Effect of propylene glycol supplementation to feed of dairy cows on some biochemical measurements

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Abstract

The periparturient period during late pregnancy and early lactation are physiologically important status for health of dairy cows and inside great metabolic stress. Propylene glycol (PG) is a substance used to prevent negative energy balance in periparturient dairy cows, since it has a beneficial effect on glucose and fat homeostasis. Fifteen clinically health pregnant cows in good nutritional condition nearly ages , weights , body condition , and 30 days before expected calving day . Animals are divided into three equal groups , the first fed a basal ration , the second was administrated 100ml liquid does of PG/os and the third group was administrated 200 ml of PG/os to every cow beside basal ration . The experimental period extended from 30 days periparturient to 30 days post parturient. Blood samples were obtained 4 hours after the onset of first feed in take or / and PG administration days. Serum was separated and analyzed for glucose, total protein, albumin, globulin, total cholesterol, triglyceride, blood urea nitrogen, creatinine, and non-esterified fatty acid (NEFA).The obtained results revealed, an improvement in the metabolic status reflected by an increase in glucose and total proteins in PG 200ml treated group at 30 days post parturient. Meanwhile, significant decreases in each of total cholesterol , triglycerides , NEFA and urea in PG supplied groups (100ml and 200ml) at 30 days post parturient . In addition a significant decrease in the creatinine level was observed in the 200 ml PG treated group at 30 days post parturient. The concentrations of albumin and globulin were uniform in all groups. It can be concluded that the use of such additional nutrient (PG) in later periparturient and in early post parturient period of cows was indicated beneficial effects in the treatment of such physiology and biochemical disturbances.

Introduction

Pregnancy and lactation are physiological status considered to modify metabolism in animals and induce stress (Rollin et al., 2010) and manifested by a decrease in voluntary feed intake, intense mobilization of body reserves, and increase in nutritional requirements. During the final days prior to calving and immediately postpartum, , a 30% dry matter intake (DMI) of dairy cows are usually decreased, as during this period a fetus grows rapidly and energy requirements for initiation of lactation are greatly increased (Rukkwamsuk et al., 2006), this phenomenon, so called negative energy
balance (NEB), induces the cows to increase mobilization of body energy reserves, mainly glycogen, fat and protein to compensate for their energy needs (Rukkwamsuk et al., 1999).

The periparturient period is important in terms for its influence on the health and the subsequent performance of dairy cows, since cows develop serious metabolic and physiological changes during these periods (Tanaka et al., 2011).

Propylene glycol (PG) is a substance used to prevent negative energy balance in periparturient dairy cows (Rukkwamsuk et al., 2005). It is used for prevention and treatment of ketosis in dairy cows. The use of such additional nutrients (PG) increases the energy in the late phase of pregnancy due to the increase in food consumption of the cattle (Toghdory et al., 2009). It also decreases NEFA concentrations (Grummer et al., 1994). It induced an optimization in metabolic parameters in peripartum period of cattle (Juchem et al., 2004). Several studies reported beneficial effects of PG on glucose and fat homeostasis in dairy cows (Nielsen and Ingvartsen, 2004).

The objective of this work is studying the effect of propylene glycol supplementation on some biochemical parameters in dairy cows.

**MATERIALS AND METHODS**

1-**Animals:** Fifteen healthy pregnant cows belonging to a private farm in Sharkia Governorate were used in this study. They are 3 - 4 years old, weighed an average of 390-440 kg were fed 3.5 - 4 kg per day of concentrate and the hay in add libitum. The cows were 30 days before expected calving day and divided into 3 equal groups: the first was fed a basal ration only with no PG, the second was supplemented with 100 ml liquid dose of PG per os per every cow and the last group was supplemented with 200 ml of PG per os per every cow. It gives to cows by drenching gun after dilution with water (1:1). All cows calved normally.

The experimental period started 30 days periparturient and extended to 30 days postparturient with adaptation and blood samples conducted on the last day.

2-**The PG:** It is a mono propylene glycol liquid Korea D2551U Batch No novy12012 novy12014 PG Corporation 1589/3 Seocho, Dong, Seocho- Seoui Korea.

3-**Blood Sampling:** Blood samples were obtained 4 hrs after the onset of first feed intake or/and PG administration days by jugular vein puncture. Then samples were centrifuged at 1500 r. p. m for 15 min after collection, and serum samples were harvested and stored at - 20°C until analysis for glucose (Oser, 1965) total protein (Doumas et al., 1981) , albumin (Bauer, 1982), globulin (Doumas and Biggs, 1972), total cholesterol (Ellefson and Caraway, 1976), triglyceride (Stein, 1987), blood urea
nitrogen (Chaney and Marbach 1962), creatinine (Young 2001) and (NEFA) non-esterified fatty acids according to Weichselbaum and M. Somogyi (1941).

4- Milk yield:

It was recorded at start of second week post parturient till end of experiment. After parturition all animals were left a week to provide colostrum for the calves and then milked after calves nourished and recording the daily amount of milk around the experiment period.

5- Statistical analyses:

Data were explored for Comparison of data between peripartum and postpartum without and with treatment doses sampling days were performed using the one-way Repeated Measure Analysis of Variance (ANOVA) was applied to evaluate the influence of the propylene glycol on the considered parameters (SPSS. 14, 2006). Differences were considered significant when p values were less than 0.05.

Results and discussion

Clinical observations: Cows were fed their diet for 60 days and no signs of lack of tolerance to the ration were noticed. On the first sampling day before the experiment, two of the cows consumed the entire amount of offered concentrate within 15 min whereas the third cow had consumed approximately half and stopped eating. The cow remained standing although it was severely affected by the incidence. After 2 hrs of feeding PG the symptoms had completely disappeared and it was observed that the cow started eating hay and the remained concentrate. By 2.5 hrs after feeding she was observed drinking water and had apparently completely recovered.

Milk yield: PG supplementation had no effect on milk yield. Initial milk yield 8.80-12.40 L/d was similar among the treatments as reported by (Borş et al 2014). Their results did not show a PG effect when supplied in the prepartum period on milk yield.

However, Fonseca et al (2004) observed higher milk yields only in the fourth and fifth lactation weeks for cows that received PG pre and postpartum. Similarly feeding cows with PG for a period from 3 weeks prepartum until 3 weeks postpartum contributed to a higher milk yield and an improvement of colostrum composition (Kupczyński et al., 2005).

Miettenen (1995) reported higher milk yield on the second test day in the treated group than in the control group. An application of propylene glycol resulted in higher milk yield observed by (Adamski et al. 2011).
In contrast, supplementation of PG by incorporating it into the concentrate had shown to increase milk yield (Baldi and Pinotti, 2006), it is in agreement with previous studies (Pickett et al., 2003).

**Blood metabolic profiles:** Concentrations of glucose in serum are significant increased on 30 days postpartum compared to another animals groups which may be due to increasing PG supplementation similar to (Castañeda-Gutiérrez et al.2009). Which would have contributed to the reduction of negative energy balance (NEB) (Adamski et al.,2011) or improve energy balance (Rukkwamsuk et al. 2010) during postpartum phase, or due to a decrease in glucose utilization by peripheral tissues, even under increased serum insulin concentrations (Chung et al., 2009).

By contrast, Garcia et al (2011), Borş et al , (2014 ) reported no change in serum glucose concentrations in cattle treated by propylene glycol .As PG is a glucogenic precursor which is firstly metabolized into propionate, then converted into glucose absorbed by rumen ( Nielsen and Ingvartsten 2004 ). This metabolic conversion may limit the utilization of endogenous glucose and prevent hypoglycaemia or even induce increase in glycaemia.

Christensen et al. (1997) failed to detect a significant effect of propylene glycol on glucose metabolism. It has been presumed that in the early weeks postpartum hypoglycaemia and hypoinsulinemia in cattle may occur due to NEB; this situation could be modified and associated to decrease in lipolysis (Castaneda-Gutierrez et al 2009).

After parturition, the concentration of glucose in blood serum decreased significantly (Adamski et al.,2011). low glucose level was associated with feeding of dry fodder ( Ramakrishna 2003 ) and insufficient nutrient intake or under nutrition ( Reynolds et al., 2003 ) or may be related to the sudden activity of the mammary gland and the increased lactose synthesis ( Djokovic et al., 2007).

Glucose level significantly decreased on postpartum compared with that prepartum (Rukkwamsuk 2010) it remained at lower levels corresponded well with other previous studies.

This decrease indicating that cows suffered some degrees of negative energy balance (Rukkwamsuk et al., 2006) and can be interpreted to be the consequence of high demands for lactose synthesis and of insufficient gluconeogenesis (Doepel et al., 2002).

However, Juchem et al., (2004) observed lower glucose concentration in prepartum cows treated with glucoplastic substances.
Nielsen and Ingvarsten (2004) stated that glucose response is rather limited even if blood samples were collected shortly after administration of PG, they suggest that the different results of blood glucose concentration could be explained by the time of blood sampling or by the allocation method.

Serum proteins levels were significantly affected from the physiological period and after PG supplement it increased significantly during lactation if compared to late gestation after PG supplementation. The variations reflect the maternal requirements of proteins need for milking and providing immunoglobulins (Mohri et al., 2007). During the postpartum period showed significant higher values in the “Glycol” group, nevertheless this trend could be due to the higher milk production (Chiofalo et al., 2008) of the animals fed the propylene glycol. Where, in early lactation (Leroy et al, 2004) have observed a significant increase of serum total protein which could be due to a decrease in serum globulin (El-Sherif and Assad 2001).

Adamski et al., (2011) recorded that the prepartum total protein concentration in blood serum was lower as compared to the one measured during the postpartum period, which was the result of the diet fed, or was observed due to an increased fetal growth, and especially the utilization of amino acids from the maternal circulation for protein synthesis in the fetal muscles (Antunovic et al. 2002). A lower total protein concentrations in the postparturient period observed by (Mordak and Nicpon 2006). Lower total protein concentrations may indicate malabsorption, malnutrition, agammaglobulinaemia.

Chibisa (2008) reported no significant variation in serum total protein (TP) concentration in the prepartum and postpartum period.

During the investigation period, no significant differences in albumin and globulin levels were observed. Albumin increased significantly during the late gestation compared to early lactation periods (Piccione et al. ; 2009). The increased total blood volume especially in late pregnancy induces an increase in glomerular filtration, which is also responsible for the increased values of albumin during late gestation and this significant increase in albumin level over dry period could be ascribed to the low protein intake during this period and dehydration (Yokus et al. 2006). The significant increase of albumin in late gestation proves the higher energy requirement for the fetal growth (Durak and Altiner 2006).

Ramakrishna (2003) reported a higher albumin concentration in animals maintained on dry fodder than those on green fodder and concentrates of higher crude protein (CP) value.
Albumins are important in transporting fatty acids and some steroid hormones. Serum albumin is a very sensitive and early nutritional indicator of protein status (Agenas et al., 2006) because its turnover is only 16 days. Pechovl et al., (2002) observed that the concentrations of albumin were uniform 2 weeks before calving until 5 weeks after calving.

In the present data PG supplementation was more significantly decreased the mean cholesterol concentration post parturient than others groups. It increased early postparturient in absence of PG when dairy cows start their milk production in agreement with (Rukkwamsuk 2010) and (Kim and Suh 2003). The reason for moderate variability of cholesterol is not clear but can probably be attributed to its metabolic variation with the blood glucose levels or insufficient nutrient intake can reduce circulatory cholesterol levels.

Variation in blood cholesterol content has been observed during pregnancy, as a precursor of the steroid hormones (Iriadam 2007). This lactation stress related change in cholesterol level could be attributed to corticosteroid mediated mobilization of body reserves (Swenson and Reece 1993).

The serum cholesterol concentrations were significantly decreased before parturition (Piccione et al., 2009), this is probably related to the role of the compound in ovary steroidogenesis, so that the total cholesterol concentrations are under control of the complex of factors.

In contrast, during late pregnancy, serum concentration of total cholesterol is increased by (Borş et al 2014). Schlumbohm et al. (1997) reported that PG supplementation has the ability to increase the cholesterol level after the end of administration due to the diminished responsiveness of target tissues towards insulin that together with an increased mobilization of fatty acids from adipose tissue make available new sources for fetal growth.

Kumar et al (2006) observed that low milk yielder cattle had higher serum cholesterol as compared to high milk yielder. (Grummer and Carroll 1988) documented the importance of cholesterol as a precursor of ovarian steroidogenesis.

While the mean cholesterol concentrations results in blood were not affected by the propylene glycol (PG) treatment during transition period (Toghdory et al, 2009, Mikula et al, 2008 and Rukkwamsuk et al., 2005).

Mean triglyceride concentration was significantly increased in late pregnancy due to the diminished responsiveness of target tissues towards insulin that, together with an increased mobilization of fatty acids from adipose tissue make available new sources for fetal growth (Schlumbohm et al., 1997). And remained lower.
throughout the postpartal study period than during the dry period and up to week 3 of lactation period by (Adamski et al 2011).

Propylene glycol supplementation induced a significant decrease of triglyceride concentration postparturient (Mikula et al; 2008 and Chiofalo et al 2008). In contrast, it did not affect the concentration of triglycerides as denoted by (Toghdory et al., 2009 and Rukkwamsuk et al., 2005).

On contrast a significant decrease was observed in triglycerides concentration during the late pregnancy (Nazifi et al. 2002) which could be explained as the effect of increased lipolysis which is hormonally regulated, and not an expression of energy deficiency (Holtenius and Hjort 1990). Circulating blood triglycerides contribute significantly to milk fat synthesis (Nazifi et al. 2002).

The blood concentration of NEFA, considered the best indicator of negative energy balance and of the lipomobilization intensity during the transition period (Civilelek and al., 2011 and Gonzales et al., 2011) and had a tendency to decrease as the lactation progressed (Garcia et al, 2011 and Adewuyi et al, 2006).

NEFA concentrations data are significantly increased in early lactating cows group compared to others ones. The rise in NEFA as showed at postpartum are useful for the animals to maximize milk synthesis with lower glucose consumption, moreover, the high growth hormone concentrations and the low insulin levels, present in blood stream during this period, stimulate a marked mobilization from adipose tissues, as confirmed by the increase in NEFA plasma levels (Wheelock et al., 2010).

While, Castaneda - Gutierrez et al, (2009) demonstrated that propylene glycol (PG) treatment during the prepartum has increased glucose concentration and significantly decreased NEFA concentration in cattle postparturient (Kupczyński et al.,2005, Rukkwamsuk et al., 2005).

Juchem et al., (2004) observed that drenching cows with glycol was an efficient method to reduce NEFA concentration most likely because the inhibition of adipose adenylate cyclase activity and lipolysis by elevated insulin concentration. Nielsen and Ingvartsen (2004) suggested that the propylene glycol reduces NEFA in cows that are too fat at calving.

Propylene glycol seemed to exert a greater effect on NEFA via insulin during extensive body fat mobilization for example the periparturient period (Vazques-Anon et al., 1994) or feed restriction.
On contrast insignificant decreased effect of PG supplementation in transition period on the concentration of NEFA before and after calving was recorded by (Mikula et al; 2008 and Moallem et al. 2007). Other explanations to why there is no effects of PG on NEFA was observed in the current study this could be due to the physiological stage of cow and the amount of PG supplemented.

Adamski et al, (2011) recorded the increased level of NEFA was found in blood of cows receiving propylene glycol as compared to prepartum period and remained elevated during the postpartum period (Kabu and Civelek 2012). Nevertheless, the observed increased level of NEFA did not indicate excessive lipolysis (Adamski et al, 2011). It can indicate short-term negative energy balance and adipose tissue catabolism (Agenas et al., 2006). Chibisa et al, (2008) stated that mobilisation of body fat stores remains limited when glucose was sufficiently provided.

No significant effect of propylene glycol treatment in the periparturient period on NEFA concentrations was detected during prepartum and postpartum periods (Castaneda-Gutierrez E et al., 2009).

The Serum urea nitrogen concentration in the PG postpartum group was significantly lower than that in the other postparturient groups which was significantly increased (Rukkwamsuk 2010) than the urea nitrogen concentrations at late pregnancy because of the increased requirement (Roubies et al., 2006).

Serum urea nitrogen is an indicator of CP intake (Sun and Christopherson 2005) as well as dietary energy - protein balance in ruminant’s diet. In dry animals low urea may indicate poor dry matter intakes (Yokus et al. 2006) as urea used for protein synthesis on the ruminohepatic pathway due to compensation of the low protein uptake during the dry period.

Elevated urea in fresh cows usually indicate an excess of dietary protein relative to energy, or breakdown of cow muscle tissue, which in domestic ruminants was ascribed to the cortisol-stimulated catabolism of proteins in the body (Silanikove 2000).

Contrary to our results, serum urea nitrogen concentrations did not differ during prepartum and postpartum periods (Rukkwamsuk et al, 2010) and Chibisa 2008) since it is not dietary energy. In agreement, Kabu and Civelek (2012) reported that serum urea nitrogen concentrations remained relatively constant in PG treated cows.

In the PG group the blood urea nitrogen (BUN) level was lower than that of the others group. Since both groups of cows received the same diet, elevated BUN levels may be the result of degraded body protein, which implies that PG can improve negative energy balance, a finding in agreement with that of (Barllard et al. 2001).
Apart from being a glucogenic precursor, propylene glycol might improve protein utilization and metabolism in the rumen (Rukkwamsuk et al., 2010).

The creatinine serum level was also significantly affected by the physiological phase and showed the higher levels during the late pregnancy and early lactation. It is recognized that during the late gestation, the mother, for the foetal maternal circulation, assumes the load of organic waste of the newborn (Ferrell, 1991). So, the increase in serum creatinine levels could be attributed to the development of the foetal musculature, which is well documented in sheep and ewes too (Roubies et al., 2006).

**Conclusion:**

Dairy cattle are prone to disturbances of glucose and lipid metabolism around the time of parturition. It can be concluded that the use of such additional nutrient (PG) in later periparturient and in early post parturient period of cows was indicated beneficial effects in the treatment of such physiology and biochemical disturbance. This case underscores the need to monitor feed quality and give careful consideration to feed substitutions based solely on costs.

**Table 1. Composition of the diets fed to both treated and untreated cows according to NRC (1989).**

<table>
<thead>
<tr>
<th>Chemical composition</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude protein % (CP)</td>
<td>14</td>
</tr>
<tr>
<td>Ash</td>
<td>7.1</td>
</tr>
<tr>
<td>Fat</td>
<td>4.3</td>
</tr>
<tr>
<td>Fiber</td>
<td>19</td>
</tr>
<tr>
<td>Carbohydrates %</td>
<td>46</td>
</tr>
<tr>
<td>Moisture %</td>
<td>9.6</td>
</tr>
<tr>
<td>NE Kcal / kg of DM</td>
<td>286.9</td>
</tr>
</tbody>
</table>
Table 2: Serum mean Concentrations of some metabolic parameters for treated and untreated propylene glycol 30 days prepartum and 30 days postpartum cows. (n=5)

<table>
<thead>
<tr>
<th>Serum concentrations</th>
<th>Groups</th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Non treated</td>
<td>Treated 100 ml PG/d</td>
<td>Treated 200 ml PG/d</td>
<td></td>
</tr>
<tr>
<td></td>
<td>30d prepartum</td>
<td>30d postpartum</td>
<td>30d postpartum</td>
<td>30d postpartum</td>
<td></td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>62.9±3.3 b</td>
<td>54.4±3.3 b</td>
<td>59.5±2.9 b</td>
<td>73.52±1.3 a</td>
<td></td>
</tr>
<tr>
<td>Total protein (g/dl)</td>
<td>6.9±0.40 ab</td>
<td>5.54±0.23 c</td>
<td>6.4±0.28 bc</td>
<td>7.5±0.43 a</td>
<td></td>
</tr>
<tr>
<td>Albumin (g/dl)</td>
<td>2.93±0.07</td>
<td>2.91±0.07</td>
<td>2.90±0.06</td>
<td>3.2±0.13</td>
<td></td>
</tr>
<tr>
<td>Globulin (g/dl)</td>
<td>3.8±0.14</td>
<td>4.02±0.05</td>
<td>4.3±0.31</td>
<td>3.97±0.10</td>
<td></td>
</tr>
<tr>
<td>Cholesterol (mg/dl)</td>
<td>138.9±27.4 b</td>
<td>211.0±11.1</td>
<td>123.36±24.3 b</td>
<td>80.9±13.6 b</td>
<td></td>
</tr>
<tr>
<td>Triglycerides (mg/dl)</td>
<td>25.3±1.87 a</td>
<td>16.63±1.4</td>
<td>11.3±1.3 c</td>
<td>11.03±1.07 c</td>
<td></td>
</tr>
<tr>
<td>NEFA (mmol/l)</td>
<td>0.24±0.02 b</td>
<td>0.68±0.04 ab</td>
<td>0.21±0.02</td>
<td>0.20±0.021 b</td>
<td></td>
</tr>
<tr>
<td>Urea nitrogen (mg/dl)</td>
<td>12.8±0.5 b</td>
<td>17.23±0.9 a</td>
<td>12.6±0.8 b</td>
<td>12.7±0.53 b</td>
<td></td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>1.24±0.09 a</td>
<td>1.12±0.04 ab</td>
<td>0.97±0.09 ab</td>
<td>0.89±0.12 b</td>
<td></td>
</tr>
</tbody>
</table>

Mean ± SE (standard errors). Values with different superscript letters are significantly different, P < 0.05.

Table 3: Effect of propylene glycol (PG) supplementation on milk yield (kg) of postpartum groups (n=5).

<table>
<thead>
<tr>
<th>Postpartum groups</th>
<th>Daily milk yield(kg)</th>
<th>21days milk yield(kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated</td>
<td>11.56 ± 0.66</td>
<td>242.8 ± 6.95</td>
</tr>
<tr>
<td>Treated 100ml PG</td>
<td>10.12 ± 0.18</td>
<td>212.8 ±1.3.89</td>
</tr>
<tr>
<td>Treated 200ml PG</td>
<td>10.36 ± 0.57</td>
<td>217.6 ± 11.9</td>
</tr>
</tbody>
</table>

Mean ± SE (standard errors).
References


Weichselbaum E. and Michael somogyi(1941) :A method for the determination of small amounts of ketone bodies 5-11Downloaded from www.jbc.org

