Volume: 2 Issue: 8 Pages: 15-22

Effect of Dosing Vitamin E and Selenium Nanoparticles in the Milk Components of Holstein Cows

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Article Info.
E-ISSN: 2583-6528
Impact Factor (SJIF): 5.231
Available online:
www.alladvancejournal.com

Received: 09/July/2023
Accepted: 18/Aug/2023

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Abstract

This experiment was carried out at the cows Al-Khalis station in Habhab district in Diyala Governorate, during the period from 1-10/2022 to 1-5/2023 using 16 Holstein multiparous cows from milk-producing cows raised at the station. The animals were randomly divided into four treatments in the last two months of pregnancy and were as follows: T1: four cows without dosing (control treatment). T2: four cows were vaccinated 3000 IU vitamin E/cow per day. T3: three cows were tested for the nanoparticle selenium in an amount of 5 mg/cow per day. T4: five cows were spayed with a combination of vitamin E 3000 IU and nano-selenium in an amount of 5 mg/cow per day. Vitamin E is Alfa tocopherol acetate powder packed in capsules after weighing and dosed directly into the animal's mouth, as well as selenium nanoparticles packed in a capsule and dosed to cows once a day in the morning. The results showed that treatment T2 exceeded the percentage of fat in milk, reaching 5.60% compared to the other dosing treatment T3 and T4, which amounted to 2.46 and 2.82%, respectively, 10 days after birth, while there were no significant differences between the treatments in the rest of the milk components, represented by the percentage of protein, lactose sugar, ash, non-fat solids and milk density, while the degree of milk freezing there was a significant superiority of treatment T1, while treatment T4 exceeded the concentration of vitamin E and selenium in milk, reaching 2.89 Ppm for vitamin E and amounted to 4.05 ppm for selenium compared to the T1 control, which recorded 0.94 and 1.98 ppm for vitamin E and selenium, respectively, and a significant increase occurred in The concentration of vitamin E in milk for the T2 treatment was recorded at 2.43 Ppm, while the T3 treatment recorded a significant increase in the concentration of selenium in milk, where it was recorded at 3.18 Ppm compared to the control treatment mentioned above, we conclude from the current study that dosing cows with vitamin E or selenium nanoparticles in previous quantities has a significant role in increasing the levels of these elements in the milk produced from cows, which increases its nutritional value.

Keywords: Holstein cows, vitamin E, Nano-selenium, milk components.

Introduction

Milk is one of the foods rich in nutrients, as it contains protein, fat, lactose sugar, vitamins and minerals, which are essential in a balanced diet (Simo *et al*., 2016) [1]. It reduces the risk of osteoporosis, cardiovascular diseases and Type II diabetes and is the best complement to a healthy meal (Gulseven, 2017) [2].

The development of industry, the increase in population growth and the standard of living have led to a constantly increasing demand for nutrients, including milk, around the world (Stellato *et al*., 2017 [3]; Aykın-Dinçer and Erbaş, 2019) [4]. A hundred years ago vitamin E was identified as an essential micronutrient for humans and animals, and since then many biological functions of vitamin E have been revealed including antioxidant and anti-inflammatory properties, however, it has been proven that the bioavailability and physiological functions of vitamin E are highly dependent on lifestyle, genetic factors and individual health conditions (Liao *et al*., 2022) [5].

Vitamin E is an essential fat-soluble nutrient whose deficiency leads to immunosuppression and oxidative stress (Qian *et al*., 2021) [6]. And also its deficiency increases the risk of developing diseases such as placental retention, uterine inflammation and mastitis in cows (Weiss, 2017 [7]; Naderi *et al*., 2017) [8]. The main function of vitamin E is to protect lipid peroxidation and get rid of free radicals *in vivo* in order to maintain the integrity of cell membrane function, in addition, vitamin E is also involved in modulating enzyme activities, gene and protein expression (Huang *et al*., 2018 [9]; Zingg, 2019 [10]; Ungurianu *et al*., 2021) [11]. Some recent studies have suggested an important role of vitamin E and some vitamins against acute infectious respiratory diseases (Pecora *et al*., 2020 [12]; Linneberg *et al*., 2021 [13]; Dharmalingam *et al*., 2021) [14].

Selenium (Se) is also an essential mineral in food due to its role in health and Prevention of eating disorders, and it is used all over the world by adding it to ruminants that are kept under an intensive production system (Ahmadi *et al*., 2018 [15]; Ahmadi *et al*., 2019 [16]; Pan *et al*., 2021) [17]. Selenium also plays an important role in the antioxidant status of animals, as it prevents oxidative stress (Kojouri *et al*., 2020 [18]; Nabi *et al*., 2020; Han *et al*., 2021) [41,20]. Selenium supplements not only improve the efficiency of the antioxidant system, but can also enhance disease resistance (Amlan and lalhriatpuii, 2020) [21]. Nanoselenium as a feed additive has been tested mainly in domestic animals, since in ruminants it is possible that nanoselenium increases rumen fermentation and digestion of food, promotes body growth and weight gain, reduces oxidative stress, improves serum antioxidant enzymes and maintains the reproductive physiology of the animal (Abdelnour *et al*., 2021) [22]. There are numerous studies in the widespread use of selenium Se and vitamin E for their antioxidant and immunoregulatory properties and its important role in the health of cattle (Bordignon *et al*., 2019) [23]. It also significantly enhanced the growth rate of calves by reducing oxidative stress during the prenatal and postpartum period of cows (Volpato *et al*., 2018) [24]. Milk cows suffer from physiological and oxidative stress before and after birth, which affects births and milk production, and vitamin E and selenium, as is known, have several benefits in improving productive and health performance as well as being effective antioxidants (Abuelo *et al*., 2019) [25]. Due to the lack of studies in Iraq showing the effect of nanoselenium and vitamin E on milk cows, therefore, the current study aimed to demonstrate the effect of dosing vitamin E and nanoselenium for milk cows after placing them in capsules in the last two months of pregnancy and two months after delivery on a daily basis to indicate the extent of their role in improving milk components and their concentration in milk for cows.

Materials and Methods

The experiment was carried out at the cows Al-Khalis station in Hebah district-AL-Khalis district in Diyala Governorate, which is 5 km away from the city, during the period from 1-10/2022 to 1-5/2023 using 16 Holstein cows from the milk-producing cows raised at the station and their ages were between 3-8 years. The cows were placed in semi-polygonal sheds 75 m long and 25 m wide containing feeders for the cow to eat from, which have an iron clamp designed so that the cow's head enters the feed to eat the feed, and the barn is equipped with self-filling non-rusting water basins dictated by water (liquefaction water) and it is equipped for the cows to drink from whenever they want the floor of the barn is a concrete casting.

The quality of the feed and its quantity varied according to the season and depending on the availability of feed materials, as the green feed represented by alfalfa is provided in bales on two meals in the amount of approximately 20-25 kg per cow and the dry coarse feed represented by hay material is served on two meals as well morning and evening after the concentrated feed and in the amount of 4-5 kg per cow in the herd, while the concentrated feed is purchased from Best Fed feed lab-Erbil-Makhmour road-The weight of each bag is 50 kg, the type of ingredients is known (alfalfa, wheat flour, bran, soybeans, oil, calcium carbonate, table salt, animal mix, molasses, sodium carbonate, Dicalcium Phosphate and antitoxin), but the proportions are unknown due to the specificity of the company's work. Chemical analysis of the proportions of the ingredients for this feed was carried out in the Nutrition Laboratory of Graduate Studies at the Faculty of Agricultural Engineering Sciences/University of Baghdad.

The source of vitamin E that was used in this experiment is alpha tocopherol acetate powder, it is packed in capsules after weighing a certain amount and is dosed directly into the mouth of the animal in an amount of 3000 IU/cow/Day and was imported through a pure platform electronic shopping program of American origin, while the source of selenium that was used in the experiment is from the company its purity is up to 99%, which is of American origin contains all the information, weighing each 5 mg of it, packed in a capsule and dosed for cows once a day in the morning (5 mg/cow/Day).

The Study was conducted on 16 Holstein Cows, the Animals were Divided into four Treatments Randomly in the Last Two Months of Pregnancy and were as Follows

T1: Four cows without dosing (control treatment).

T2: Four cows were dosing 3000 IU vitamin E/cow per day.

T3: Three cows were dosing for the nanoparticle selenium in an amount of 5 mg/cow per day.

T4: Five cows were dosing with a combination of vitamin E 3000 IU and Nano selenium in an amount of 5 mg/cow per day.

Studied Qualities

Measuring the Proportions of Milk Components

The proportions of milk components were measured using the milk analyzr milk component analyzer in the dairy laboratory affiliated to the AL-Khalis cows station, where 50 ml of milk sample was placed for each cow after the milk samples were collected during milking in cans that are tightly covered, and then the samples are placed for freezing until they are analyzed in time for the analysis, they are thawed gradually and the bottles from the ratio (fat, protein, lactose sugar, Ash, milk density, non-fat solids and freezing point) of milk These components were estimated in the milk every month for each cow after calving.

The Concentration of Vitamin E and Selenium in Milk

Estimation of Vitamin E

Vitamin E can then be concentrated in milk in a laboratory of the Ministry of Science and technology in Baghdad, where 10 ml of the milk sample was immersed in 50 ml of ethanol for 30 minutes in a water bath at 85 degrees Celsius. The solution is left to cool and then filtered into a separating funnel. Add 10 ml Heptane and shake the solution for 5 minutes. After that, 20 ml of 1.25% sodium sulfate is added and the solution is shaken again for two minutes, allowed to separate into layers. The total tocopherol was determined by UV-VIS 545

Nm and a volume of 0.5 ml of tocopherols in ethanol was processed in the same way as the sample, using it as a standard (Santhosh *et al*., 2013) [26].

Selenium Estimation

The Selenium element under study was also estimated in a laboratory belonging to the Ministry of Science and technology in Baghdad, as reported in APHA (2017) [27], where the method included the following:

50 mL was taken for each sample of milk, the samples were placed inside a clean glass flask with a capacity of 100 ml, 5 ml of concentrated nitric acid was added to digest the samples, the flask was heated on the hot plate, and the heating continued on the hot plate until it reached the pre-drying stage, and 5 ml of concentrated nitric acid was added again to the samples while continuing to heat, in order to obtain a precipitate, leaving the solution to cool, and then we complete the volume to 25 filled with distilled water, the solution was filtered using a membrane filtration membrane filter (0.45 µm.) The samples were ready to estimate the concentrations of heavy metals in them, the absorbency of these digested samples was measured Using an atomic Absorption device (atomic Absorption) of the type SHEMADZU AA 7000.

Using the design of complete random sectors (RCBD) Random Complete Block Design as a sector to remove its influence from the experiment in analyzing the results according to the SPSS program, the significance of differences between the averages of transactions was tested according to the Duncan multi-range test at a significance level of 0.05, regardless of the significance of the F test in the table of analysis of variance.

Results and Discussion

Measuring the Proportions of Milk Components the Percentage of Fat in Milk

Table 1 shows. Effect of dosing vitamin E and selenium nanoparticles on the percentage of fat in the milk of Holstein cows, noting that there are significant differences between the transactions in the months of the experiment, ten days after birth, the T2 dosing treatment was superior, which recorded 5.60% compared to Treatments T3 and T4, they recorded a percentage of fat in the milk reached 2.46 and 2.82%, respectively, while there was no significant difference 4.27% two months after birth, the same is the case for the fourth treatment, which amounted to 4.30% two months after birth, while the T3 treatment was 2.33% one month after However, two months after the birth, we note that all the dosage transactions recorded a computational increase compared to transaction T1, which faced a computational decrease of 2.57 %. The reason for this may be attributed to the effect of vitamin E dosing, which led to an improvement in the fat content of milk and helps in the formation of fatty acids (Weiss and Wyatt, 2003) [28]. Which is consistent with what Santoshi *et al*. (2018) [19] came up with when adding a mixture of sodium citrate at a ratio of 25 g/kg feed and vitamin E in an amount of 1000 IU/Animal/Day for thirty Sahiwal Sahiwal multi-born cows, while it did not agree with what Wu *et al*. (2023) [23] came up with when adding vitamin E in an amount of 12000 IU/cow/day for multi-born Holstein cows, during three consecutive periods, each period included 18 one day.

Table 1: Effect of dosing vitamin E and selenium nanoparticles two months before and after calving on the percentage of fat in the milk of Holstein cows (mean ±standard error).

Duration Treatments	10 Days from Birth	A Month after Birth	Two Months after Birth
T1: Control without dosing	4.10 ±0.56ab	3.35±0.55	2.57±0.16
T2: Vitamin E dosage 3000 IU/cow	5.60± 0.96a	4.45±1.05	4.27±1.23
T3: 5mg/cow Nano Selenium dosing	2.46±0.34b	2.33±0.06	3.56±0.52
T4: Dosing vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	2.82±0.18b	3.06±0.76	4.30±0.82
Significant level	0.033	0.400	0.527

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

The Percentage of Protein in Milk

Table 2. It is noted that there were no significant differences between the Treatments during ten days after birth and one and two months after birth, and the values were similar in all dosing Treatments in the period of 10 days after birth, T2, T3 and T4 recorded a close value of 3.65, 3.66 and 3.38%, respectively, in addition to the control treatment T1, which amounted to 3.35%, and in the first month after birth, T2, T3 and T4 recorded a close value it amounted to 3.40, 3.16 and 3.06%, respectively, in addition to the control treatment T1, which amounted to 3.32%, as well as in two months after birth, the treatment T2, T3 and T4 recorded a value The approaches amounted to 3.10, 3.10 and 3.00%, respectively, in addition to the T1 control Treatments, which amounted to 3.27%.

Table 2: The effect of dosing vitamin E and selenium nanoparticles two months before and after birth on the percentage of protein in the milk of Holstein cows (mean ±standard error).

Duration Treatments	10 Days from Birth	A Month after Birth	Two Months after Birth
T1: Control without dosing	3.35±0.15	3.32±0.16	3.27±0.07
T2: Vitamin E dosage 3000 IU/cow	3.65±0.23	3.40±0.14	3.10±0.00
T3: 5mg/cow Nano Selenium dosing	3.66±0.29	3.16±0.03	3.10±0.20
T4: Dosing vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	3.38±0.09	3.06±0.10	3.00±0.06
Significant level	0.694	0.382	0.142

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

The Percentage of Lactose Sugar in Milk

Table 3 is shown. The effect of dosing vitamin E and selenium nanoparticles on the percentage of lactose sugar in the milk of Holstein cows, it is noted from the table that there are significant differences between the Treatments during the two months after birth, as the control treatment without dosing exceeded and recorded 4.92%, which did not differ significantly from the second and third treatment, which recorded 4.62 and 4.63%, respectively, on the fourth treatment, which is the dosing of a mixture of vitamin E and selenium nanoparticles, which recorded 4.44%, also there are no significant differences between the transactions My Account increased in the first month of the trial for the T2 and T3 treatment, which amounted to 5.42 and 5.50% 10 days after birth, respectively، And the second treatment, which reached an arithmetic increase in the first month of the experiment, was observed for a treatment of 5.15% a month after birth. The results did not agree with the findings of Bagnicka *et al* (2017) [31] when selenium was added in an amount of 6 mg selenium/cow/day over 90 days in Polish Holstein-Frisian cows. With the findings of Ianni *et al* (2019) [32] when selenium was added at a rate of 0.45 mg/kg of feed over a period of 63 days to the suspension of Friesian cows during the milk production period. While the results agreed with what Zhang *et al*. (2020) [33] found when adding selenium at three levels, low by 0.1 mg selenium/kg feed, medium by 0.2 mg selenium/kg feed and high by 0.3 mg selenium/kg feed.

Table 3: Effect of dosing vitamin E and selenium nanoparticles two months before and after birth on the percentage of lactose sugar in milk% of Holstein cows (mean ±standard error).

Duration Treatments	10 days from Birth	A month after Birth	Two months after Birth
T1: Control without dosing	5.07±0.22	5.17±0.28	4.92±0.11a
T2: Vitamin E dosage 3000 IU/cow	5.42±0.39	5.15±0.22	4.62±0.04ab
T3: 5mg/cow Nano Selenium dosing	5.50±0.41	4.64±0.03	4.63±0.31ab
T4: Dosing vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	5.06±0.13	4.64±0.15	4.44±0.05b
Significant level	0.783	0.280	0.073

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

The Percentage of Ash in Milk

It can be seen from Table 4. Effect of dosing vitamin E and selenium nanoparticles on the percentage of ash in the milk of Holstein cows, showing no significant differences between the coefficients in the percentage of ash ten days after birth, one month after birth and two months after birth, but an arithmetic increase was observed after 10 days after birth for the T3 dosing treatment, which amounted to 0.80% compared to the T1 control treatment, which recorded the lowest percentage of ash in milk was 0.67%, while it decreased at the T2 and T4 dosing treatments, reaching 0.77 and 0.72%, respectively. A month after the birth, an arithmetic increase was observed for the T2 treatment, which recorded 0.72%, compared to the T4 dosage treatment, which recorded an arithmetic decrease in the percentage of ash in milk of 0.64%, while the T1 and T3 Treatments recorded the same value of 0.70 and 0.70%, respectively.

Table 4: Effect of dosing vitamin E and selenium nanoparticles two months before and after birth on the percentage of ash in milk% of Holstein cows (mean ±standard error).

Duration Treatments	10 Days from Birth	A Month after Birth	Two Months after Birth
T1: Control without dosing	0.67±0.02	0.70±0.04	0.67±0.02
T2: Vitamin E dosage 3000 IU/cow	0.77±0.04	0.72±0.02	0.65±0.02
T3: 5mg/cow Nano Selenium dosing	0.80±0.05	0.70±0.00	0.63±0.03
T4: Dosing of vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	0.72±0.02	0.64±0.02	0.60±0.00
Significant level	0.261	0.246	0.210

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

The Percentage of non-fat Solids in Milk

Table 5 is showed. Effect of dosing vitamin E and selenium nanoparticles on the percentage of non-fat solids in the milk of Holstein cows, and the table shows that there are significant differences between the transactions during two months after birth in the percentage of non-fat solids, as it shows that the treatment T1 has increased significantly and recorded the highest percentage of non-fat solids amounting to 9.00% compared to the dosing treatment T4, which recorded a percentage of non-fat solids in milk amounting to 8.16%, while it is noted that the treatment T2 and T3 have recorded a significant increase compared to treatment T4, amounting to 8.42 and 8.43%, respectively, while no significant differences were observed at ten days from birth and a month after birth, but an arithmetic rise of 10 is noted The days after delivery of the dosing T2 and T3 treatment, which recorded the highest percentage of non-fat solids at 9.97 and 10.03%, respectively, the dosing T4 treatment recorded a value of 9.28%, while the T1 transaction recorded the lowest value compared to all dosing Treatments at 9.25%, an arithmetic increase was also observed in the two months after delivery for the T2 transaction, which amounted to 9.24% and the dosing T2 and T3 Treatments reached 8.46 and 8.44%, respectively, while Treatments T1 recorded a value of 9.20%. The results did not agree with the findings of Kay *et al*. (2005) [34] when adding vitamin E in an amount of 10,000 IU/cow on a daily basis during a period of 21 days after birth in Holstein-Frisian dairy cows, the results also did not agree with the findings of Khalifa *et al*. (2016) [33] injection of 10 ml of vitamin E and selenium mixture, as each 1 ml of the mixture contains vitamin E in an amount of 150 ml acetate and selenium in an amount of 1.67 mg Sodium selenite as an inorganic substance was injected into cows once every two weeks and for 60 days.

Table 5: Effect of dosing vitamin E and selenium nanoparticles two months before and after birth on the percentage of non-fat solids in milk% of Holstein cows (mean ± standard error)

Duration Treatments	10 Days from Birth	A Month after Birth	Two Months after Birth
T1: Control without dosing	9.25±0.38	9.20±0.43	9.00±0.19a
T2: Vitamin E dosage 3000 IU/cow	9.97±0.68	9.42±0.41	8.42±0.11ab
T3: 5mg/cow Nano Selenium dosing	10.03±0.74	8.46±0.20	8.43±0.58ab
T4: Dosing of vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	9.28±0.24	8.44±0.26	8.16±0.08b
Significant level	0.74	0.20	0.071

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

Milk Density

Table 6. It showed Effect of vitamin E and selenium nanoparticle dosing on the milk density of Holstein cows and notes that there are significant differences between the Treatments two months after birth, as transaction T1 recorded the highest value in milk density of 1.031 g/cm3, while all the addition coefficients (T2 vitamin E dosing Treatments, T3 selenium nanoparticle dosing Treatments, T4 vitamin E and selenium nanoparticle dosing transaction) had similar values recorded a slight decrease in milk density of 1.028, 1.028, 1.026 g/cm3, respectively, within ten days after delivery and a month after delivery, no significant differences were observed between all the coefficients. Also, all Treatments recorded similar ratios in the remaining periods, this may be due to the fact that when production decreases, the intensity increases.

Table 6: Effect of dosing vitamin E and selenium nanoparticles two months before and after birth on the milk density (g/cm^3) of Holstein cows (mean ±standard error)

Duration Treatments	10 Days from Birth	A Month after Birth	Two Months after Birth
T1: Control without dosing	1.031±0.001	1.031±0.001	1.031±0.000a
T2: Vitamin E dosage 3000 IU/cow	1.032±0.003	1.031±0.000	1.028±0.001b
T3: 5mg/cow Nano Selenium dosing	1.035±0.003	1.031±0.000	1.028±0.000b
T4: Dosing of vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	1.032±0.000	1.029±0.000	1.026±0.000b
Significant level	0.833	0.284	0.001

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

Degree of Immobilization

Table 7 showed Effect of dosing vitamin E and selenium nanoparticles two months before and after birth in the freezing point of milk for Holstein cows, as it was noted that there were no significant differences between the Treatments in all months of the experiment in freezing point, but an arithmetic increase was observed for Treatment T2, which recorded the highest freezing point value of 0.670-compared to the dosing Treatments T3 and T4, which recorded a clear decrease of 0.429-and 0.477-respectively, while Treatment T1 recorded a slight decrease compared to Treatment T2, reaching 0.601-for 10 days after delivery. During the month after the birth, an arithmetic increase is observed for the T2 Treatments, where a value of 0.615 was recorded-on all dosing Treatments T3 and T4 amounted to 0.549-and 0.541-respectively, in addition to the T1 Treatment without dosing amounted to 0.593-, as for the two-month period after the birth, it is noted that all dosing Treatments T2, T3 and T4 recorded a convergent arithmetic increase of 0.542-and 539-0.And 0.521-respectively, in addition to the T1 Treatment, which registered a slight increase of 0.587 –

Table 7: Effect of dosing vitamin E and selenium nanoparticles two months before and after birth on the freezing point(-C degree) of milk for Holstein cows (mean ± standard error)

Duration Treatments	10 Days from Birth	A Month after Birth	Two Months after Birth
T1: Control without dosing	0.601±0.028	0.593±0.033	0.587±0.019
T2: Vitamin E dosage 3000 IU/cow	0.670±0.047	0.615±0.038	0.542±0.014
T3: 5mg/cow Nano Selenium dosing	0.429±0.183	0.549±0.008	0.539±0.045
T4: Dosing of vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	0.477±0.104	0.541±0.023	0.521±0.010
Significant level	0.479	0.393	0.264

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

Concentration of Vitamin E and Nano-selenium in Milk

It can be seen from Table 8. The presence of significant differences between the coefficients in the concentration of vitamin E in milk, as the treatment of dosing the mixture of vitamin E and nano-selenium exceeded and was 2.89 Ppm on all Treatments, as well as the treatment of dosing vitamin E surpassed the two Treatments (control, nano-selenium dosing), but no significant differences were observed between the treatment of control and nano-selenium dosing, as for the concentration of nano-selenium, the treatment of dosing the mixture of vitamin E and nano-selenium also exceeded, as it was 4.05 Ppm on all Treatments (control without dosing, vitamin E supplementation treatment) while no significant differences were observed between the control treatment and the Dosing of vitamin E, the reason for this may be attributed to vitamin E and selenium, which work synergistically in maintaining the body's condition as an antioxidant, which increased their concentration in milk for the T4 treatment, and dosing of vitamin E or selenium nanoparticles for cows increased the absorption of these elements in the body and increased the utilization of these elements and thus increased their concentration secreted in milk, which increased the nutritional value of that milk produced.

Also, some sources indicated that feeding cows with Selenium resulted in an increase in Se concentrations in blood and milk (Seboussi *et al*., 2016 [38]; Sun *et al*., 2017 [39]; Sun *et al*., 2019) [37]. These results are consistent with what Kay *et al*. (2005) [34] reached when supplementing vitamin E in an amount of 10,000 IU/cow on a daily basis for a period of 21 days after birth in Holstein-Frisian dairy cows. The results were consistent with the findings of Meglia *et al*. (2006) [40] when vitamin E was dosed at an amount of 1000 IU/day for three weeks before delivery and 14 days after delivery in dairy cows. The results also agreed with the findings of Han *et al* (2021) [41,20] when treating feed at two levels, the first treatment is a control treatment without addition and the second treatment is the addition of nano-Se selenium in an amount of 0.30 mg/kg feed in the milk of dairy Holstein cows during 30 days of the milk production period. I also agreed with what Netto *et al*. (2022) [42] came up with when adding a mixture of selenium in an amount of 2.5 mg/kg dry matter and vitamin E in an amount of 1000 IU/cow to the hangover of Holstein cows on a daily basis during the 12-week period of milk production.

Table 8: Effect of vitamin E and nano-selenium dosing of Holstein cows on the concentration of vitamin E and selenium in milk Ppm (mean ± standard error)

Duration Treatments	Concentration of Vitamin E Ppm	Selenium Concentration Se Ppm
T1: Control without dosing	0.94±0.02 c	1.98±0.06c
T2: Vitamin E dosage 3000 IU/cow	2.43±0.08 b	2.06±0.02c
T3: 5mg/cow Nano Selenium dosing	1.08±0.00 c	3.18±0.05b
T4: Dosing of vitamin E and Selenium nanoparticles in the amount of 3000 IU/cow and 5 mg/cow, respectively	2.89±0.01 a	4.05±0.03a
Significant level	0.000	0.000

*Averages with different letters within the same column differ from each other significantly at a probability level of 0.05 according to the Duncan multi-range test.

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