Indian J Anim Health (2022), 61(2): 335- 340
DOI: https://doi.org/10.36062/ijah.2022.11222

Research Article

Fat-soluble vitamins supplementation favours peri-parturient health in Murrah buffaloes

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Abstract

Fat-soluble vitamins, due to their antioxidant properties, have long been linked to the reproductive performance of dairy animals. Pregnant (-30 day) Murrah buffaloes (n=22) were distributed among control (C, n=8), treatment 1 (T_1, n=7) and treatment 2 (T_2, n=7). Buffaloes were fed either a control diet as per standard (ICAR, 2013) recommendations (C) or supplemented with vitamin A @ 75,000 IU daily for the whole study and vitamin E @ 1500 IU from -30 to +30 days of the study, followed by 500 IU daily up to 90 days of calving in T_1 group, vitamin A @ 1,50,000 IU daily for whole study and vitamin E @ 3000 IU from -30 to +30 days of study, followed by1000 IU daily up to 90 days of calving in T_2 group. Treatments helped in reducing the time of the expulsion of the placenta (4.02, 4.09 and 5.51 hours in T_1, T_2 and C, respectively) and somatic cell count (P>0.05) as compared to the control group. Supplementation of vitamins at a higher level (T_2) helped to improve the cyclicity % (85.71) of buffaloes as compared to the other two groups (T_1, 42.86% and C, 37.5%), in addition, to the complete lack of any post-partum reproductive disorders, including mastitis, due to a higher level of supplementation (T_2), while the control group showed one each incidence of metritis, mastitis, retained placenta, and prolapse of the uterus as well. The study shows that a higher dose of vitamins A and E could improve peri-parturient health in buffaloes.

Key words: Buffaloes, Cyclicity, Mastitis, Peri-parturient disorders, Vitamins

Highlights

- Supplementation of fat-soluble vitamins improves the estrus cycle in Murrah buffaloes.
- The prevalence of post-partum reproductive disorders in buffaloes can be decreased by supplementing with fat-soluble vitamins at higher doses.
- Supplementation of fat-soluble vitamins can reduce the incidence of mastitis.

INTRODUCTION

India is the leading country having livestock population in the world. Buffalo is a prime dairy animal of India (BAHS, 2019), but buffaloes are especially prone to the silent estrus/anestrus of cyclicity after parturition. Data indicates that only about 34–49% of buffaloes showed estrous within the first 90 days after parturition, which even extends up to 150 days in 31–42% of buffaloes (El-Wishy, 2007). Delay in resuming cyclicity is a critical issue for a dairy enterprise as it directly diminishes the sustainability of economic returns. Post-parturient disorders are also a big constraint on the economic viability of the dairy sector. Mastitis, metritis, retention of the placenta, and prolapse of the uterus are the major hurdle which interfere with the normal physiology of the body and directly or indirectly affects the sustainability of the dairy industry by affecting production and hampering reproduction performances.

During the peri-parturient period, blood levels of these vitamins dramatically reduce (Goff and Stabel, 1990; Weiss *et al.*, 1994), and dairy animals become vulnerable to different

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post-parturient disorders (Block, 2010). Vitamin A and E have a direct involvement in normal reproduction and immunity due to their role in epithelium membrane integrity and antioxidant role. In addition to enhancing immunity status, vitamin A and E supplementation have been observed to enhance milk quality, udder health (Pottier *et al.*, 2006), and reproduction status (Wilde, 2006) of dairy animals. The hypothesis of this study is based on the fact that these vitamins could help in maintaining optimally peri-parturient health in buffaloes.

MATERIALS AND METHODS

The study was performed on Murrah buffaloes maintained at the Animal Farm Section of ICAR-Central Institute for Research on Buffaloes, Hisar, Haryana, India, from December 2017 to June 2018. The farm is located 212 meters above the mean sea level. The animal experimentation was carried out on 22 multiparous buffaloes in the advanced stage of pregnancy (30 days before the expected date of parturition) after being distributed randomly into three groups having 8 buffaloes in the control group and 7 buffaloes in each treatment group on the basis of parity, most probable producing ability (MPPA) of milk and body weights.

Buffaloes were individually fed as per the nutrient requirements (ICAR, 2013) during the study period, using a concentrate mixture, wheat straw, and green berseem. Buffaloes of group T_1 were fed additionally supplemented with vitamin A @ 75,000 IU daily for the whole study and vitamin E @ 1500 IU from -30 to +30 days of study, followed by 500 IU daily up to 90 days of calving in group T_1, vitamin A @ 1,50,000 IU daily for whole study and vitamin E @ 3000 IU from -30 to +30 days of study, followed by1000 IU daily up to 90 days of calving in group T_2, daily after mixing with concentrate mixture during morning feeding.

Representative samples of feed offered were collected, processed, and analyzed for proximate principles (AOAC, 2012), neutral detergent fiber (NDF), and acid detergent fiber (ADF) content (Van Soest *et al.*, 1991).

Animals were examined for ultrasonography using a real-time B-mode ultrasound scanner (Just Vision 200, Model 320A, Toshiba, Japan) equipped with an intraoperative 7.0 MHz micro convex transducer with an interval of one month for the initial 3 months of the post-partum period i.e. between days 25 to 35, 55 to 65, and 85 to 95 after parturition. Observations of the expulsion of the placenta in (hrs) buffaloes were recorded after parturition. The somatic cell counts of milk were recorded with the help of Ekomilk somatic cells analyzer on the fortnightly basis for 12 weeks. Cases of post-parturient reproductive disorders like retention of the placenta, metritis, and prolapse of the uterus were recorded with incidences of mastitis in buffaloes.

All the data were analyzed with the help of Statistical Package for Social science (SPSS, 2010) version 16 software using the General Linear Model; multivariate for comparison among groups, repeated measures were applied to determine the difference among different periods within the same group.

RESULTS

Grouping of animals: Twenty-two buffaloes in the advanced stage of pregnancy (-30 days) were selected and grouped into three equal groups based on their most probable producing ability (MPPA) for milk (average 2184.58 kg), parity (average 2.27), and body weights (average 589.22 kg). Eight buffaloes were assigned to control, while seven each were in two treatment groups, i.e. T_1 and T_2.

Chemical composition of feeds: The chemical composition of representative feed samples collected during the experimental period indicated that the feed could fulfil the nutrient requirements (ICAR, 2013) of buffaloes. Concentrate mixture, wheat straw, and green berseem had 19.21%, 3.08%, and 16.19% crude protein and 56.25%, 44.79%, and 47.9% nitrogen-free extract, respectively.

Expulsion of placenta: The time interval (in hours) of the expulsion of the placenta after calving and their overall mean ± SE values in treatment as well as in control groups have been presented in Table 1. Although a non-significant (p>0.05) effect of supplementation of vitamins A and E was observed, a clear-cut trend was apparent (P=0.126 and 0.144). This treatment group had early (T_1, 4.02±0.23 and T_2, 4.09±0.43 hrs) expulsion of the placenta as compared to the control (5.51±0.96 hrs) group buffaloes.

Somatic cell count of Murrah buffalo's milk: There was no significant (P>0.05; Table 2) effect on SCC of treatment was observed at different fortnightly intervals except at day 96, where somatic cell counts were significantly (P<0.05; Table 2) lower in both the treatment groups as compared to control. The overall mean value also showed a trend; the somatic cell count in the milk of the treatment groups was lower (T_2 and T_1) compared to the control group, indicating a positive effect on milk quality due to vitamin supplementation. Somatic cell count was also significantly (P<0.05) reduced in both treatment groups at day 96 of the study with numerically lower values in both the treatment groups throughout the study.

Cyclicity percentage: Data regarding cyclicity (%) in buffaloes at the monthly interval (day 25-35, 55-65, and 85-95) is presented in Table 3.

Table 1. Effect of vitamin A and E supplementation on the expulsion of the placenta (hours) in buffaloes

C (N=8)	T_1 (N=7)	T_2 (N=7)	SEM	P value			
				C*T_1	C*T_2	T_1*T_2	Overall
5.51±0.96	4.02±0.23	4.09±0.43	0.39	0.126	0.144	0.941	0.261

Table 2. Effect of vitamin A and E supplementation on somatic cell count (^{1000}/cm^3) of Murrah buffalo's milk

DIM	C (n=8)	T_1 (n=7)	T_2 (n=7)	SEM	P value			
					C*T_1	C*T_2	T_1*T_2	Overall mean
6	150.00 ±22.35	126.83 ±99.63	136.00 ±44.31	34.45	0.403	0.595	0.764	0.690
21	371.50 ±261.58	106.67 ±93.05	94.14 ±4.14	98.61	0.488	0.268	0.680	0.523
36	187.25 ±77.28	128.33 ±93.34	101.29 ±8.33	40.45	0.749	0.399	0.264	0.504
51	393.63 ±258.98	117.40 ±101.81	105.57 ±15.57	98.82	0.620	0.251	0.518	0.508
66	247.00 ±78.54	108.17 ±109.35	94.86 ±4.86	45.17	0.783	0.180	0.295	0.368
81	141.25 ±26.76	114.33 ±26.82	95.29 ±5.29	13.26	0.958	0.164	0.193	0.297
96	176.13 ±19.76	109.17 ±13.25	95.86 ±4.66	11.23	0.004	0.001	0.531	0.002
P value	0.168	0.738	0.363					
Overall mean	238.11 ±101.99	164.80 ±43.32	111.94 ±8.70	39.59	0.592	0.208	0.472	0.442

Table 3. Effect of vitamin A and E supplementation on cyclicity (%) of buffaloes

Month	C(N=8)	T_1(N=7)	T_2(N=7)
1	2(25%)	2(28.57%)	3(42.86%)
2	0	0	3(42.86%)
3	1(12.5%)	1(14.29%)	0
Overall	3(37.5%)	3(42.86%)	6(85.71%)

Cyclicity % data indicates the superiority of supplementation in treatment groups; thus, cyclicity (%) was higher (85.71%) in group T_2, followed by T_1 (42.86%) and least in the control (37.5%) group.

Peri-parturient disorders: As per different peri-parturient disorders are concerned, the treatment group at a higher dose rate (T_2) of vitamin A and E supplementation did not show any of the observed peri-parturient disorder incidences. While, at a lower dose of supplementation (T_1), one each case was reported for metritis as well as mastitis, although buffaloes of the control group were more prone to different peri-parturient disorders as out of 8 buffaloes, four different buffaloes showed cases of metritis, retained placenta, mastitis as well as prolapse of the uterus.

DISCUSSION

The treatment group had early (T_1, 4.02±0.23 and T_2, 4.09±0.43 hrs) expulsion of the placenta as compared to the control (5.51±0.96 hrs) group buffaloes. Likewise, Bourne *et al*. (2007) also indicated that Vitamin E supplementation during the dry period is associated with a reduced risk of retention of fetal membrane and that the synthetic forms of Vitamin E are more effective than the natural compound. Oliveira *et al.* (2015) also reported a reduction (392 min vs. 490 min) in time of the expulsion of the placenta due to β-carotene supplementation in HF cows. Kaliannan *et al*. (2020) also reported earlier (6.9 hrs) shedding of fetal membranes due to prepartum injection of vitamin A against control cows (9.75 hrs); likewise, Panda *et al.* (2005) also reported a 17% reduction in retention of foetal membranes in buffaloes due to supplementation of vitamin E. Retained foetal membranes are a common problem in dairy farms, and it can influence the risk of other diseases and fertility of dairy cows (Dubuc *et al*., 2010).

The overall mean value of SCC also showed a trend in the somatic cell count in the milk of treatment groups was lower (T_2 and T_1) compared to the control group, indicating a positive effect on milk quality due to vitamin supplementation. In contrast, Oldham *et al.* (1991) did not find any effect on somatic cell count due to supplementation of 280 IU of vitamin A /kg body weight as compared to the group fed 75 IU /kg body weight. No response may be due to a sufficient level of vitamin A in the control group, while most of the previous researchers reported a reduction in somatic cell count and lower incidences of mastitis due to supplementation of vitamin E (Baldi *et al*., 2000; Politis *et al*., 2004; Panda and Kuar, 2008). Kuczynska *et al*. (2015) reported an inverse correlation between somatic cell count and beta-carotene concentration in milk. Likewise, Oliveira *et al*. (2015) also reported a reduction in incidences of subclinical mastitis due to the supplementation of beta carotene @ 1.2 g/cow/day. Our result also lines with Collet *et al*. (2017), who reported no significant effect on the SCC of milk on supplementation of vitamins A and E with trace minerals. Contrary to our result, Singh *et al.* (2020) reported that supplementation of vitamin E (1g/day/cow) significantly reduced the number of SCC in the milk.

The percentage of cyclicity was highest in the higher supplementation group, followed by the lower supplemental group, with the least in the control group; clear-cut emphasizes the favourable/promising impact of supplemental vitamins and the effect may be due to the antioxidant properties of vitamins which lead to least oxidative stress in treatment group supplied with a higher level of those vitamins. Bian *et al.* (2007) also reported reduced days from calving to heat due to vitamin A supplementation in HF cows. De ondarza *et al.* (2009) also reported a 22% pregnancy rate compared to 11% in the control group due to the supplementation of β-carotene in Holstein cows. In contrast, Oliveira *et al.* (2015) did not report any improvement in % conception at first service for day 90 of parturition, but in their experiment only a prepartum (14 days) supplementation of β-carotene was carried out in HF cows. Similar to the present observation, Khan *et al.* (2016) also reported a positive influence of vitamin E supplementation (@ 300IU/head/day in dairy cows) in the form of significantly increased progesterone concentration, which leads to quality graffian follicles with decreased number of follicles. In a study, Ahmed *et al.*(2017) supplemented vitamins A, D_3 and E daily in water from 4 weeks prepartum to seven weeks post-partum and reported a significant reduction in subclinical endometritis with shorter days open (97.5 vs. 152.1) with a higher pregnancy rate at day 120 (50 vs. 44%) and 150 (75 vs. 56.33) post-partum. Bhateshwar *et al.* (2021) also found improved fertility due to β-carotene supplementation in lactating cows, whereas Hye *et al.* (2020) found no improvement in fertility rate after giving β-carotene.

The buffaloes of the control group were more prone to different peri-parturient disorders as out of 8 buffaloes four different buffaloes showed cases of metritis, retained placenta, mastitis as well as prolapse of the uterus. Similarly, Kaliannan *et al.* (2020) did not reportany post-partum complications due to prepartum injection of vitamin A in dairy cows.

Supplementation of fat-soluble vitamins can help in improving the cyclicity percentage and reducing the incidences of post-parturient health disorders in buffaloes which may be due to their antioxidant role.

Conflict of interest: The authors have no conflict of interest to declare for this study.

Author's contribution: VM and AKV: Conceived and designed the research; V: Conducted experiments; RKS: Contributed new reagents or analytical tools; VM and V: Analyzed the data; V: Wrote the manuscript; VM and V: Edited and revised the manuscript as well as prepared it for journal submission. All authors read and approved the manuscript.

Ethics declaration: The care and treatment of all the animals utilized in the experiment were strictly in accordance with accepted guidelines for treating animals humanely. All animal-using experimental procedures were carried out in accordance with laws governing animal welfare.

REFERENCES

Ahmed EA, Elsayed DH, Kilany OE and El-Beltagy MA, 2017. Multivitamins preventive therapy against subclinical endometritis in buffaloes: its correlation to NEFA and oxidative stress. Reprod Biol, 17(3): 239-245, doi: 10.1016/j.repbio.2017.05.008

AOAC, 2012. Official Methods of Analysis of the Association of Official Analytical Chemistry, 19th edn., AOAC, International, Washington, USA

BAHS (Basic Animal Husbandry Statistics), 2019. Department of Animal Husbandry and Dairying, Krishi Bhawan, New Delhi

Baldi A, Savoini G, Pinotti L, Monfardini E, Cheli F *et al.*, 2000. Effects of vitamin E and different energy sources on vitamin E status, milk quality and reproduction in transition cows. J Vet Med A Physiol Pathol Clin Med, 47: 599-608, doi: 10.1046/j.1439-0442.2000.00323.x

Bhateshwar V, Rai DC and Duarv RK, 2021. Effect of beta-carotene supplementation on plasma carotene content and fertility of lactating crossbred Sahiwal cows. Indian J Anim Res, 55(8): 905-909

Bian SB, Elliott R, Immig I and Sun DF, 2007. The influence of β-carotene supplementation on post-partum disease and subsequent reproductive performance of dairy cows in China. J Anim Feed Sci, 16(2): 370-375, doi: 10.22358/jafs/74563/2007

Block E, 2010. Transition cow research–What makes sense today? In Proceedings High Plains Dairy Conference, pp 75-98

Bourne N, Laven R, Wathes DC, Martinez T and McGowan M, 2007. A meta-analysis of the effects of vitamin E supplementation on the incidence of retained foetal membranes in dairy cows. Theriogenology, 67(3): 494-501, doi: 10.1016/j.theriogenology.2006.08.015

Collet SG, Demeda MA, Taffarel GV, Taffarel L, Girardini LK *et al.*, 2017. Effect of injectable trace mineral supplement and vitamins A and E on production and milk composition of Holstein cows. Rev Ciênc Agrovet, 16: 463-72, doi: 10.5965/223811711642017463

De Ondarza MB, Wilson JW and Engstrom M, 2009. Case study: effect of supplemental β-carotene on yield of milk and milk components and on reproduction of dairy cows. Prof Anim Sci, 25(4): 510-516, doi: 10.15232/S1080-7446(15)30742-7

Dubuc J, Duffield TF, Leslie KE, Walton JS and LeBlanc SJ, 2010. Risk factors for postpartum uterine diseases in dairy cows. J Dairy Sci, 93: 5764-5771, doi: 10.3168/jds.2010-3429

El-Wishy AB, 2007. The postpartum buffalo: II. Acyclicity and anestrus. Anim Reprod Sci, 97(3-4): 216-236, doi: 10.1016/j.anireprosci.2006. 03.003

Goff JP and Stabel JR, 1990. Decreased plasma retinol, α-tocopherol and Zn concentration during the periparturient period: effect of milk fever. J Dairy Sci, 73: 3195-3199, doi: 10.3168/jds.S0022-0302(90)79010-8

Hye N, Klein-Jöbstl D, Blessing A, Burmeister J, Hamann N *et al.*, 2020. Effect of two postpartum intramuscular treatments with β-carotene (Carofertin®) on the blood concentration of β-carotene and on the reproductive performance parameters of dairy cows. Theriogenology, 148: 1-7, doi: 10.1016/j.theriogenology.2020.02.042

ICAR, 2013. Nutrient Requirements of Cattle and Buffalo. 3rdedn., Indian Council of Agricultural Research, New Delhi

Kaliannan R, Mani S, Mahakrishnan P, Subramaniyan P, Venkatraman V *et al.*, 2020. Effect of injectable anti-oxidant vitamins (A, E & SE) in the separation and expulsion of fetal membranes and involution in dairy cows. J Pharm Innov, 9(9): 12-14

Khan I, Qureshi MS, Akhta S and Ali I, 2016. Fertility improvement in cross-bred dairy cows through supplementation of vitamin E as antioxidant. Pakistan J Zool, 48(4): 923-930

Kuczynska B, Kapusta AL, Puppel KA, Nalecz-Tarwacka T, Budzinski A *et al.*, 2015. Relationships between milk beta-carotene concentrations and the cytological quality of cow's milk. Ann Wars Univ Life Sci-SGGW, Anim Sci, 54(1): 45-49

Oldham ER, Eberhart RJ and Muller LD, 1991. Effects of supplemental vitamin A or β-carotene during the dry period and early lactation on udder health. J Dairy Sci, 74: 3775-3781, doi: 10.3168/jds.S0022-0302(91)78569-X

Oliveira RC, Guerreiro BM, Junior NM, Araujo RL, Pereira *et al.*, 2015. Supplementation of prepartum dairy cows with β-carotene. J Dairy Sci, 98(9): 6304-6314, doi: 10.3168/jds.2014-9037

Panda N and Kaur H, 2008. Effect of vitamin E supplementation on mastitis and milk quality in buffaloes. Indian J Anim Nutr, 25: 118-124

Panda N, Kaur H and Mohanty TK, 2005. Reproductive performance of dairy buffaloes supplemented with varying levels of vitamin E. Asian-Aust J Anim Sci, 19(1): 19-25, doi: 10.5713/ajas.2006.19

Politis I, Bizelis I, Tsiaras A and Baldi A, 2004. Effect of vitamin E supplementation on neutrophil function, milk composition and plasmin activity in dairy cows in a commercial herd. J Dairy Res, 71: 273, doi: 10.1017/S002202990400010X

Pottier J, Focant M, Debier C, De Buysser G, Goffe C *et al.*, 2006. Effect of dietary vitamin E on rumen biohydrogenation pathways and milk fat depression in dairy cows fed high-fat diets. J Dairy Sci, 89(2): 685-692, doi: 10.3168/jds.S0022-0302(06)72131-2

Singh AK, Bhakat C, Kumari T, Mandal DK, Chatterjee A *et al.*, 2020. Influence of pre and postpartum alpha-tocopherol supplementation on milk yield, milk quality, and udder health of Jersey crossbred cows at tropical lower Gangetic region. Vet World, 13: 2006-2011, doi: 10.14202/vetworld.2020.2006-2011

SPSS, 2010. Statistical packages for Social Sciences, Version 16, SPSS Inc, Illinois, USA

Van Soest PJ, Robertson JB and Lewis BA, 1991. Methods for dietary fiber, neutral detergent fiber and non-starch polysaccharides in relation to animal nutrition. J Dairy Sci, 74: 3583-3597, doi: 10.3168/jds.S0022-0302(91)78551-2

Weiss WP, Hogan JS and Smith KL, 1994. Use of α-tocopherol concentrations in blood components to assess vitamin E status to dairy cows. Agric Practice, 15: 5-8

Wilde D, 2006. Influence of macro and micro minerals in the peri-parturient period on fertility in dairy cattle. Anim Reprod Sci, 96(3-4): 240-249, doi: 10.1016/j.anireprosci.2006.08.004

Received- 27.08.2022, Accepted- 20.10.2022, Published- 05.11.2022 (Online), 01.12.2022 (Print)

Section Editor: Dr. A. K. Patra, Associate Editor