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Effect of Propylene Glycol and Calcium Propionate on Some Morphometric, Productive, Reproductive and Metabolic Parameters in Drenching Egyptian Water-Buffalo Heifers

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Abstract

Propylene glycol (PG) and calcium propionate (CP) in saline solutions were drenched twice weekly to 18 Egyptian water-buffalo heifers at the last 8 weeks of gestation, besides a control group. The overall means of both treatments (particularly CP) significantly increased ($P \leq 0.05$) the feed intake of all ingredients, live body weight, reproductive performance; teat diameter, size, side area, area and length; calve birth weight, as well as daily and total milk production compared with the control. Both treatments significantly ($P \leq 0.05$) reduced blood urea nitrogen, β-hydroxybutyrate and non-essential fatty acids' values; but increased blood insulin, T3, and T4, concentrations and T3/T4 ratio. The CP treatment elevated ($P \leq 0.05$) also blood glucose level and milk fat and protein but reduced milk lactose %. Sampling time affected significantly ($P \leq 0.05$) daily feed intake, milk fat and lactose % as well as blood Na, K, Ca, P, T4 and insulin values, in addition to body condition score, live body weight, and udder form.

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Introduction

Buffalo is the main source of milk in Egypt (Agricultural Economic Institute, 1997). Propionate is the primary precursor to glucose synthesis, and can provide 90% of the sugars to the animal (Bergman, 1975). Liu *et al.*, (2009) found that the addition of calcium propionate helps in the fermentation processes of rumen and maintains the balance of energy in lactating animals. Propylene glycol (PG) is a glycogen precursor that is both quickly absorbed from the rumen and converted to glucose (Nielsen and Ingvartsen, 2004). The PG is used in the treatment of ketosis caused by imbalances in the energy balance (McClanahan *et al.*, 1998). The PG has been shown to have a decreased effect of dry matter intake (Formigoni *et al.*, 1996). Postnatal doses of PG resulted in a significantly lower βHBA concentration in plasma (Butler *et al.*, 2006).Addition of gluconeogenic precursors, such as sodium or calcium salts of propionate, increased ruminal propionate production (Harmon and Avery, 1987). The transition period (three weeks before and after parturition) is an important period for animal health and the productive performance of milk;during this period significant change occur in the endocrine and metabolism of dairy animal (DeFrain *et al.*, 2005).Therefore, the objectives of the present study were assessing to analyze the effects of supplementing propylene glycol (PG) and calcium propionate (CP) as sources of energy to primiparous Egyptian buffaloes during the late gestation and early lactation on feed intake, body conditions score and body weight,some blood parameters, milk yield and milk composition, and udder measurements, i.e. on productive and reproductive performance and some metabolic parameters.

Materials and Methods

The objectives of this investigation were planned to study the effects of drenched 3 L of saline solution (NaCl 0.9%) without any additives, 300 ml of propylene glycol (PG) dissolved in 3L of a saline solution, and calcium propionate (CP) (300g calcium propionate dissolved in 3L of saline solution), respectively twice weekly by using esophagus feeder pump to all animals, on feed intake, udder measurements (preened post-partum), milk yield, milk composition, body condition score (BCS), blood parameters and reproductive performance of buffalo heifers.

Experimental animal

A total number of 18 Egyptian primiparous buffaloes clinically free of diseases and healthy at the last 8 weeks of gestation (before 2 month of late gestation), were used in this study. The buffalo heifers' average live body weight was 500 ± 25 kg and aged 35±5 months.

Buffalo heifers were divided into three groups (6 / each) based on live body weight (LBW), body condition scores (BCS) and expected calving. Heifers of the 1st group control (C) and 2nd group G_1(PG) were drenched 3 liters of a saline solution (NaCl 0.9%) without any additives and 300 ml of propylene glycol (PG) dissolved in 3 liters of a saline solution, respectively. The 3rd group G_2(CP),was drenched 335g of calcium propionate dissolved in 3 liters of a saline solution twice weekly through the esophagus using esophagus feeder pump to all animal.

Tested diets

The animals were fed individually concentrate feed mixture (CFM) which composes of (25% un decorticated cotton meal, 43% yellow corn, 25% wheat bran, 3.5% molasses, 2% limestone, 1% common salt and 0.5% minerals mixtures), berseem (clover, *Trifolumalexandrinum*) hay and rice straw which were adjusted consistent with the physiological and productive stage, whereas fresh water and mineral blocks were available as free choice. All heifers were fed daily at 9 a.m. and 4 p.m. the recommended requirements according to NRC (1985) as given in Table1and the chemical composition of the feed stuffs is illustrated in Table 2. Which composed of 28% underacted COHON seed meal. 30% wheat bran, 25% yellow corn. 10% rice bran 3.5% molasses, 2% limestone, 1% berseem hay (BH) and rice straw (RS) in order to meet the nutritional allowance of buffalo cows during the late gestation and early lection periods according to (NRC, 2001) while fresh water and minerals blocks were available as free choice.

The animals were fed on the following tested diets

The control group: C + BH + RS + CFM

The 1st treated group (G_1) BH + RS + CFM plus drenched saline solution and 300 ml propylene glycol.

The 2st treated group (G_2) BH + RS + CFM plus drenched saline solution and 300 g calcium propionate.

Composition of propylene glycol, calcium propionate and saline solution

Propylene glycol GPR $CH_3H_8O_2$ (GP). Min. assay; 99.5%, Alpha Chemicals Company.

Calcium propionate GPR $CH_6H_{10}O_4$ (GP). Alpha Chemicals Company, laboratory chemicals.

Saline solution consists of commercial sodium chloride (NaCl).

Body condition score (BCS) and Milk production

Throughout the feeding period (preened post-partum), body condition score (BCS) as changes in live body weight was recorded monthly for each animal. Milk yield was individually recorded twice daily in morning at 6 A.M. and afternoon at 5 P.M. hand milking throughout lactation period of 8 weeks. Representative milk samples (2x/day) were well mixed and analyzed weekly for estimating the fat %, protein % and lactose. About 10 ml by placing it in the refrigerator on a freezing degree were added to preserve the milk until analysis using Milko-scan (Milko-scane, 150, Italy).

Blood sampling

Blood samples were collected from jugular vein in heparinized test tubes at 28 days pre-partum, at calving and 8 weeks postpartum from all heifers after eating one hours in morning twice a weekly and then centrifuged at 4000 rpm/ 15 minutes for plasma separation, then stored at -20ºC until performing biochemical analysis. Protein fractions (total protein, albumin and globulin), kidney function parameters (creatinine and urea-N), liver function enzymes [aspartate amino transfers (AST) and alanine amino transferase (ALT), plasma beta-hydroxy butyrate (BHB)]activity and some minerals' concentration (Na, K, Ca and P) was estimated in the plasma using commercial kits (Sigma kit 310.A, Sigma Diagnostics). While the plasma concentrations of triiodothyronine (T_3) and thyroxin (T_4) hormones were evaluated by the radioimmunoassay (RIA) procedure using the coated tubes kits (Diagnostic Products Corporation, 105 Angeles, CA, USA).

Placental drop and uterine involution

The time required for complete placental in each animal was immediately calculated. The reproductive tract of each animal was rectally palpated once/ tow day post-partum and once / three day after that to assess the uterine involution. Live body weight of calves at birth was recorded individually after parturition.

Observation of estrus

Buffaloes of all groups were visually observed for estrus behavior using teaser bull introduced for 3 times/ day at 6 a.m., 12 a.m. and 4 p.m. The teaser bull was allowed to run with females for 30 minutes on each occasion. The following symptoms were used as indicators for estrus including response to teaser bull: bellowing, segregation and restlessness, standing female, frequent urination, response to finger massage, vaginal mucous discharge, sniffing the vulva following the female, resting chine on female rump, mounting the female and standing for mounting by bull.

Reproductive parameters

Post-partum first ovulation interval (PPFVI) was determined by subtracting four days from the time at which plasma progesterone concentration time of the 1st occurrence of standing post-partum estrus was recorded as post-partum first estrus interval (PPFOI). Buffaloes that were detected in standing estrus were served naturally from a fertile bull and then post-partum first service interval (PPFSI) was recorded. Rectal palpation was preformed 60 d after date of service for pregnancy diagnosis, thereafter length of days open (DO), service period (SP), and number of service per conception (NS/C) were recorded. Conception rate was calculated as percentage of buffaloes diagnosed pregnant proportional to the total number of buffaloes served. This was recorded for the 1st, 2nd and 3rd service. Gestation period (GP) was recorded as an interval from conception date to parturition date, and calving interval (CI) was computed as gestation period length plus days open.

Statistical analysis

Data were subjected to analysis of variance (ANOVA), using the General Linear Model procedure (GLM) of the SAS software package (2004). The used model was:

$$Yijk = \mu + Ai + Tj + ATij + eijk$$

Where, Yijk is an observation of performance traits.

Ai = is the fixed effect of group.
Tj= is the fixed effect of treatment.
ATij= the interaction between group and time.
eijk = is the random error.

Significant differences between the mean treatments were compared by using Duncan's test (1955).

Results and Discussion

Feed intake

Results in Table3 show the average daily dry matter (DM) intake for concentrate feed mixture (CMF), rice straw (RS) and berseem hay were significantly($P \leq 0.001$) highest in treatment PG, CP and control group and average total feed intake in pre-partum period and post-partum data are significantly($P \leq 0.001$) highest between treatment PG and CP than control group.It could be noticed that the increase in DM intake from the treatment groups was probably due to drenched PG and CP, hence to the milk production through experimental period. Similarly, results were obtained by Cozzi *et al.*, (1996), who found increased DM intake of dairy cows in the middle of lactation when it's fed of PG administration compared with control group. Some results were observed too by Miyoshi *et al.*, (2001), they drenched the propylene glycol to Holstein cows during early feeding, after drenched propylene glycol there was an increase in the dry matter intake. Generally, the feeding values during pre-partum and post-partum period of female buffalo are show in Table3.

Average daily feed intake

Average daily feed intake as dry matter (DM) from different feed stuffs as calculated from TDN and DCP in the diets are presented in Table 4.Female buffalo received the requirement of TDN and DCP according to (NRC 2001).

The feeding by drenched 3L of saline solution (NaCl 0.9%) without any addictive, 300ml of propylene glycol (PG) dissolved in saline solution and calcium propionate (CP), 300ml dissolved in 3L of saline solution, respectively produced twice weekly (Abdel-Latif *et al.*, 2016).

Blood biochemical parameters

The effect of PG and CP drenched by orally on concentration of total protein (TP), albumin (Al), globulin (Gl) and albumin / globulin ratio as well as liver function (AST, ALT, and BUN) with time pre-partum (Time 1), at birth (Time 2) and post-partum (Time3) of female buffalo of the experimental group at various ages are shown in Table5.

Concentration of total protein

Overall mean of concentration of total proteins in plasma of female buffalo to 3 time in Table5 was not affected significantly (P>0.05) by drenched PG and CP with saline solution, although there was little improvement with control group compared with treatment groups. Analysis of variance revealed insignificant (P>0.05) effect of group and time and its interaction with PG and CP drenched with saline solution dissolved by oral feeder pump of total protein in blood plasma was almost higher in control group than those treatments and the improved in time 2 and 3 with treatment group compared with control group. The obtained results are in agreement with those reported by Abdel-Latif *et al.*, (2016), who found that drenched PG and CP dissolved in saline solution showed insignificant change in TP concentration in blood plasma of female buffalo.

Concentration of albumin

Overall mean of concentration of albumin (Alb) did not significantly (P>0.05) differ in blood plasma in female buffalo in different experimental groups (Table5) but it tended to be higher in control group than in treatment group. Also, the effect of times and its interaction with treatment group was not significant (P>0.05).Regarding the change in (Alb) concentration at different times, it increased from time 1 to time 3 with CP group and overall mean between groups. These results revealed insignificant decreased concentration, but did not reach the significant level of differences. The obtained values of albumin concentration agree with that obtained by Gabr *et al.*, (2017).

Concentration of globulin

Overall mean of globulin (glo) concentration in blood plasma of female buffalo in PG treatment group was nearly similarly to that in CP treatment group but those were increased than control group. Results in Table5revealed significant ($P \leq 0.05$) increase with CP treatment and control group than PG treatment. The results agree with those of Abdel-Latif *et al.*, (2016).On the other hand, Gabr *et al.*, (2017) on primiparous buffalo found no differences in blood biochemical parameters as affected by drenched saline solution of 300 ml PG and 300 g CP twice weekly.

Concentration of albumin / globulin ratio

Ratio of concentration of Alb / glo in blood plasma in female buffalo was significantly differ ($P \leq 0.05$) with control group witch similar in significantly with PG treatment group than CP treatment group in overall mean (Table5) and analysis of variance showed significance ($P \leq 0.05$) with interaction (group x time). These results are in agreements with those of Abdel-Latif *et al.*, (2016). They treated multiparous Egyptian buffalo by drenched saline solution dissolved it is 300 ml PG and 300 g CP, respectively. Their results were significantly differed ($P \leq 0.05$) at 28 d pre-partum, at calving and 56d post-partum period.

Activity of AST and ALT, and BUN concentration

Aspartate aminotransferase (AST) and alanine aminotransferase (ALT) are enzymes recognized as indices of protein reserve mobilization in the organism in the case of negative energy balance. Activity of AST enzyme is show in Table6. Analysis of variance shows that ALT activity in blood plasma of female buffalo was differ significant (P<0.05), it increased in control group and CP treatment group than that fed PG treatment group. This trend was observed in all groups at all times studies. These findings of significant increase in activity of AST and ALT enzymes and BUN concentration as indicators for liver function as affected by drenched saline solution dissolved with propylene glycol PG and calcium propionate CP with animal received this treatment. This trend was obtained by Klebaniuk *et al.*, (2009) when their studies were carried out on dairy cows. The objective of the study was to determine the effect of the dose and application period of glycogenic additive (GA) to high-yield dairy cows at the periparturient period and early lactation the metabolic disorder incidence and some blood biochemical parameters. They were divided into 5 groups, a control one (K) and four experimental groups received diets with GA, calcium propionate and propylene glycol mixture in loose form at two levels (300 or 450 g / head / d) and during two different periods (4 or 7 weeks). The glycogenic supplement reduced AST activity and slightly elevated ALT activity. On the other hand, Gabr *et al.*, (2017) published that 12 primiparous Egyptian buffalo were received drenched saline solution 3L dissolved 300m (PG) and 300g (CP) twice weekly pre- and post-partum (2 month). The observed results in blood plasma AST and ALT values were within the normal ranges and did not affect both immunity status and liver function. Moreover, the treatments (PG) and (CP) affected the activity of BUN when compared with the control group. The results were significantly (P< 0.05) differed at 28d pre-partum, at- calving and 56d post-partum periods. Abdel-Latif *et al.*, (2016) found that PG feeding or orally drenched, indicating that blood urea nitrogen (BUN) decreased compared with control group (Lien *et al.*, 2010).

Concentration of T3, T4 and Insulin

The analysis of variance shows that concentration of hormones T3, T4, and insulin as well as glucose was affected significantly by feeding system of PG and CP, at different 3 times and their interaction; however, non-esterified fatty acids NEFA was increased with control group than those treatment group. The observation in Table7 showed significant (P<0.001) increase in blood plasma of female buffalo in PG and CP treatment group with T3, T4, T3/T4 ratio, and insulin hormones and glucose compared with control group. These findings indicated significant increase in concentration of different parameter in Table7 in blood plasma of female buffalo as affected by drenched saline solution 3L dissolved in 300m from PG and 300g from CP used feeder pump by oral for female buffalo. The same trend was obtained by Gabr *et al.*, (2017), they reported that effect of PG and CP as an energy source for primiparous Egyptian buffalo (8 weeks before delivery) on the production of milk and some blood biochemical parameters was studies, 18 cow's buffalo were used with average live body weight (plup) of 500± 25 kg and the age of 5 month ± 35. The animals were classified into three comparable groups. The animals in the first group served as control with 3L of saline solution (0.9% NaCl) without any additions the animals in group (PG) were soaked in 300ml of dissolved PG in 3L of brine and the third group CP animal were macerating 335g of CP dissolved 3L of saline solution. The authors showed that the treated animals with PG and CP showed better results than the control group in T3, since T4 showed the highest in all treatment groups. Concentration of insulin in blood plasma of female buffalo showed the same trend was obtained by Liu *et al.*, (2009) and Abdel-Latif *et al.*, (2016).

Concentration of glucose (mg/dl)

Concentration of glucose (Table 7) in blood plasma of female buffalo in experimental groups fed drenched saline solution (0.9 NaCl) dissolved in 300m PG and 300g CP for treatment group and the analysis of variance showed that concentration of glucose was significantly (P< 0.0001) increased with treatment groups than control group, being the values were 68.22, 70.78 and 47.11 g/dl, respectively. The same trend was obtained by Pickelt (2003).

Concentration of Beta hydroxybutyrate (βHB), g/mol.

Averages of three times of female buffalo as attacked by drenched saline solution (0.9 NaCl) dissolved in 300ml PG and 300g CP is presented in Table8.Analysis of variance of βHB enzyme revealed significant (P< 0.05) effect on treatment groups of female buffalo at time 1, time 2 and time 3.Data shown in Table8reflected significant (P< 0.05) increased with control group than those PG and CP treatment groups and no difference between treatment group of female buffalo fed PG and CP treatment by oral feeder pump, but the lowest value was with CP treatment in T1, T2 and T3, respectively.

The results are in agreement with those of Stokes and Goff (2001) when they fed cows with 9.5L + 0.68 kg of calcium propionate, 9.5L of water + 300ml (310g) propylene glycol (PG or CP) 9.5L of water non-treatment, respectively. The cows received a dose drench allocated within 4 hours of after birth health events were recorded during calving and the first 15 days of milk (DIM). They reported that plasma β-hydroxy-butyrate was lowest at birth but was not affected by the treatment. The same trend was obtained by Pickett (2003), DeFrain *et al.*, (2005) and Lu *et al.*, (2009).

Concentration of urea and creatinine

Analysis of variance showed none significant (P>0.05) difference between groups and interaction for female buffalo fed PG and CP treatment as a source of energy. Results observed in Table8during three times and overall mean for blood plasma urea and creatinine revealed the highest value in control group than those treatment groups. On the other hand, the lowest value was with animal fed PG and CP treatment than was in control group for female buffalo. Urea and creatinine showed no significant (P>0.05) differences between treatment and control groups. In contrast, βHB results showed significant differences between the treated groups and the control group. The concentrations of βHB in the PG group at CP group were lower compared with control group. These results were supported by the results obtained by Kristensen and Raun (2007) who stated that injecting 650g of propylene glycol into the rumen reduced plasma levels of βHB. Furthermore, the plasma concentration of βHB was linearly decreased with the complement of PG and CP.

At the same time, Hoedemaker *et al.*, (2004) showed that glycogen supplementation showed no significant effect on urea concentration when milking cows were supplemented with a loose mixture of calcium propylene and propylene glycol at two levels (300 and 450 g / day / day) for different periods of time. On the other hand, the results did not coincide with the results obtained by Moallem *et al.*, (2007) who found no discrimination in the plasma levels between the control and treatment groups. On the other hand, Ballard *et al.*, (2001) recorded a significant difference between animals supplied with energy supplements (beet pulp, sugar cane, propylene glycol and calcium propionate) and the group control in creatinine concentration in blood. Recently, the results are in agreement with Gabr *et al.*, (2017) who infused/drenched the animals with propylene glycol or calcium propionate.

Average weekly milk yield

Average weekly milk yield as actual milk yield of female buffalo in different experimental groups through eight successive lactation weeks are presented in Table 9.As found for average daily milk yield, as weekly milk yield was affected significantly (P<0.001), since it increased by treatment and lactation weak and affected significantly (P<0.001), but there was an improving effect of feeding PG and CP dissolved in 3L saline solution (0.9 NaCl) on Average weekly milk yield. Results present in Table 9 shown significant (P<0.001) increase in CP treatment 1st week than PG and control groups but different between different weeks for along experimental groups. Total milk yield weekly from 1st to 8th weeks showed significant (P<0.001) increased with treatment group (207.142, 183.417 and 2.250 kg milk in CP, PG and control group, respectively) these increased with CP treatment due to animals were fed calcium elements with energy, and importance of calcium element for milking animals. Analysis of variance showed the significant effect between treatment groups compared with control for lactation period from (1:8 weeks).

Average daily milk yield

Average daily milk yield as actual milk yield (ADMY) of female buffalo as effected by PG and CP treatment dissolved in 3L saline solution (0.9% Nacl) for fed buffalo cows by used oral pump through, the first to eight lactation weeks are shown in Table9.Analysis of variance revealed significant (P< 0.001) effect of feeding and lactation week on ADMY for female buffalo. Results in Table9 show that daily milk yield as ADMY was significantly (P<0.001) higher for female buffalo fed this treatment than those control group which not milking for along experimental after calving compared with affected treatment groups. Moreover, the treated animals received more energy compared with control group during the eight lactation weeks. The daily milk yield of female buffalo significantly (P<0.001) increased, being 3.275 and 3.699kg with PG and CP group than 0.040 for control group, respectively. The results are in agreement with those reported by Cozzi *et al.*, (1996), Pehrson *et al.*, (1998) and Lucci *et al.*, (1998). Also, Stokes and Goff (2001) reported that a filed study was conducted to assess the effect of drenched oral with addition of energy or calcium plus energy. The treatments were 9.5L + 0.68 kg of calcium propionate, 9.5L of water + 300ml (310g) propylene glycol (PG or CP) 9.5L of water non-treatment respectively. The cows received a dose drench allocated within 4 hours of after birth and again after 24 hours after birth. Health events were recorded during calving and the first 15 days of milk (DIM). They found that milk production was better in animals treated with PG than other treatment. The same results were published too with Gabr *et al.*, (2017).

Milk composition

Average milk fat percentage of female buffalo in different experimental groups at lactation weeks in Table10.Fat parentage in milk was affected significant (P<0.01) only by treatment group; however, the effect of lactation week with treatment group was significantly (P<0.05), it increased with treatment group (Table10). But its interaction with treatment group was not significantly (P>0.05).Overall percentage of milk fat was significantly (P<0.01) higher in CP group than in PG group and the results showed nearly the same percentage. Average percentage of fat in milk female buffalo in control group was no found because the female buffalo were not milking from parity No.1 and this action back to no effect of control group during lactation weeks. Generally, fat percentage in milk treatment group was improved and increased with CP group perhaps because the effect of using calcium propionate in CP group in feeding buffalo cows. The results agree with those of Gavanaand Motorga (2009) and Liu *et al.*, (2009).Analysis of variance revealed that there was significant effect with group and lactation weeks for buffalo cows fed treatment than control group (that not milked). On the other hand, the good trend found by Gabr *et al.*, (2017) when they fed multiparous buffalo cows pre-and post-partum 300ml and 300g PG and CP, respectively, dissolved in saline solution (0.9 NaCl), the results showed no differences in milk fat in milk female buffalo.

Milk protein and lactose percentage

Milk protein and lactose percentage of female buffalo in different treatment group during successive lactation weeks (Tables11 and 12, respevtively) was affected significantly by feeding PG and CP dissolved in saline solution (0.9 NaCl), and its interaction not effected significantly with lactation week. Percentage of protein and lactose in milk of female buffalo was significantly higher in PG and CP group as overall mean. Throughout lactation weeks, lactose percentage was the highest in milk of female buffalo in PG treatment during the 1st and 8th week than CP treatment group. During the period of lactation weeks, protein percentage was the highest in milk of female buffalo cows in PG treatment than CP treatment for a long periods and overall mean showed the significantly (P<0.05) increased value in treatment group. The same trend was found with DeFrian *et al.*, (2005) and Gavanaand Motorga (2009). On the other hand, Gabr *et al.*, (2017) found no significantly difference between the treatment groups.

Milk elements (Na, K, Ca, and P) percentage

Percentages of milk elements in buffalo cows' milk(Tables13, 14, 15 and 16) were not affected significantly (P>0.05) between treatment group for a long period from 1st and 8th weeks between PG and CP treatment group and different elements in milk buffalo cows were fed propylene glycol and calcium propionate dissolved in 3L saline solution (0.9 NaCl).Analysis of variance showed no significant (P>0.05) difference between treatment group and not significant also in overall mean for every milk element. The present results agree with those reported by Lien *et al.*, (2010) and Gabr *et al.*, (2017).

Body condition score (BCS)

Average body condition score (BCS) of female buffalo in experimental group during successive lactation weeks is presented in Table 17. Analysis of variance of BCS revealed significant (P<0.001) effect of PG and CP treatments and lactation weeks on BCS of female buffalo and interaction was significant (P<0.05) too. Overall mean of BCS was significantly (P<0.05) higher in buffalo cows of PG and CP treatment groups than of control group, being 4 vs. 4.029 and 2.830, respectively. Throughout different lactation weeks, female buffalo in CP showed the highest BCS with 1st to 4th weeks than that PG and control group but the highest BCS increase was in PG than those in CP and control groups beginning in 5th to 8th weeks. The lowest values were recorded with control group. The present results on female buffalo are in contrast to those obtained on dairy cows by Cozzi *et al.*, (1996) who found that BCS was improved by feeding PG that added with corn silage to CP (14.7%&41.1%, NDF DM). Lucci *et al.*, (1998) reported that in cows that fed two laboratories 300ml of

PPG 300ml and water control, the BCS was assessed in days 10.0, 15, 30, 45 and 60 relative to the date of birth and they found BCS had no effect in both treatments from birth to 60 day post-partum and the results were in agreement with those of Abdel-Latif *et al.*, (2016).

Live body weight (LBW)

Average weekly live body weight (LBW) of female buffalo during 1 to 8 weeks of the study as affected by PG and CP treatment groups is presented in Table 18. Analysis of variance of LBW revealed significant (P<0.05) effect PG and CP on LBW of female buffalo at pre-and post-partum from 1 and 8weeks of age. LBW was significantly differed between experimental groups. Between 1 and 8 weeks of age, average weeks of age average weekly of female buffalo showed pronounced increased in LBW for all groups. Female buffalo fed 300ml PG and 300g CP dissolved in saline solution (0.9 NaCl) the highest value as in CP treatment than those in PG and control groups. Similar results were obtained by Cozzi *et al.,* (1996). The diets were fed to brown Italian cow's rumen and was 0, 200 or 400g of day 1PG added with corn silage to CP (14.7%, 14.1% NDF DM). They found that using propylene glycol administration improved the body weight. Also, when feeding the PG originally drenched, the BCS and LBW was higher than the control group (MaciejAdamski *et al.,* 2011 and Abdel-Latif *et al.*, 2016).

Live body weight at pre-birth (kg)

Means and stander error of live body weight of buffalo heifers in different experimental groups at pre-birth shown in Table 19. Analysis of variance shows that the effect of PG and CP treatment on LBW of buffalo heifers was significant (P<0.001), since it increased with PG and CP treatment compared with control and moreover the increase was higher in CP treatment than those PG and control group. The same trend was in birth weight (kg) for experimental groups, for buffalo cows in the treatment groups.

Changes in LBW at calve birth (kg)

The trend of changes in LBW of calve birth (kg) among the experimental groupsis noticed from Table19. At calve birth, there was a significant (P<0.05) increase in PG and CP treatment than control group but the increase was better in CP treatment groups fed 300g CP dissolved in 3L saline solution containing (0.9 NaCl) than those PG treatment and control group. The same trend was obtained in live body weight and changes in LBW at calve birth (kg) with MaciejAdamski *et al.*, (2011) and Abdel-Latif *et al.*, (2016).They published that feeding propylene glycol by using 18 primiparous Egyptian buffalo that were selected during the last pregnancy at the end of 8 weeks of gestation and the period of early feeding with an average of 500± 25kg vivo and age 35± 5 month to evaluate the effect of PG and CP treatment group for animals fed experimental group. The animals were divided into 3 similarly groups (6/each) based on LBW, the animals were drenched 3 liters of saline solution (0.9% NaCl) without any other supplements; whereas in the second groups' animals (PG) were given 300ml of dissolved PG was drenched in 3 liters of saline solution, and in the third CP group, animals were supplemented with 335g of dissolved CP in 3 liters of brine. All treatments were given through the esophagus via esophagus deeding tube twice / week through the experimental period vivo weight of the primparous buffaloes and their calves at birth and the interval from the calves to the first estrus were recorded the treatment group. They concluded that it was cleared that PG and CP supplementation had a significant increase of LBW (kg) of primiparous buffaloes during the late gestation period (65.28days pre-partum) and pre-birth when compared with control group, as well as during the suckling period 28 and 58 days post-partum (Table 19).

Change in LBW at estrus (kg)

The trend of changes in LBW of female buffalo at estrus, average LBW of animals were affected by feeding for experimental group. The results are shown in Table 19, significant (P<0.001) higher LBW (kg) was increased of animal fed CP treatment than those PG treatment and control group. The values were 621.67, 596.83 and 535.5 kg, respectively. The results are in agreement with the findings of Miyoshi *et al.,* (2001), who fed 36 Holstein cows the PG and reported that PG improved first estrous cycle in control cows.

Birth weight of calves

Average birth weight of calves born in different treatment groups (Table20) was significantly (P<0.05) affected by treatment groups for buffalo cows compared with control group. These increased backs to effect of treatment. Analysis of variance showed significant (P<0.05) differences for animals treatment group and control group.

Table.1 Amounts of DM, TDN and CP required for pre-partum and post-partum feeding of buffalo heifers

Items	DM (Kg)	TDN (Kg)	CP (Kg)
8 weeks Pre-partum	10 – 11	5.63	1.21
8 weeks Milk Production (5.7% fat)			
3 – 5 Kg Milk	11 - 12	6.54	1.03

Table.2 Chemical composition (% on DM basis) of feed ingredients given to primiparous buffaloes along the period of experimental.

Chemical analysis	Dietary ingredients		
	CFM	Hay	Rice Straw
DM	91.20	86.70	88.40
OM	93.90	78.10	78.36
CP	15.70	15.50	2.96
CF	14.23	30.93	35.83
EE	3.13	2.13	2.23
NFE	60.84	38.54	37.34
Ash	6.10	12.90	21.64

Table.3 Calculated feeding value and energy (% on DM basis) of feed ingredients given to primiparous buffaloes along the period of experimental.

Feeding Value% of DM	CFM	Hay	Rice straw
TDN %	58.68	63.96	60.47
DCP %	11.52	11.32	- 0.71
DE (M Cal/kg DM) *	2.587	2.820	2.666
ME (M Cal/kg DM) *	2.1763	2.398	2.243
NE (M Cal/kg DM) *	1.438	1.447	1.362

Total digestible nutrients (TDN) = 129.39- 0.9419 (CF+ NFE); Digestible crude protein (DCP) = 0.9596 CP- 3.55; Digestible energy (DE) = 0.04409 (TDN %); Metabolizable energy (ME) = 1.01(DE) - 0.45; Net energy (NE) = 0.0245 (TDN %) - 0.12 (*: according to NRC, 2001).

Table.4 Effect of calcium propionate (CP) and propylene glycol (PG) on daily feed intake

	Time	All Treatment			Overall mean
		C	PG	CP	
CFM	Pre	4.16±0.05	4.44±0.05	4.45±0.05	4.35^{b}±0.03
	Post	4.21±0.03	5.60±0.03	5.82±0.03	5.21^{a}±0.01
Overall mean		4.19^{c}±0.03	5.02^{b}±0.03	5.13^{a}±0.03	
Hay	Pre	2.49±0.02	2.62±0.02	2.67±0.02	2.59^{b}±0.01
	Post	2.53±0.01	2.80±0.01	2.91±0.01	2.75^{a}±0.01
Overall mean		2.51^{c}±0.01	2.71^{b}±0.01	2.79^{a}±0.01	
RsS	Pre	4.16±0.04	4.36±0.04	4.45±0.04	4.32^{b}±0.02
	Post	4.21±0.02	4.66±0.02	4.85±0.02	4.58^{a}±0.01
Overall mean		4.19^{c}±0.02	4.51^{b}±0.02	4.65^{a}±0.02	

a-b: Means in the same column, within each feedstuff, superscripted with different letters, significantly (P≤0.05) differ.

Table.5 Least squares mean of some blood parameters (total protein – albumin – globulin - albumin/ globulin ratio) for female buffalo

	Group / Time	Control	Treatment group PG	Treatment group CP	Overall Mean
Total protein g/dl	TIME1	7.84±0.43	7.11±0.43	7.34±0.43	7.43^{a}±0.25
	TIME2	7.62±0.43	7.64±0.43	7.68±0.43	7.65^{a}±0.25
	TIME3	7.62±0.43	7.38±0.43	7.82±0.43	7.61^{a}±0.25
Overall Mean		7.7^{a}±0.25	7.38^{a}±0.25	7.61^{a}±0.25	
Albumin g/dl	TIME1	4.21±0.33	4.15±0.33	3.91±0.33	4.09^{a}±0.19
	TIME2	4.38±0.33	3.96±0.33	4.03±0.33	4.12^{a}±0.19
	TIME3	4.32±0.33	4.06±0.33	4.29±0.33	4.23^{a}±0.19
Overall Mean		4.30^{a}±0.19	4.06^{a}±0.19	4.08^{a}±0.19	
Globulin g/dl	TIME1	3.63±0.29	2.96±0.29	3.43±0.29	3.34^{a}±0.17
	TIME2	3.25±0.29	3.68±0.29	3.65±0.29	3.53^{a}±0.17
	TIME3	3.30±0.29	3.32±0.29	3.53±0.29	3.38^{a}±0.17
Overall Mean		3.39^{a} ±0.17	3.32^{a}±0.17	3.54^{a}±0.17	
albumin / globulin ratio	TIME1	1.17±0.15	1.42±0.15	1.15±0.15	1.25^{a}±0.09
	TIME2	1.36±0.15	1.08±0.15	1.19±0.15	1.21^{a}±0.09
	TIME3	1.32±0.15	1.23±0.15	1.22±0.15	1.26^{a}±0.09
Overall Mean		1.28^{a}±0.09	1.24^{a}±0.09	1.19^{a}±0.09	

a: No significant (P>0.05) differences.

Table.6 Least squares mean of some blood parameters (AST – ALT – BUN) for female buffalo

	Group / Time	Control	Treatment group PG	Treatment group CP	Overall Mean
AST,μ/l	TIME1	63.33±3.49	63.67±3.49	60.67±3.49	62.56^{a}±2.01
	TIME2	64.33±3.49	56.33±3.49	59±3.49	59.89^{a}±2.01
	TIME3	57.67±3.49	56±3.49	60±3.49	57.89^{a}±2.01
Overall Mean		61.78^{a}±2.01	58.67^{a}±2.01	59.89^{a}±2.01	
ALT, μ/l	TIME1	30±2.43	31.33±2.43	27.67±2.43	29.67^{a}±1.40
	TIME2	31±2.43	27±2.43	28±2.43	28.67^{a}±1.40
	TIME3	29.33±2.43	26.67±2.43	27.33±2.43	27.78^{a}±1.40
Overall Mean		30.11^{a}±1.4	28.33^{a}±1.4	27.67^{a}±1.40	
BUN,mg/dl	TIME1	20±1.70	12.67±1.70	13±1.70	15.22^{a}±0.98
	TIME2	20±1.70	14±1.70	13.33±1.70	15.78^{a}±0.98
	TIME3	19.33±1.70	13±1.70	15.67±1.70	16.00^{a}±0.98
Overall Mean		19.78^{a}±0.98	13.22^{b}±0.98	14^{b}±0.98	

a-b: Means in the same row superscripted with different letters, significantly (P≤0.05)differ.

Table.7 Least squares mean of some blood parameters (T3, T4, T3/T4 ratio, glucose, insulin, and NEFA) for female buffalo

	Group Time	Control	Treatment group		Overall Mean
			PG	CP	
T3,ng/ml	TIME1	3.36±0.28	5.08±0.28	4.80±0.28	4.41^{a}±0.16
	TIME2	3.09±0.28	5.36±0.28	5.02±0.28	4.49^{a}±0.16
	TIME3	2.93±0.28	5.40±0.28	5.14±0.28	4.49^{a}±0.16
Overall Mean		3.13^{b}±0.16	5.28^{a}±0.16	4.99^{a}±0.16	
T4,ng/ml	TIME1	15.653±0.646	19.130±0.646	18.863±0.646	17.88^{b}±0.37
	TIME2	16.693±0.646	18.797±0.646	19.223±0.646	18.24^{ab}±0.37
	TIME3	16.653±0.646	20.667±0.646	20.560±0.646	19.29^{a}±0.37
Overall Mean		16.33^{b}±0.37	19.53^{a}±0.37	19.55^{a}±0.37	
T3/ T4 ratio	TIME1	0.22±0.02	0.27±0.02	0.25±0.02	0.25^{a} ±0.01
	TIME2	0.19±0.02	0.28±0.02	0.26±0.02	0.24^{a}±0.01
	TIME3	0.18±0.02	0.26±0.02	0.25±0.02	0.23^{a}±0.01
Overall Mean		0.19^{b}±0.01	0.27^{a}±0.01	0.25^{a}±0.01	
Insulin U/ml	TIME1	5.90±1.75	10.27±1.75	11±1.75	9.06^{b}±1.01
	TIME2	6.79±1.75	13.85±1.75	18±1.75	12.88^{a}±1.01
	TIME3	5.65±1.75	12.23±1.75	11.57±1.75	9.81^{b}±1.01
Overall Mean		47.11^{b}±1.82	68.22^{a}±1.82	70.78^{a}±1.82	
Glucose mg/dl	TIME1	47.67±3.14	67±3.14	67.67±3.14	60.78^{a}±1.82
	TIME2	47.33±3.14	71±3.14	76±3.14	64.78^{a}±1.82
	TIME3	46.33±3.14	66.67±3.14	68.67±3.14	60.56^{a}±1.82
Overall Mean		47.11^{b}±1.82	47.11^{b}±1.82	70.78^{a}±1.82	
(NEFA) Eq/L	TIME1	381.33±27.78	289.3±27.78	261.3±27.78	310.67^{a}±16.04
	TIME2	377.67±27.78	271±27.78	223.7±27.78	290.78^{a}±16.04
	TIME3	374.33±27.78	255±27.78	256.3±27.78	295.22^{a}±16.04
Overall Mean		377.78^{a}±16.04	271.8^{b}±16.04	247.1^{b}±16.04	

a-b: Means in the same row superscripted with different letters, significantly (P≤0.05) differ.

Table.8 Least squares mean of some blood parameters (BHB – Urea – Creatinine) for female buffalo

	Item	Control	Treatment group		Overall Mean
			PG	CP	
Urea, mg/dl	TIME1	69.51±3.78	62.98±3.78	65.05±3.78	65.85±2.18
	TIME2	67.56±3.78	67.73±3.78	68.06±3.78	67.78±2.18
	TIME3	67.56±3.78	65.43±3.78	69.30±3.78	67.43±2.18
Overall Mean		68.21^{a}±2.18	65.38^{a}±2.18	67.47^{a}±2.18	
Creatinine, mg/dl	TIME1	0.71±0.04	0.64±0.04	0.66±0.04	0.67±0.02
	TIME2	0.69±0.04	0.69±0.04	0.69±0.04	0.69±0.02
	TIME3	0.69±0.04	0.67±0.04	0.7±0.04	0.69±0.02
Overall Mean		0.69±0.02	0.67±0.02	0.68±0.02	
Beta-hydroxybutyrate (βHB),g·mol	TIME1	26±2.38	19.33±2.38	14.67±2.38	20^{a}±1.37
	TIME2	26.33±2.38	18±2.38	16.33±2.38	20.22^{a}±1.37
	TIME3	25.67±2.38	15.67±2.38	13±2.38	18.11^{a}±1.37
Overall Mean		26^{a}±1.37	17.67^{b}±1.37	14.67^{b}±1.37	

a-b: Means in the same row superscripted with different letters, significantly (P≤0.05) differ.

Table.9 Least squares mean of period milk production (weekly) for female buffalo

Group / Weeks	Control	Treatment group	
		PG	CP
Pried	1.5^{b}±2.922	51.167^{a}±2.922	56^{a}±2.922
1	1.750^{b}±3.069	17.917^{a}±3.069	19.25^{a}±3.069
2	0.5^{b}±2.963	23.417^{a}±2.963	22.75^{a}±2.963
3	0^{b}±2.929	25.583^{a}±2.929	23.5^{a}±2.929
4	0^{b}±1.124	25.083^{a}±1.124	23^{a}±1.124
5	0^{b}±2.726	21.5^{a}±2.726	28.917^{a}±2.726
6	0^{b}±2.979	22.167^{a}±2.979	27.75^{a}±2.979
7	0^{b}±3.540	24.5^{a}±3.540	32.15^{a}±3.540
8	0^{b}±3.090	23.25^{a}±3.090	29.825^{a}±3.090
Daily	0.040^{b}±0.168	3.275^{a}±0.168	3.699^{a}±0.168
Total	2.25^{b}±9.395	183.417^{a}±9.395	207.142^{a}±9.395

a-b: Means in the same row superscripted with different letters, significantly ($P \leq 0.05$) differ.

Table.10 Least squares mean of milk fat % for female buffalo

Lactation / Weeks	Treatment group		Overall Mean
	PG	CP	
1	6.282±0.234	6.488±0.234	6.385^{b}±0.165
2	6.477±0.234	6.757±0.234	$6.617a^{b}$±0.165
3	6.695±0.234	6.887±0.234	6.791^{b}±0.165a
4	6.240±0.234	7.002±0.234	6.621^{b}±0.165a
5	6.532±0.234	7.022±0.234	6.777^{b}±0.165a
6	6.847±0.234	7.175±0.234	7.011^{a}±0.165
7	6.692±0.234	7.1±0.234	6.896^{b}±0.165a
8	6.768±0.234	7.137±0.234	6.953^{a}±0.165
Overall Mean	6.566^{b}±0.083	6.946^{a}±0.083	

a-b: Means in the same row or column superscripted with different letters, significantly ($P \leq 0.05$) differ.

Table.11 Least squares mean of milk total protein % for female buffalo

Lactation / Weeks	Treatment group		Overall Mean
	PG	CP	
1	4.805±0.113	4.310±0.113	4.558^{a}±0.080
2	4.765±0.113	4.423±0.113	4.594^{a}±0.080
3	4.715±0.113	4.462±0.113	4.588^{a}±0.080
4	4.578±0.113	4.562±0.113	4.57^{a}±0.080
5	4.712±0.113	4.665±0.113	4.688^{a}±0.080
6	4.657±0.113	4.748±0.113	4.703^{a}±0.080
7	4.687±0.113	4.710±0.113	4.698^{a}±0.080
8	4.672±0.113	4.728±0.113	4.7^{a}±0.080
Overall Mean	4.699^{a}±0.040	4.576^{b}±0.040	

a-b: Means in the same row superscripted with different letters, significantly ($P \leq 0.05$) differ.

Table.12 Least squares mean of milk lactose % for female buffalo

Lactation Weeks	Treatment group		Overall Mean
	PG	CP	
1	5.153±0.139	5.133±0.139	5.143[c]±0.098
2	5.615±0.139	5.568±0.139	5.592[b]±0.098
3	6.013±0.139	5.752±0.139	5.883[a]±0.098
4	6.1±0.139	5.830±0.139	5.965[a]±0.098
5	6.172±0.139	5.962±0.139	6.067[a]±0.098
6	6.107±0.139	5.828±0.139	5.968[a]±0.098
7	6.14±0.139	5.898±0.139	6.019[a]±0.098
8	6.125±0.139	5.862±0.139	5.993[a]±0.098
Overall Mean	5.928[a]±0.049	5.729[b]±0.049	

a-c: Means in the same row or column superscripted with different letters, significantly (P≤0.05) differ.

Table.13 Least squares mean of milk Na % for female buffalo

Lactation Weeks	Treatment group		Overall Mean
	PG	CP	
1	1.5867±0.0237	1.5583±0.0237	1.5725[c]±0.0168
2	1.6433±0.0237	1.6317±0.0237	1.6375[b]±0.0168
3	1.6950±0.0237	1.6667±0.0237	1.6808[ab]±0.0168
4	1.6467±0.0237	1.6933±0.0237	1.67a[b]±0.0168
5	1.6933±0.0237	1.715±0.0237	1.7042[a]±0.0168
6	1.7133±0.0237	1.725±0.0237	1.7192[a]±0.0168
7	1.7017±0.0237	1.7183±0.0237	1.71[a]±0.0168
8	1.7083±0.0237	1.7217±0.0237	1.715[a]±0.0168
Overall Mean	1.674[a]±0.008	1.679[a]±0.008	

a-c: Means in the same column superscripted with different letters, significantly (P≤0.05) differ.

Table.14 Least squares mean of milk K % for female buffalo

Lactation Weeks	Treatment group		Overall Mean
	PG	CP	
1	63.220±0.945	62.088±0.945	62.654[c]±0.669
2	65.48±0.945	65.085±0.945	65.283[b]±0.669
3	67.558±0.945	66.373±0.945	66.966[ab]±0.669
4	65.708±0.945	67.450±0.945	66.579[ab]±0.669
5	67.527±0.945	68.383±0.945	67.955[a]±0.669
6	68.242±0.945	68.762±0.945	68.502[a]±0.669
7	67.885±0.945	68.572±0.945	68.228[a]±0.669
8	68.063±0.945	68.667±0.945	68.365[a]±0.669
Overall Mean	66.71[a]±0.334	66.923[a]±0.334	

a-c: Means in the same column superscripted with different letters, significantly (P≤0.05) differ.

Table.15 Least squares mean of milk Ca% for female buffalo

Lactation Weeks	Treatment group		Overall Mean
	PG	CP	
1	23.137±0.346	22.722±0.346	22.929^{c}±0.245
2	23.965±0.346	23.817±0.346	23.891^{b}±0.245
3	24.725±0.346	24.290±0.346	24.508^{ab}±0.245
4	24.045±0.346	24.683±0.346	24.364^{ab}±0.245
5	24.713±0.346	25.027±0.346	24.870^{a}±0.245
6	24.975±0.346	25.165±0.346	25.070^{a}±0.245
7	24.847±0.346	25.095±0.346	24.971^{a}±0.245
8	24.91±0.346	25.130±0.346	25.020^{a}±0.245
Overall Mean	24.415^{a}±0.122	24.491^{a}±0.122	

a-c: Means in the same column superscripted with different letters, significantly (P≤0.05) differ.

Table.16 Least squares mean of milk phosphorous% for female buffalo

Lactation Weeks	Treatment group		Overall Mean
	PG	CP	
1	36.873±0.552	36.217±0.552	36.545^{c}±0.390
2	38.195±0.552	37.967±0.552	38.081^{b}±0.390
3	39.410±0.552	38.717±0.552	39.063^{ab}±0.390
4	38.328±0.552	39.342±0.552	38.835^{ab}±0.390
5	39.390±0.552	39.888±0.552	39.639^{a}±0.390
6	39.807±0.552	40.110±0.552	39.958^{a}±0.390
7	39.598±0.552	40±0.552	39.799^{a}±0.390
8	39.7±0.552	40.055±0.552	39.878^{a}±0.390
Overall Mean	38.913^{a}±0.195	39.037^{a}±0.195	

a-c: Means in the same column superscripted with different letters, significantly (P≤0.05) differ.

Table.17 Least squares mean of body condition score (BCS) for female buffalo

Group weeks	Control	Treatment group		Overall Mean
		PG	CP	
Pre-partum	2.735±0.119	3.344±0.119	3.669±0.119	3.249^{b}±0.068
Post-partum	2.825±0.168	3.817±0.168	3.957±0.168	3.533^{a}±0.097
2	2.880±0.168	3.9±0.168	3.953±0.168	3.578^{a}±0.097
3	2.928±0.168	3.978±0.168	3.947±0.168	3.618^{a}±0.097
4	2.853±0.168	4.147±0.168	4.125±0.168	3.708^{a}±0.097
5	2.793±0.168	4.225±0.168	4.163±0.168	3.727^{a}±0.097
6	2.823±0.168	4.186±0.168	4.144±0.168	3.718^{a}±0.097
7	2.809±0.168	4.206±0.168	4.154±0.168	3.723^{a}±0.097
8	2.822±0.168	4.197±0.168	4.152±0.168	3.723^{a}±0.097
Overall Mean	2.830^{b}±0.054	4^{a}±0.054	4.029^{a}±0.054	

a-b: Means in the same row or column superscripted with different letters, significantly (P≤0.05) differ.

Table.18 Least squares mean of weight (kg) for female buffalo

Group / Weeks	Control	Treatment group PG	Treatment group CP	Overall Mean
Pre-partum	498.833±4.815	523.083±4.815	533.583±4.815	518.5^{c}±2.780
Post-partum	499.667±6.809	552.167±6.809	573.667±6.809	541.833^{b}±3.931
2	499.667±6.809	552.167±6.809	573.667±6.809	541.833^{b}±3.931
3	499.667±6.809	552.167±6.809	573.667±6.809	541.833^{b}±3.931
4	499.667±6.809	552.167±6.809	573.667±6.809	541.833^{b}±3.931
5	511.667±6.809	567.333±6.809	590.167±6.809	556.389^{a}±3.931
6	511.667±6.809	567.333±6.809	590.167±6.809	556.389^{a}±3.931
7	511.667±6.809	567.333±6.809	590.167±6.809	556.389^{a}±3.931
8	511.667±6.809	567.333±6.809	590.167±6.809	556.389^{a}±3.931
Overall Mean	504.907^{c}±2.206	555.676^{b}±2.206	576.546^{a}±2.206	

a-c: Means in the same row or column superscripted with different letters, significantly (P≤0.05) differ.

Table.19 Least squares mean of some reproductive performance for female buffalo

Group / Item	Control (CO)	PG	CP
Week 8 pre-partum	487.17^{b}±4.58	500.17a^{b}±4.58	507.00^{a}±4.58
Week 4 pre-partum	510.50^{b}±5.05	546.00^{a}±5.05	560.17^{a}±5.05
Pre-birth kg	545.33^{c}±5.96	596.33^{b}±5.96	621.33^{a}±5.96
Birth weight kg	499.67^{c}±5.52	552.17^{b}±5.52	573.67^{a}±5.52
Calve Weight kg	27.08^{b}±1.24	30.42^{b}±1.24	34.25^{a}±1.24
After 4 weeks kg	511.67^{c}±5.53	567.33^{b}±5.53	590.17^{a}±5.53
After 8 weeks kg	524^{c}±5.84	582.83b±5.84	604.67^{a}±5.84
At estrus kg	535.50^{c}±5.03	596.83^{b}±5.03	621.67^{a}±5.03

a-c: Means in the same row superscripted with different letters, significantly (P≤0.05) differ.

Table.20 Least squares mean of birth weight kg of calves in buffalo cows in treatment groups

Item	Control (Co)	Treatment group PG	Treatment group CP
Calve birth (kg)	27.25^{b}±1.21	30.42^{b}±1.21	34.25^{a}±1.21

a-b: Means denoted with different superscripts are significantly different at (P<0.05)

Table.21 Least squares mean of treatment groups on post-partum duration of placental drop and uterine involution in buffalo cows in treatment groups

Item	Control (Co)	Treatment group PG	Treatment group CP
Placenta drop interval / Hour	12.25^{a}±1.21	6.42^{b}±1.21	4.92^{b}±1.21
Uterine involution (Day)			
Normal uterine position	40.50^{a}±2.49	29.33^{b}±2.49	26.33^{b}±2.49
Cervical closure	32.83^{a}±2.42	22.83^{b}±2.42	20.33^{b}±2.42
Uterine horns symmetry	52.83^{a}±2.13	34.83^{b}±2.13	31.67^{b}±2.13

a-b: Means denoted with different superscripts within the same row are significantly different at (P<0.05)

Table.22 Least squares mean of post-partum first estrus interval (PPFOI) and service period length (SP) in buffalo cows in treatment groups.

Item		Treatment group	
	Control (Co)	PG	CP
PPFOI (day)	82.33^{a}±4.70	53.50^{b}±4.70	48.50^{b}±4.70
Servile Period	46.17^{a}±8.34	9.83^{b}±8.34	3.67^{b}±8.34

a-b: Means denoted with different superscripts within the same row are significantly different at (P<0.05)

Table.23 Least squares mean of number of services per-conception NS/C, days open (DO), gustation period (GP) and calving interval (CI)

Item		Treatment group	
	Control (Co)	PG	CP
Number of service preconception	2.17^{a}±0.24	1.33^{b}±0.24	1.17^{b}±0.24
Days open, d	128.50^{a}±9.81	63.33^{b}±9.81	52.17^{b}±9.81
Gestation period, d	307.83^{a}±3.62	314.83^{a}±3.62	311.83^{a}±3.62
Calving interval, d	436.33^{a}±9.41	378.17^{b}±9.41	364^{b}±9.41

a-b: Means denoted with different superscripts within the same row are significantly different at (P<0.05)

Table.24 Least squares mean of distances between teats for female buffalo

		Control	Treatment Group		Overall Mean
			PG	CP	
Distances between teats	Pre-partum	7.927±0.308	8.494±0.308	7.917±0.308	8.113^{b}±0.178
	Post-partum	8.518±0.308	9.839±0.308	8.5±0.308	8.952a±0.178
Overall Mean		8.222^{b}±0.218	9.167^{a}±0.218	8.208^{b}±0.218	
Distance 2=d between right behind to lift behind	Pre-partum	9.959±0.412	8.750±0.412	8.25±0.412	8.986^{b}±0.238
	Post-partum	10.959±0.412	10.372±0.412	9.5±0.412	10.277^{a}±0.238
Overall Mean		8.222^{b}±0.218	9.167^{a}±0.218	8.208^{b}±0.218	
Distance 3=d between right front to right behind	Pre-partum	9.751±0.374	9.883±0.374	9.072±0.374	9.569^{b}±0.216
	Post-partum	10.051±0.374	11.561±0.374	10.006±0.374	8.986^{b}±0.238
Overall Mean		10.459^{a}±0.291	9.561^{b}±0.291	8.875^{b}±0.291	
Distance 4=dB tween right front to right behind	Pre-partum	11.5±0.429	9.761±0.429	10.467±0.429	10.576^{b}±0.247
	Post-partum	12.479±0.429	10.994±0.429	11.356±0.429	11.610^{a}±0.247
Overall Mean		9.901^{b}±0.264	10.722^{a}±0.264	9.539^{b}±0.264	

a-b: Means denoted with different superscripts within the same row or column (within each measure) are significantly different at (P<0.05)

Table.25 Least squares mean of teat diameter for female buffalo.

		Control	Treatment Group		Overall Mean
			PG	CP	
Diameter 1 right front teat	Pre-partum	1.976±0.101	2.728±0.101	2.750±0.101	2.484^{b}±0.058
	Post-partum	2.484±0.101	3.150±0.101	3.006±0.101	2.880^{a}±0.058
Overall Mean		2.230^{b}±0.072	2.939^{a}±0.072	2.878^{a}±0.072	
Diameter 2right behind teat	Pre-partum	1.827±0.108	2.839±0.108	2.817±0.108	2.494^{b}±0.062
	Post-partum	2.133±0.108	3.233±0.108	3.156±0.108	2.841^{a}±0.062
Overall Mean		1.980^{b}±0.076	3.036^{a}±0.076	2.986^{a}±0.076	
Diameter 3lift behind teat	Pre-partum	1.851±0.107	3.100±0.107	2.611±0.107	2.521^{b}±0.062
	Post-partum	2.501±0.107	3.489±0.107	2.889±0.107	2.960^{a}±0.062
Overall Mean		2.176^{c}±0.076	3.294^{a}±0.076	2.750^{b}±0.076	
Diameter 4lift front teat	Pre-partum	2.027±0.175	2.883±0.175	2.650±0.175	2.520^{b}±0.101
	Post-partum	2.634±0.175	3.256±0.175	3.194±0.175	3.028^{a}±0.101
Overall Mean		2.331^{b}±0.124	3.069^{a}±0.124	2.922^{a}±0.124	

a-b: Means denoted with different superscripts within the same row or column (within each measure) are significantly different at ($P<0.05$)

Table.26 Least squares mean of teat length for female buffalo

		Control	Treatment Group		Overall Mean
			PG	CP	
Length 1 right front teat	Pre-partum	3.868±0.249	4.594±0.249	6.028±0.249	4.830^{b}±0.144
	Post-partum	4.676±0.249	4.983±0.249	7.111±0.249	5.590^{a}±0.144
Overall Mean		4.272^{c}±0.176	4.789^{b}±0.176	6.569^{a}±0.176	
Length 2 right behind teat	Pre-partum	4.468±0.232	5.156±0.232	5.961±0.232	5.195^{b}±0.134
	Post-partum	5.384±0.232	5.539±0.232	6.667±0.232	5.863^{a}±0.134
Overall Mean		4.926^{b}±0.164	5.347^{b}±0.164	6.314^{a}±0.164	
Length 3 lifts behind teat	Pre-partum	4.042±0.274	5.133±0.274	6.050±0.274	5.075^{b}±0.158
	Post-partum	5.034±0.274	6.256±0.274	6.889±0.274	6.060^{a}±0.158
Overall Mean		4.538^{c}±0.194	5.694^{b}±0.194	6.469^{a}±0.194	
Length 4 lift front teats	Pre-partum	3.951±0.228	5.194±0.228	6.150±0.228	5.099^{b} ±0.132
	Post-partum	4.668±0.228	5.756±0.228	6.572±0.228	5.665^{a} ±0.132
Overall Mean		4.309^{c}±0.161	5.475^{b}±0.161	6.361^{a}±0.161	

a-b: Means denoted with different superscripts within the same row or column (within each measure) are significantly different at ($P<0.05$)

Table.27 Least squares mean of teat size for female buffalo

		Control	Treatment Group		Overall Mean
			PG	CP	
size 1 right front teat	Pre-partum	11.934±3.002	27.401±3.002	37.033±3.002	25.456^{b}±1.733
	Post-partum	24.553±3.002	39.474±3.002	51.649±3.002	38.559±1.733
Overall Mean		18.244^{c}±2.123	33.437^{b}±2.123	44.341^{a}±2.123	
size 2right behind teat	Pre-partum	12.766±3.130	33.229±3.130	38.731±3.130	28.242^{b}±1.807
	Post-partum	21.523±3.130	46.116±3.130	52.788±3.130	40.142^{a}±1.807
Overall Mean		17.144^{b}±2.213	39.673^{a}±2.213	45.759^{a}±2.213	
size 3 lift behind teat	Pre-partum	11.628±3.254	39.397±3.254	32.920±3.254	27.982^{b}±1.879
	Post-partum	27.368±3.254	61.101±3.254	45.466±3.254	44.645^{a}±1.879
Overall Mean		19.498^{c}±2.301	50.249^{a}±2.301	39.193^{b}±2.301	
size 4 lift front teat	Pre-partum	13.277±11.057	33.868±11.057	34.719±11.057	27.288^{b}±6.384
	Post-partum	27.092±11.057	48.142±11.057	68.834±11.057	48.023^{a}±6.384
Overall Mean		20.185^{b}±7.819	41.005^{b}±7.819	51.777^{a}±7.819	

a-b: Means denoted with different superscripts within the same row or column (within each measure) are significantly different at (P<0.05)

Table.28 Least squares mean of side area teat for female buffalo

		Control	Treatment Group		Overall Mean
			PG	CP	
Side area 1 of the teat	Pre-partum	23.758±2.956	39.062±2.956	52.517±2.956	38.446^{b}±1.707
	Post-partum	37.096±2.956	49.417±2.956	67.467±2.956	51.326^{a}±1.707
Overall Mean		30.427^{c}±2.090	44.239^{b}±2.090	59.992^{a}±2.090	
Side area 2 of the teat	Pre-partum	26.105±2.965	46.126±2.965	53.334±2.965	41.855^{b}±1.712
	Post-partum	36.336±2.965	56.432±2.965	65.883±2.965	52.884^{a}±1.712
Overall Mean		31.221^{c}±2.096	51.279^{b}±2.096	59.609^{a}±2.096	
Side area 3 of the teat	Pre-partum	23.790±3.163	49.953±3.163	49.723±3.163	41.155^{b}±1.826
	Post-partum	40.041±3.163	68.887±3.163	62.384±3.163	57.104^{a}±1.826
Overall Mean		31.916^{b}±2.236	59.420^{a}±2.236	56.054^{a}±2.236	
Side area 4 of the teat	Pre-partum	25.112±4.655	46.270±4.655	51.487±4.655	40.956^{b}±2.687
	Post-partum	38.759±4.655	58.674±4.655	67.896±4.655	55.110^{a}±2.687
Overall Mean		31.936^{b}±3.291	52.472^{a}±3.291	59.691^{a}±3.291	

a-b: Means denoted with different superscripts within the same row or column (within each measure) are significantly different at (P<0.05)

Table.29 Least squares mean of measurement the teat area for female buffalo

Measurement		Control	Treatment Group		Overall Mean
			PG	CP	
Measurement the teat area 1	Pre-partum	30.030±3.563	51.156±3.563	64.578±3.563	48.588^{b}±2.057
	Post-partum	47.284±3.563	65.186±3.563	81.841±3.563	64.770^{a}±2.057
Overall Mean		38.657^{c}±2.519	58.171^{b}±2.519	73.209^{a}±2.519	
Measurement the teat area 2	Pre-partum	31.615±3.669	58.926±3.669	66.051±3.669	52.197^{b}±2.118
	Post-partum	44.204±3.669	72.983±3.669	81.828±3.669	66.338^{a}±2.118
Overall Mean		37.909^{c}±2.594	65.954^{b}±2.594	73.939^{a}±2.594	
Measurement the teat area 3	Pre-partum	29.398±3.805	65.336±3.805	60.557±3.805	51.764^{b}±2.197
	Post-partum	50.646±3.805	88.231±3.805	75.634±3.805	71.504^{a}±2.197
Overall Mean		40.022^{c}±2.691	76.783^{a}±2.691	68.096^{b}±2.691	
Measurement the teat area 4	Pre-partum	31.827±7.218	59.694±7.218	62.637±7.218	51.386^{b}±4.167
	Post-partum	50.283±7.218	75.503±7.218	87.241±7.218	71.009^{a}±4.167
Overall Mean		41.055^{b}±5.104	67.599^{a}±5.104	74.939^{a}±5.104	

a-b: Means denoted with different superscripts within the same row or column (within each measure) are significantly different at (P<0.05).

The heist values in PG and CP treatment groups compared the control group back to the heavier LBW of born calves in PG and CP was associated with higher score of body condition of their dams thought post-partum and different sources of energy which fed dams.

Duration of placental drop

Data presented in Table21show averages post-partum duration of placental drop in different treatment groups. Analysis of variance presented revealed that this duration was affected significantly (P<0.05) by groups. Mean duration from parturition until drop of the fetal membranes was significantly (P<0.05) lower in treated groups PG and CP than that of the control group. These lowest values consider a good result with experimental group compared with control values. The present duration of fetal membrane drop is in accordance with the normal time needed from calving to release of fetal membranes, being 1-12 hours as reported by Stokes *et al.*, (2001), Kara *et al.*, (2009) and Abdel-Latif *et al.*, (2016).These results indicated lower duration of placenta drop in buffalo cows fed treatment with those fed control.

Post-partum uterine involution

The complete post-partum uterine involution to allow the return of the uterus to its normal non-pregnant size and condition was determined by recording the interval from parturition up to uterine symmetry in both of gravid and non-gravid homes. Also, the interval of the return of uterus to its normal position prior to pregnancy and post-partum cervical closure interval were recorded (Table 21). The interval required for pregnant uterus to return intrapelvic was affected significantly (P<0.05) by treatment groups later in control buffaloes (32.83d) than in PG (22.83d) and CP (20.33d). Post-partum uterine horns symmetry occurred earlier in treated groups (PG and CP groups) than in control about (34.83 and 31.67 VS 52.83 d). The group differences were statistically significant (P<0.001).The obtained interval of uterine horns symmetry in each experimental group was associated with the interval of placenta drop and the interval of return the uterus to its normal position prior to pregnancy. A general time of fetal membranes drop and uterine involution of treated cows my indicated normal parturition during pre-treatment season. It was reported that the uterine involution is considered to be complete when both uterine horns had returned to equal or almost equal non-graved size (Abdel-Latif *et al.*, 2016).

Post-partum first estrus interval

Least squares mean of post-partum first estrus interval (PPFOI) of buffalo cows in different treatment groups are shown in Table22. Analysis of variance of post-partum first estrus interval (PPFOI) in buffalo cows revealed significant (P<0.001) highest value of treatment groups on post-partum first estrus interval (PPFOI), although there was tendency of longer PPFOI in CP than in control group and PG. That means that PPFOI of buffalo cows fed PG treatment was earlier by about 5 and 29 days than those fed PG and control diet, respectively. The present length of PPFOI is in good agreement with that reported before by different authors (El-Moghazy, 2003 and Abdel-Latif *et al.,* 2016).

Service period length

Least squares mean of service period length (SP) of buffalo cows in different treatment groups are shown in Table37. Analysis of variance shows that SP length in buffalo cows was affected significantly by fed treatment group (Table22). Concerning all buffalo cows of CP treatment group at the 1st service period length was significant (P<0.05) higher in control group compared with PG and CP treatment groups. But the perfect period at the 1st service, SP length was 3.67d. The corresponding periods were 46.17 and 9.83 days in control group and PG treatment, respectively. It is worth noting that, all buffalo cows in CP treatment (95.4%), service period was about (4.6%). The improvement was in CP treatment followed by PG treatment compared control group. This is in accordance with the results of Angulo-Cubillan *et al.,* (1999). Also, in Egyptian buffaloes, mean of SP was found to range from 19 to 76 d (El-Moghazy, 2003).

Number of services per-conception

Least squares mean of number of services per-conception NS/C of buffalo cows in different treatment groups are shown in Table 23. Analysis of variance shows that number of services per-conception NS/C in buffalo cows was affected by fed treatment group was fed 300ml PG and 300g CP dissolved in 3L saline solution (0.9% NaCl) twice weekly and fed of female buffalo by pump feeder by oral. All buffalo cows in CP treatment group were conceived from the 1st service and NS/C was conception affected, since it significantly (P<0.05) increased with control group in NS/C than those in PG and CP treatment groups. The corresponding numbers were 2.17 and 1.33 services conception in buffalo cows in the control group an PG treatment group compared CP group that it lower in value. These results in CP and PG treatment groups in NS/C were good results compared with control group for female buffalo. Number of services per conception in Egypt buffaloes ranged between 1.3 and 2.4 (El-Wardani, 1995).Under small holdings condition, El-Moghazy (2003) found that overall mean percentage of animals required 1, 2, 3, 4 services for conception were 79.4, 15.1, 3.0 and 2.5%, respectively.

Days open

Least squares mean of days open (DO) of buffalo cows in different treatment groups are presented in Table 23. Analysis of variance revealed that DO was affected significantly by treatment groups. Results presented in Table41 showed affected significant (P<0.001) increased in control group compared with treatment groups. Buffalo cows in CP group showed the shortest interval in DO (52.17 d), while DO in PG and control group were nearly similarly, being 63.33 and 128.50 d, respectively. Values of DO in buffaloes of all treatment groups were in range from 52.17 to 128.50 d from three groups. These results agree with those of El-Wardani *et al.,* (1998) and El-Moghazy (2003).

Gestation period (GP)

Least squares mean of gestation period (GP) of buffalo cows in different treatment groups are presented in Table 23. Analysis of variance revealed that GP in buffalo cows was not affected by feeding treatment groups. Data show non- significant effect between treatment groups and control group but there was an improvement in GP treatment groups compared control group (436.33⁻ 378.17and 364, respectively).The differences in birth weight and age were reported by Wing (1899), who concluded that many cows show a marked individual trait throughout pregnancy.

Calving interval

Least squares mean of calving interval (CI) of buffalo cows in different treatment groups are presented in Table23. Analysis of variance showed that CI in buffalo cows was affected significantly by fed treatment groups. As a result of shorter PPFOI, and SP and in turn shorter DO in buffalo cows in treatment (PG and CP) groups. The CI was affected significantly (P<0.001), since it increased in control group than those in treatment groups. Shorter CI (364 d) was obtained for animals in CP treatment followed by those in PG (378.17 d) in treatment. However, the control group buffaloes showed the longest in CI (436.33d).These results are in agreement with those found by Mahdy *et al.,* (2001).Management practices and the physiological process of she buffalo are largely responsible to along CI, while the hendiadys component is almost negligible.

However, at conditions of small farmers, CI is shorter, being 402 – 453d (El-Wardany *et al.*, 1998 and El-Moghazy 2003).

Distances between teats (cm)

Data in Table 24 show that distance between teats for pre-and post-partum affected by treatment groups significantly (P<0.05) increased for female buffalo were fed treatment group in PG group but the values were similar between with CP treatment and control group in pre-and post-partum. Moreover, the same trend of values in overall mean between groups. Analysis of variance reflects the significant effect on animals fed the high energy from PG and CP that dissolved in 3L saline solution (0.9% NaCl).

Distance between behind of quarter right and left behind quarter (cm)

Results shown in Table24 for animals fed treatment groups affected significantly (P<0.001), since it increased in control group than those in PG and CP treatment groups in pre-partum period, but the values in distance between quarter of right and quarter of left behind were significantly nearly in PG treatment and control group in post-partum period. Overall mean was significantly increased in PG value of treatment than those in values of control group and CP treatment.

Distance between of quarter of right front and quarter of right behind (cm)

Data presented in Table24 are shown affected significantly (P<0.05) increased in PG treatment than those control group and CP treatment distance between of quarter of right front and quarter of right behind. In different period (pre-and post-partum), the values in CP treatment were nearly similar with value in control group. On the other hand, the overall mean was affected and significantly in control group than those treatment groups. Analysis of variance shows the significantly lower values for animals fed treatment groups compared with control group.

Distance between right front to right behind (cm)

Data in Table24show non-significance between groups but the increase was high in control group than those treatment groups but the CP treatment were nearly similar from value in control group 10.467 and 4.5, respectively in pre-partum but the values were increased in distance between right front to right behind control group than those in treatment groups. Overall mean reflected significantly (P<0.05) increased in PG treatment compared with control group and CP group these increase may back to the PG consider good a source of energy for fed animals. These results agree with those given by Akhtar *et al.*, (1999), they found studied 433 swamp buffalo to find the relationship between different teat's measurements and milk production. It was found that the distance between the fore and the hind teats increased until 3rd lactation and gradually decreased later. The same results were obtained with Chandrasekar *et al.*, (2016).

Diameter 1 of right front teat

Results presented in Table25of average diameter of teat for female buffalo fed treatment groups was significantly (P<0.001) increased in period of pre-partum and similar results with in post-partum for animals fed treatment than control group. Analysis of variance reflected significant effect between treatment groups. Overall mean showed significant (P<0.05) increase in diameter 1 of right front teat treatment groups compared with control group by the highest value was in PG treatment than control group and it was nearly similar with animals fed CP treatment.

Diameter 2 of right behind teat

Results in Table 25showed non-significant difference between treatment groups compared with control group in pre-and post-partum period. Analysis of variance showed the animals drenched treatment was not affected in interaction. Overall mean showed the significant (P<0.001) increase in treatment groups compared with control group. The highest values were in PG and CP treatment was nearly with it than the control group, being 3.036, 2.986 and 1.980, respectively in diameter2 of right behind teat.

Diameter 3 of left behind teat

The diameter of right behind teat is shown in Table25.Significantly (P<0.05) increased values were measured with animals fed treatment groups compared with control group in pre-and post-partum and the same trend was in overall mean showed significantly higher values in PG than those of CP treatment and control group in diameter 3 of left behind teat. Analysis of variance in Table 44showed affected animals fed experimental group in diameter 3 of left behind teat for groups' period.

Diameter 4 of left front teat

In this 4 of left front teat, the animals fed experimental group in two periods (pre-and post-partum) was not affected by treatment groups compared with control group but the highest value was in PG than those in CP treatment and control group in pre-and post-partum period. Analysis of variance showed the effect of time and not affected interaction. Overall mean showed in diameter 4 of left front teat significant (P<0.001) in PG treatment group than those fed CP treatment and control group and lowest value was in control group. The same results were obtained by Tilki *et al.*, (2005) and Prasad *et al.*, (2010). Also, there were significant effects on the equivalence of characters such as diameter of the front teats and the diameter of the posterior teats (Yosbams Espinosa-Nunez *et al.,* 2013).

Length of right front teats (cm)

Results in Table 26 show significantly (P<0.05) increased in length of right front teats (cm) in pre-and post-partum with animals in fed treatment groups than in fed control group and the same trend was in overall mean in length of right front teat and the highest value was in CP treatment than in those PG treatment and control group, but the control value was near from PG treatment and the highest value in length of right front teats (cm) in post-partum period this observation was also in overall mean between groups. Analysis of variance in Table 46 showed significantly effect in length of right front teats (cm).

Length of 3 right behind teats (cm)

The same significant effect in right behind teats, these results back levels of calcium effect and making good effect during the metabolism of diets in body female buffalo and overall mean effect significantly in effect of 3 rights behind teats but the improvement was in post-partum period.

Length of lefts behind teats (cm)

Analysis of variance revealed non-significant effect between groups for animal fed experimental group and control. Data (Table 26) showed significant effect (P<0.001) in overall mean for experimental groups compared with control group and the highest value was in CP treatment than those in PG and control group for pre-and post-partum period.

Length of 4 left front teats (cm)

The same trend in length 3 lifts behind teats resulted showed no effect between groups for animals fed treatment groups and control group, but the overall mean showed (Table26) significant (P<0.05) increase in treatment groups than in control group in length of 4 left front teats and the values were in CP treatment than those in PG and control groups. The results are in agreement with those found by Baghdasar (2011) and Chandrasekar *et al.,* (2016).

Size 1 of right front teats

Results presented in Table 27reflected no effect between treatment groups and control group in pre-and post-partum for animals fed PG and CP dissolved in 3L saline solution (0.9% NaCl) fed by pump feeder. Analysis of variance showed no differences between groups in size 1 of right front teats. Overall mean shown presented significantly (P<0.001) highest value in CP treatment group than those in PG treatment and control group but the highest value was in CP treatment group followed by PG treatment than the lowest in control group (44.341, 33.437 and 18.2444, respectively).

Size of left behind teats

The results in Table 27showed no significant effect between treatment groups and control in pre-and post-partum but the teats size improvement after post-partum in values than pre-partum period for animals were fed treatment groups compered control group. Overall mean showed significant (P<0.001) effect between time and treatment groups compared with in control in size right behind teats and the improvement was in post-partum period in this measure.

Size 3 of left behind teats

Results showed significantly (P<0.05) effect between treatment group than control size 4 left front teat in PG group followed with CP treatment compared control group but the improvement was post-partum period in overall mean and highest significant with in overall mean in PG treatment and CP treatment compared control group (being 20.249, 39.193 and 19.498, respectively).

Size 4 of left front teats

Analysis of variance calculated an effect in size 4 for female buffalo and control groups. Data shown in Table 27revealed significantly higher size with treatment groups than control group in pre-and post-partum time but the improvement in size 4 was in post-partum period. Overall mean between groups showed significant ($P<0.05$) higher size in CP treatment followed by PG treatment compared control group but the highest value was in CP treatment than in control group in size 4 left (being 51.777, 41.005 and 20.185, respectively) for animals fed treatment groups than control group. The results in all measurements size of teats were in agreement with those reported by Gajbhiye *et al.*, (2007) and Prasad *et al.,* (2010).

Side area of teat

The following Table 28 illustrates least squares mean and analysis of variance, respectively of side area of teats. There was significant ($P \leq 0.001$) effect of dietary treatments and time but not their interaction on this measurement.

Teat area

Table 29 presents least squares mean and analysis of variance, respectively of teat area. There was significant effect of dietary treatments ($P \leq 0.001$) and time ($P \leq 0.01$ and 0.001) but not their interaction on this measurement.

Conflict of interest statement

Authors declare that they have no conflict of interest.

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