciferols in sheep affected by osteodystrophy. *J. Dairy Sci.* 62:567-572.
- MATSCHINER, J.T., 1970. Characterization of vitamin K from the contents bovine of rumen. *J. Nutr.* 100:190-196.
- MAY, B.J., 1982. The Minimum vitamin A requirement for growing and finishing lambs. M.S. Thesis, Angel State Univ., S. Angel Tex, USA.
- MCELROY, L.W., GOSS, H., 1940. To quantitative study of vitamins in the rumen contents of sheep and cows fed vitamins low-diets. I. Riboflavin and vitamin K. *J. Nutr.* 20:527-533.

- MOORE, T., 1957. Vitamin A. Elsevier Publishing Company, Amsterdam, The Netherlands.
- MYERS, G.S. JR., EATON, H.D., ROUSSEAU, J.E. JR., 1959. Relative value of carotene from alfalfa and vitamin A from dry carrier fed to lambs and pigs. *J. Anim. Sci.* 18:288-293.
- NATIONAL RESEARCH COUNCIL, 1989. Nutrient Requirements of Dairy Cattle. 6th rev. ed. National Academy Press, Washington, DC, USA.
- PADH, H., 1991. Vitamin C: newer insights into its biochemical functions. *Nutr. Rev.* 49:65-70.
- RAMMELL, C.G., 1983. Vitamin E status of cattle and sheep. The background review. *New Zeal. Vet. J.* 31:179-191.
- RIFICI, V.A., KACHADURIAN, A.K., 1993. Dietary supplementation with vitamins C and its inhibits in vitro oxidation of lipoproteins. *J. Am. Coll. Nutr.* 12:631-637.
- SAVOINI, G., DELL'ORTO, V., PIVA, G., 2001. Qualità dell'acqua di abbeverata per gli animali. In: *Metodi di analisi delle acque per uso agricolo e zootecnico*. Franco Angeli Editore, Milano, Italy, pp 1-15.
- SMITH, K.L., CONRAD, H.R., 1987. Vitamin E and selenium supplementation for dairy cows. RCD 7442. pp 47-56 in *Proc. Hoarse Technical The role of vitamins on animal performance immune and response*, Daytona Beach, USA.
- TENGERDY, R.P., 1980. Disease resistance: immune response. In: L.J. Machhlin (ed.) *Vitamin E: to comprehensive treatise*. Marcel Dekker, NY, USA, pp 429-444.
- ULLREY, D.E., 1972. Biological availability of fat-soluble vitamins: vitamin A and carotene. *J. Anim. Sci.* 35:648-653.
- WEBER, F., 1983. Biochemical mechanisms of vitamin A action. *Proc. Nutr. Soc.* 42:31-39.
- WINDHOLZ, M., BUDAVARI, S., BLUMETTI, R.F., OTTERBEIN, E.S., 1983. *The Merck Index: An Encyclopedia of Chemicals, Drugs and Biologicals*. 10th rev. ed. Merck and Co., New Jersey, USA.