EFFECT OF DIETARY PROTEIN DEGRADABILITY ON FERTILITY OF DAIRY CATTLE

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Abstract

Excess protein (0.45 kg/day) was fed lactating Holstein cows, primarily in the form of degradable protein (soybean meal) or undegradable protein (fish meal). Cows were superovulated, inseminated, and the eggs/embryos were collected 6 days later, examined, and incubated in vitro for 8 days. Blood urea nitrogen was slightly higher in cows fed soybean meal and slightly lower in cows fed fish meal as compared to cows fed the control ration. The source or amount of protein had no effect on the number of embryos recovered; however, embryos recovered from cows fed fish meal were less developed when recovered and fewer advanced to the blastocyst or hatched blastocyst stage during in vitro incubation. Further investigation is warranted to determine if fish meal contains substances which may be detrimental to fertility.

(**Key words:** Protein, Dairy, Fertility)

Introduction

It is known that feeding high levels of dietary protein during the early stages of lactation (breeding period) in dairy cattle is detrimental to fertility (Jordan and Swanson, 1979; Folman et al., 1981); however, the mechanism by which this is accomplished is unknown. Ferguson and Chalupa (1989) have suggested that the degradable protein content of the ration may be more important than the level of protein. Therefore, this experiment was designed to provide further evidence toward this hypothesis.

Materials and Methods

Eighteen high producing multiparous Holstein cows were randomly assigned to one of three groups after day 45 of lactation. Cows assigned to the control group were fed a ration containing 100% of their daily protein (degradable and undegradable) requirements (NRC); cows in the soybean group were fed 0.45 kg crude protein from soybean meal (0.36 kg degradable protein) daily in addition to the ration of the control group; and cows in the fish meal group were fed 0.45 kg crude protein from fish meal (0.36 kg undegradable protein) daily in addition to the ration of the control group. The three rations were isocaloric. The cows were maintained on these treatments for 27 days.

After 60 days postpartum, but during the treatment period, and having observed at least one estrous cycle, the cows were synchronized with prostaglandin $F_{2\alpha}$ (Lutalyse)

and subjected to a standard superovulatory protocol (36 mg pFSH administered over 4 days beginning on day 12 after administration of Lutalyse). A second injection of Lutalyse was given 14 days after the first to cause ovulation. Embryos were recovered on day 6 after insemination by non-surgical flushing, examined for stage of development and quality and then incubated in vitro (suspended in microdrops) for 8 days (192 h) in Ham's F-12 media, containing 1.5% bovine serum albumin, at 37°C in an atmosphere of 5% CO_2-95% air. The embryos were examined every 24 h for development to the blastocyst and hatched blastocyst stages and transferred to fresh media. Blood samples were collected from each cow during the treatment period, prior to superovulation, for subsequent analysis of blood urea nitrogen.

Results and Discussion

Although there were no significant differences between treatments in blood urea nitrogen (BUN) levels (Table 1), BUN levels were higher in cows from the soybean group and lower in cows fed the fish meal. This would be expected since soybean meal is relatively high in degradable protein whereas fish meal is lower in degradable protein (i.e. high in bypass protein). Feedstuffs containing higher levels of degradable protein have more of their protein metabolized in the rumen, overloading the rumen microorganism's capacity to convert ammonia to bacterial protein. Consequently, the excess ammonia is converted to urea which is recycled via saliva or excreted. Excess ammonia or urea in the blood could be a factor responsible for decreased fertility. However, Ferguson et al. (1988) has reported that fertility did not decrease until BUN levels exceeded 20 mg/dl.

The superovulatory response was quite variable among cows; there were no significant differences between treatments (Table 2). Three cows in the control group did not respond to pFSH and embryos were not recovered in three cows. All cows responded in the soybean group while embryos were not recovered in two cows in the fish meal group and one of these two cows did not respond to pFSH.

There were significant differences in the stage of development of embryos at the time of collection on day 6 after insemination (Table 3). Day 6 embryos should be at the morula to early blastocyst stage of development. The code 1 stage of development (1 cell) may contain unfertilized embryos. Embryos from cows fed the control ration were the most advanced while embryos from cows fed fish meal were the least advanced. This does not fit our hypothesis that embryos should be of higher quality in cows fed fish meal since these cows had lower BUN levels (Table 1) which should provide a better uterine environment, resulting in more advanced embryos.

The quality (a subjective measure of morphology) of the embryos collected on day 6 showed the same treatment differences as stage of development (Table 4). Embryos were of higher quality from cows fed the control or soybean rations than from cows fed fish meal.

Table 5 presents measurements which combine the stage of development with quality of the embryos at the time of collection - a combination of Tables 3 and 4. Embryos from both supplementary protein groups were significantly lower in quality x development score than embryos from the control group. In addition, embryos from cows fed the fish meal were significantly lower in quality and development than embryos from cows fed soybean meal.

Table 6 presents data on the proportion of embryos which advanced to the blastocyst stage during culture in vitro. There were no differences in rate of development between embryos collected from cows fed the control or soybean-based rations but a lower proportion of embryos from cows fed fish meal advanced to the blastocyst stage.

After examination of embryos which had advanced to the blastocyst stage during in vitro culture, the embryos were evaluated for those which had advanced to the hatched blastocyst stage of development. Similar results were observed (Table 7); embryos collected from cows fed the fish meal ration were significantly less advanced than embryos from cows fed the other two rations.

Table 1. Blood urea nitrogen (BUN) levels in Holstein cows during early lactation fed three rations differing in source of protein.

Source of protein	BUN (mg/dL)
Control	10.7 ± 1.28
Soybean Meal	11.1 ± 2.75
Fish Meal	9.6 ± 1.77

Table 2. Recovery rate of embryos from lactating cows superovulated with pFSH.

Source of protein	Recovery rate (# embryos/cow)
Control	3.8 ± 2.30
Soybean Meal	5.8 ± 2.30
Fish Meal	6.0 ± 2.30

Table 3. Stage of development[1] of superovulated embryos at time of collection on day 6 after insemination.

Source of protein	Development Stage
Control	6.9 ± .43[a]
Soybean Meal	5.2 ± .35[b]
Fish Meal	4.1 ± .34[c]

[1]Embryo development code: 1=1 cell or unfertilized; 2=2-3 cell; 3=4-5 cell; 4=6-8 cell; 5=16 cell; 6=morula; 7=early blastocyst; 8=blastocyst; 9=expanded blastocyst.
[a,b,c]Means within columns showing unlike superscripts differ ($P<.05$).

Table 4. Quality[1] of superovulated embryos at time of collection on day 6 after insemination.

Source of protein	Embryo quality
Control	3.3 ± .25[a]
bean Meal	2.8 ± .20[a]
Fish Meal	2.0 ± .20[b]

[1]Morphology (quality) code: 1=poor; 2=fair; 3=good; 4=excellent.
[a,b]Means within columns showing unlike superscripts differ (P<.05).

Table 5. Quality x development scoring[1] of embryos at time of collection on day 6 after insemination.

Source of protein	Quality X Development
Control	23.9 ± 2.00[a]
Soybean Meal	15.4 ± 1.64[b]
Fish Meal	9.9 ± 1.60[c]

[1]Refer to footnote of Table 3 for development coding and to the footnote of Table 4 for quality coding.
[a,b,c]Means within columns showing unlike superscripts differ (P<.05).

Table 6. Proportion of embryos collected from superovulated cows which advanced to the blastocyst stage of development after 192 h culture in vitro.

Source of protein	Percent of embryos advancing to blastocysts
Control	100.0[a]
Soybean Meal	96.4[a,b,]
Fish Meal	81.8[b]

[a,b]Means within columns showing unlike superscripts differ (P<.05).

Table 7. Proportion of embryos collected from superovulated cows which advanced to the hatched blastocyst stage of development after 192 h culture in vitro.

Source of protein	Percent of embryos advancing to hatched blastocysts
Control	85.7[a]
Soybean Meal	85.7[a]
Fish Meal	45.5[b]

[a,b]Means within columns showing unlike superscripts differ (P<.05).

We remain at a loss in attempting to explain the mechanism by which high levels of dietary protein are detrimental to fertility. From these studies it would appear that, either the amount of degradable and undegradable protein in the ration has no effect on fertility, or that fish meal contains a factor which may be detrimental to fertility. Since in every instance the quality and development of embryos from cows fed fish meal was lower than those from cows fed the control or soybean meal rations (even though the soybean meal and fish meal rations contained similar quantities of excess protein), it would appear that fish meal may contain a factor(s) which is detrimental to fertility.

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