# Glycerol as a Feed Ingredient in Dairy Rations

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## Introduction

Glycerol, also known as glycerin, glycerine, or as propane-1,2,3-triol, 1,2,3-propanetriol, 1,2,3-trihydroxypropane, glyceritol, and glycyl alcohol, is a colorless, odorless, hygroscopic, and sweet-tasting viscous liquid. It is a sugar alcohol with high a solubility index in water. There are a wide range of applications for glycerol in the food, pharmaceutical, and cosmetic industries.

The term 'bio-diesel' is used to describe the methyl or sometimes ethyl esters produced from oilseed crops. Every 10 gallons of biodiesel produced generates about 7.6 lb of crude glycerol. According to the National Biodiesel Board, the production of biodiesel in the U.S. over the next decade is expected to grow (http://www.biodiesel.org/). Current annual production is 395 million gallons and planned expansions in the biodiesel industry are expected to drive that capacity to more than 1.1 billion gallons within the next 18 months annually, generating more than 800 million pounds of glycerol. Corresponding price projections suggest that glycerol could be priced competitively with grains as a source of energy for livestock. The value of glycerol in this regard may be further amplified with increasing diversion of corn and other grains to ethanol production. Although there is supporting evidence for use of glycerol for transition cows, there is little information that examines the use of glycerol as a macro-ingredient in rations for lactating dairy cows. This review will explore some of the attributes and issues pertinent to glycerol as a feed for lactating dairy cows and highlight results from a recent research study at Purdue University where the value of glycerol was examined as a replacement for corn grain.

## Glycerol Production and Quality Concerns

Most biodiesel is currently produced by a reaction that utilizes a base catalyzed transesterification of the oil. For soy diesel production, soybean oil is reacted with an equal weight of a short chain alcohol (usually methanol but sometimes ethanol) in the presence of a catalyst (sodium hydroxide; caustic soda or potassium hydroxide; potash) to yield biodiesel and crude glycerol. This process requires low temperature and pressure, yields high conversion (98%) with minimal side reactions and reaction time, and results in direct conversion of soybean oil to biodiesel with no intermediate compounds. The biodiesel is separated from the glycerol by gravity separation or by centrifugation. Because most commercial biodiesel production utilizes a 6 to 1 molar ratio of alcohol to oil, or excess alcohol, to drive the reaction to completion, methanol can partition to the glycerol and biodiesel phases.

Alcohol is removed from biodiesel and glycerol phases by flash evaporation or by distillation to recover, and re-use it. The resulting glycerol contains unused catalyst and soaps which are then neutralized by the addition of acid to produce crude

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glycerin containing 80 to 88% glycerol. Further purification of crude glycerin to 99% or higher purity is needed for use in the cosmetic and pharmaceutical industries. Impurities devalue crude glycerol; high levels of residual catalyst, salts, and methanol may be problematic in the using of glycerol as a livestock feed. Recent evaluation of crude glycerol from soy biodiesel production indicates a glycerol content of 76.2% and as much as 7.98% fat, 0.05% protein, and 2.73% ash. The latter was composed of 11 ppm Ca, 6.8 ppm Mg, 53 ppm P, and 1.2% Na (Thompson and He, 2006).

Glycerin is generally recognized as safe for use in animal feed (FDA, 2006, 21 C.F.R. 582.1320). Although food grade glycerol is safe in this regard concerns have been expressed relative to contaminant levels in crude glycerol from biodiesel production. Methanol levels are of particular concern and the methanol content of crude glycerol should be less than 0.5%. A recent regulatory letter issued by FDA indicates that methanol levels higher than 150 ppm could be considered unsafe for animal feed.

## Glycerol for Transition Cows at Low Inclusion Levels

The use of glycerol in the treatment of ketosis was reported as early as 1954 (Johnson et al., 1954), and evaluation of glycerol as well as propylene glycol as a ketosis treatment was further explored in the 1970's (Fisher et al., 1971, 1973). More recently, the value of glycerol has been examined as a preventative aid for metabolic problems associated with transition cows. Goff and Horst (2001) used up to 3 L in ketosis treatment and prevention, and DeFrain et al. (2004) fed 1.89 lb/day to transition dairy cattle. While these studies demonstrate the potential value of glycerol in treating ketosis, there is a lack of data to examine the value of glycerol as a primary ration ingredient for post-transition dairy cattle. Feeding rates for transition cows range from 5 to 8 % of the dietary DM.

## Feeding Studies Using Higher Inclusion Levels of Glycerol

Feeding studies have typically been lower from 150 to 472 g/day (Fisher et al., 1971, 1973; Kalili et al., 1997). There are only a handful of studies with glycerol feeding rates that approach 5% or more of the ration on a dry matter **(DM)** basis. Schröder and Südekum (1999) fed 10% glycerol to dairy cattle, effectively replacing over one-half of the starch in the diet, without negatively affecting intake, ruminal digestibility, rumen microbial synthesis, or total tract nutrient digestibility in steers. Feeding 3.6% glycerol to mid-lactation dairy cows was without effect on intake, milk production, or gross milk composition but slightly altered the profile of fatty acids in milk and increased rumen propionate and butyrate concentrations at the expense of reduced acetate concentration (Khalil et al., 1997). Feeding 1.89 lb/day of glycerol to +21 days relative to calving (5.4% of ration DM) did not have any effects on milk production or feed intake (DeFrain et al., 2004). Feeding 500 ml of glycerol, or approximately 3.1% of ration DM, from 3 weeks prior to calving through 70 days in milk caused an increase milk yield and milk protein content (Bodarski et al., 2005). Taken together, these experiments indicate that glycerol may be added to diets for lactating cows to a level of at least 10% of DM without deleterious effects, and in some cases, beneficial effects on milk production and composition have occurred.

## Energy Value for Glycerol

Because glycerol has not been used as a macro ingredient, the estimates of net energy of lactation **($NE_L$)** are not available for typical feeding scenarios. Schröder and Südekum (1999) reported estimates from 0.9 to 1.03 Mcal/lb with energy values decreasing for higher starch diets, and recently, DeFrain et al. (2004) reported 0.86 Mcal/lb when feeding glycerol in early lactation. There is uncertainty in the energy value for glycerol due to the amounts fed previously and unknown interactions with other ration components.

## Rumen Metabolism of Glycerol

Glycerol is fermented to volatile fatty acids **(VFA)** in the rumen. Early reports of glycerol fermentation indicated that glycerol was almost entirely fermented to propionate (Johns et al., 1953; Garton et al., 1961). Other reports indicate an increase in acetic and propionic acids (Wright, 1969) or increased propionic and butyric acids (Czerkawski and Breckenridge, 1972). In vitro glycerol fermentation using rumen fluid inoculum from cows adapted to glycerol feeding indicates increased production of propionate and butyrate at the expense of acetate (Remond et al., 1993). Studies using 14$^{o}$C labeled glycerol indicate that that most of the glycerol was found in propionate (Bergner et al., 1995). Rumen microbes adapt to glycerol feeding as the rates of glycerol disappearance from rumen fluid are more rapid after 7 days of glycerol feeding to donor animals used as a source of rumen-fluid (Kijora et al., 1998). In studies where 15 to 25% glycerol was added, most of the glycerol disappeared within 6 hours (Bergner et al., 1995).

The maximal rates of glycerol disappearance in the rumen determined using in vitro fermentors is 0.52 to 0.62 g/hour (Remond et al., 1993). There is lack of agreement for in vivo disappearance from the rumen by microbial metabolism. Estimates from disappearance of a 200 g dose of glycerol indicate that more than 85% of glycerol in the rumen disappears within 2 hours in cattle acclimated to glycerol feeding (Kijora et al., 1998). Other data using a dose of 240 g of glycerol indicate rumen disappearance rates ranging between 1.2 to 2.4 g/hour (Remond et al., 1993). Likewise, there have been reports suggesting that a portion of the glycerol entering the rumen can be absorbed directly (Remond et al., 1993). The fate of any absorbed glycerol is metabolism in the liver and requires glycerol kinase (Lin, 1977), and this enzyme is responsible for channeling glycerol into the triose phosphate step of glycolysis/gluconeogenesis. When glucose demands are high, such as the case for lactating cows, the fates of absorbed glycerol or propionate produced by rumen fermentation are likely to be identical.

## Feeding Experiments with Glycerol at Purdue University

The objective of our experiment was to evaluate the value of glycerol as a replacement for corn grain in diets of lactating dairy cattle. Sixty lactating Holstein cows were housed in individual tie stalls at the Purdue Dairy Research and Education Center and adjusted to a basal diet for a 2 week period. Cows were then assigned to diets containing 0, 5, 10, or 15% glycerol (99.5% USP/FCC, Kosher grade) as a percentage of ration DM. The basal (0 glycerol) ration was balanced to meet exceed or NRC (2001) requirements and contained corn silage, alfalfa haylage, hay, dry-rolled corn, vitamins, and minerals (Table 1). Corn was replaced by an equivalent amount of food grade glycerol and corn gluten feed. The addition of corn gluten feed adjusted for the protein removed with corn grain. Diets were offered once daily for ad libitum intake (5 to 10% weighbacks), feed refusals were measured daily and feed intake determined by difference. Cows were milked twice daily and milk samples were obtained weekly at two consecutive milkings and analyzed for fat, protein, lactose, total solids, milk urea N, and somatic cells.

Glycerol was well-tolerated by the cows, and there were no differences in DM intake or milk production when the entire 8 week experimental period is considered (Table 2). Feed intake was reduced by inclusion of 15% glycerol during the first 7 days of the trial. Negative effects on intake were only evident during the first week of the test and differences were not detected for the subsequent 7 weeks. Recovery of intake within 7 days suggests that achieving a feeding rate of 15% glycerol might be best accomplished with a protocol that gradually introduces glycerol into the ration.

Milk production and composition were not altered in response to glycerol feeding with the exception of decreased milk urea nitrogen in response to glycerol. These changes were observed at all levels of glycerol feeding. Reduced MUN concentrations suggest improved use of dietary protein by rumen bacteria and reduced losses as ammonia. Cows fed the highest amount of glycerol gained the most weight during the 8 week feeding period. Cows fed 10 and 15% glycerol gained more weight than cows fed 5% glycerol or the control diet. Weight gain for the control cows and 5% glycerol did not differ.

Estimates of $NE_L$ for the diets were calculated from intake, production data, and body weight **(BW)** changes. The energy content of each ration was calculated for each cow over the experimental period using total energy expenditure (milk, maintenance, and BW gain) with DM intake. An estimate of $NE_L$ (Mcal/lb) for each diet was determined from $NE_L$ used (Mcal) divided by DM consumed for the corresponding interval. Estimated energy values for the diets were 0.70, 0.70, 0.71, and 0.72 ± 0.02 Mcal/lb and were not different ($P = 0.90$). The lack of differences in this regard suggests that glycerol can be substituted for corn without adjustments for the energy content. However, the feed energy value of crude glycerol is likely to be less than that of pure glycerol and must be adjusted for the levels and energy content of the impurities. It should be noted that the energy values of the TMR determined by chemical analysis in Table 1, are slightly higher than the estimates determined by difference of milk produced and BW change. These differences may reflect the effects of increasing intake and therefore passage rate to reduce the NE value of the rations.

Results from this study clearly indicate that glycerol is a valuable feed ingredient for lactating dairy cows. Glycerol can be included as a macro ingredient in diets for lactating dairy cows without any deleterious effects. Therefore, feeding glycerol in place of corn is an alternative strategy for formulating diets for lactating cows when corn is not priced favorability.

These data point to the feeding value of glycerol when fed in pure form; however, depending on the level and composition of impurities, the feeding value of crude glycerol cannot be implied directly from these results.

## Summary

Previously published research and recent work completed at Purdue University indicate the value of glycerol as a feed for lactating dairy cattle. Increased production of biodiesel and resulting glycerol when combined with an increased demand for corn in ethanol production may warrant use of glycerol as livestock feed. Although issues exist relative to the composition of crude glycerol, there does not appear to be any detrimental impact of feeding glycerol up to at least 15% of the total ration DM. Caution should be used; however, when introducing glycerol to the diet as approximately 7 days is required to adapt the rumen to glycerol feeding.

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**Table 1.** Diet composition.

| | Glycerol (% of DM) | | | |
|---|---|---|---|---|
| Ingredient | 0 | 5 | 10 | 15 |
| Corn silage | 31.94 | 31.94 | 31.94 | 31.88 |
| Alfalfa haylage | 10.00 | 10.00 | 10.00 | 9.98 |
| Alfalfa hay | 12.16 | 12.16 | 12.16 | 12.14 |
| Soybean hulls | 7.66 | 7.66 | 7.66 | 7.64 |
| 48% Soybean meal | 6.62 | 6.62 | 6.62 | 6.61 |
| Roasted soybeans | 5.40 | 5.40 | 5.40 | 5.39 |
| Fish meal | 0.66 | 0.66 | 0.66 | 0.66 |
| Urea | 0.30 | 0.30 | 0.30 | 0.30 |
| Megalac-R®[1] | 0.98 | 0.98 | 0.98 | 0.98 |
| Corn, ground | 20.00 | 14.20 | 8.40 | 2.79 |
| Glycerol | - | 5.00 | 10.00 | 14.97 |
| Corn gluten meal | - | 0.80 | 1.60 | 2.40 |
| Mineral/vitamin | 4.28 | 4.28 | 4.28 | 4.27 |
| Chemical analysis, % of DM[2] | | | | |
| Crude protein | 18.1 | 17.5 | 17.9 | 18.1 |
| ADF | 19.1 | 19.2 | 19.4 | 19.3 |
| NDF | 30.9 | 32.4 | 29.7 | 31.0 |
| $NE_L$, Mcal/lb | 0.77 | 0.76 | 0.77 | 0.77 |
| Ca | 1.03 | 1.01 | 1.06 | 1.05 |
| P | 0.41 | 0.39 | 0.41 | 0.41 |
| Mg | 0.34 | 0.31 | 0.32 | 0.33 |
| K | 1.88 | 1.85 | 1.88 | 1.88 |
| Na | 0.25 | 0.24 | 0.28 | 0.27 |

[1]Church and Dwight Co., Inc., Princeton, NJ.
[2]DM = Dry matter, ADF = acid detergent fiber, NDF = neutral detergent fiber, and $NE_L$ = net energy for lactation.

**Table 2.** Effect of glycerol on feed intake, milk production, body weight (BW) change, and body condition score (BCS) change.[1]

| Item | Glycerol (% of DM) 0 | 5 | 10 | 15 | SEM | P[2] |
|---|---|---|---|---|---|---|
| Milk production, lb/day | 81.4 | 81.2 | 82.1 | 80.0 | 1.3 | 0.71 |
| Feed intake, lb/day | 52.8 | 53.9 | 54.1 | 53.0 | 1.2 | 0.82 |
| Efficiency, milk/feed, lb/lb | 1.56 | 1.52 | 1.52 | 1.53 | 0.04 | 0.85 |
| Milk fat, lb/day | 2.93 | 2.81 | 2.92 | 2.80 | 0.14 | 0.88 |
| Milk protein, lb/day | 2.19 | 2.28 | 2.33 | 2.28 | 0.09 | 0.78 |
| Milk lactose, lb/day | 3.66 | 3.71 | 3.88 | 3.68 | 0.18 | 0.84 |
| Milk solids, lb/day | 9.50 | 9.53 | 9.85 | 9.47 | 0.43 | 0.91 |
| SCC, 1000 cells/ml | 275 | 490 | 137 | 144 | 111 | 0.10 |
| Milk urea N, mg/dl | 12.5[a] | 10.9[b] | 10.7[b] | 10.2[b] | 0.4 | <0.05 |
| Milk fat, % | 3.70 | 3.52 | 3.58 | 3.58 | 0.11 | 0.69 |
| Milk protein, % | 2.79 | 2.84 | 2.86 | 2.89 | 0.06 | 0.62 |
| Milk lactose, % | 4.64 | 4.62 | 4.70 | 4.66 | 0.07 | 0.89 |
| Milk solids, % | 12.05 | 11.89 | 12.03 | 12.04 | 0.19 | 0.91 |
| BCS change[3] | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 | 0.91 |
| BW change, lb[3] | 69.4[a] | 89.6[ab] | 109.3[b] | 113.5[b] | 10.2 | <0.05 |

[1]SEM = Standard error of mean and SCC = somatic cell count.
[2]Probablilty that treatment means are equal.
[3]Change observed over the 8 weeks of the trial.
[ab]Means with different superscripts differ (P < 0.05).