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Cover Page Footnote

Hailey Hilfiker is a May 2020 honors program graduate with a major in Animal Science with a Pre-Professional concentration. Beth Kegley is the faculty mentor and a Professor in the Department of Animal Science. Rick Rorie is a committee member and a Professor in the Department of Animal Science. Jeremy Powell is a committee member and a Professor in the Department of Animal Science. This research was made possible through generous funding from the Bumper's Honors College Undergraduate Research committee, as well as an Honors College Research Grant.

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Meet the Student-Author

Hailey Hilfiker

I am originally from Piggott, Arkansas, where I grew up showing cattle on a local, regional, state, and national level. From these experiences, I developed a passion for large animals and began raising purebred Shorthorn cattle. I began my undergraduate career at the University of Arkansas as a major in Animal Sciences with a pre-professional concentration. I have served as the livestock activities chair for the Block and Bridle Club in the animal science department and a student ambassador through the college. I have also had the opportunity to study through the Bumpers Honors College and gain hands-on experiences through my research. In the summer of 2019, I began my research on beef nutrition and reproduction. I graduated Magna Cum Laude and was recently accepted to the University of Missouri's College of Veterinary Medicine, where I will obtain my doctor of veterinary medicine degree. My time at the University of Arkansas has been made possible by many individuals. I would like to thank Dr. Beth Kegley for serving as my honors mentor and for her confidence in me and continual support throughout this project. I also would like to thank Dr. Jeremy Powell and Dr. Rick Rorie for serving on my thesis committee and their help throughout this process. To my family, thank you for your continual encouragement and support while achieving my goals.

Hailey collecting blood samples from heifers for her study at the University of Arkansas System Division of Agriculture's research farm at Savoy.

Research at a Glance

- Phosphorus is an important mineral for growth and performance in beef cattle and is thought to be linked to reproductive performance.
- Data did not show any negative effects of removing phosphorus from free choice mineral but was not advantageous with regard to fertility and growth performance.
- Producers in the area where pastures have been fertilized with livestock manure could purchase mineral with or without phosphorus.

Impact of phosphorus intake on beef heifer growth performance and conception rates

Hailey Hilfiker,[*] Beth Kegley,[†] Rick Rorie,[§] and Jeremy Powell[‡]

Abstract

In Northwest Arkansas, soil phosphorus concentrations have increased where livestock manures have been repeatedly applied, leading many to question if supplementing phosphorus in this area is necessary. The effects of phosphorus intake on beef heifer growth performance and conception rates were investigated. In this study, crossbred Angus heifers (n = 72), approximately 30 days after weaning, were stratified by body weight (average initial weight 251 ± 3.9 kg) and allocated randomly into 8 groups. Groups were assigned randomly to 1 of 2 treatments. Treatments were delivered through either a free-choice-mineral mix that contained no supplemental phosphorus (CON), or a free-choice-mineral mix with 4% supplemental phosphorus and identical concentrations of other supplemental minerals (4PMIN). Heifers grazed 2.42 ha mixed grass pastures with a history of livestock manure application and were supplemented with soy hulls (0.5% of body weight) daily. Data were analyzed using the mixed procedures of SAS with group as the experimental unit. Total mineral intake through day 112 did not differ ($P = 0.55$) between treatments. On days 84 and 112, any heifers greater than 273 kg body weight (n = 58) had an ultrasound evaluation of their reproductive tract. Reproductive tract score (1, infantile to 5, corpus luteum present) did not differ ($P = 0.65$) due to treatment. Body weights were not different ($P \geq 0.59$) through day 264, 409 ± 6.0 kg and 412 ± 6.0 kg for CON and 4PMIN, respectively. When grazing pastures with a history of livestock manure application, heifers did not need supplemental phosphorus throughout the breeding season.

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Introduction

Nutrition has a major influence on the growth and productivity of livestock. To help an animal achieve its genetic potential, a well-balanced diet of protein, vitamins, and minerals is a necessity. While there are different nutrient requirements for each stage of an animal's life, it is well known that phosphorus is a crucial component to the feed ration of any livestock species. In recent decades, producers have used livestock manure as a fertilizer for their pastures, leading many to believe phosphorus concentrations in those areas are higher than average. Because of this, there has been much discussion on whether it is truly beneficial to add phosphorus to the diets of beef cows. The environmental aspect of this conversation is supported by excess phosphorus in the soil. While price discourages some producers from adding phosphorus to feed rations, studies have shown that well-balanced diets provide shorter anestrus cycles, or when the animal is not cyclic (Ciccioli et al., 2003). Furthermore, nutritionally compromised cows have difficulty maintaining adequate body condition scores to exhibit estrous (Hess et al., 2004). As an industry, cattle producers are in need of nutritional programs to increase and maintain fertility in their herds. In order to achieve a highly concentrated period of calving, early onset of puberty in replacement females is crucial (Diskin and Kenny, 2016).

Phosphorus is a crucial nutrient in animal health and well-being, with over three-fourths of the mineral being found in the body and is abundant in the bones and teeth of many species (Karn, 2001). The benefits of feeding phosphorus include increased cellular growth, development of musculoskeletal growth, and maintenance of body weight. Not only has phosphorus been shown to be vital to animal growth and well-being, but deficient amounts can cause reproductive problems, with previous studies finding that beef heifers fed higher levels of phosphorus continue to cycle later in the season over heifers that were fed diets lower in phosphorus (Call et al., 1978).

This study aims to examine the effects of phosphorus intake on weanling beef heifer growth performance and conception rates. One group was grazed on pasture with a decades-long history of livestock manure application, fed grain with minimal amounts of phosphorus, and given no supplemental phosphorus in a mineral mix, while the other was grazed on pasture with the same type of forage, fed the same grain, and given supplemental phosphorus in their mineral mix.

Materials and Methods

Animals and Management

For this experiment, heifers (n = 72) were weaned in May 2019 from the University of Arkansas System Division of Agriculture's Cow-Calf Unit in Fayetteville. Approximately 30 days after weaning, heifers were weighed, stratified by body weight, and divided into eight groups. Following this, groups were assigned randomly to one of two dietary treatments. Group A was supplemented with phosphorus, and group B was given no supplemental phosphorus. Treatments were delivered through free choice mineral (Table 1). All groups had identical mineral feeders in their pasture, mineral was constantly available, and mineral feeders were moved with groups as they rotated pastures every 28 days. Feeders were checked daily, and mineral additions were recorded. Every 28 days, the mineral remaining in feeders was weighed, and mineral disap-

Table 1. Composition of free choice minerals for heifers.

Ingredient	Control	Supplemental P
Calcium, %	20	20
Phosphorus, %	0	4
Salt, %	24 to 26	24 to 26
Magnesium, %	0.2	0.2
Potassium, %	0.1	0.1
Copper, mg/kg	2,500	2,500
Selenium, mg/kg	26	26
Zinc, mg/kg	10,000	10,000
Vitamin A, IU/kg	440,000	440,000
Vitamin D3, IU/kg	22,000	22,000
Vitamin E, IU/kg	22	22

pearance for each group was calculated and expressed on a grams/heifer each day basis. Heifers remained in 8 groups except during the breeding season (days 168 to 223); during this period, heifers were kept in 2 groups (1 group/treatment). Heifers remained on their appropriate mineral treatment, and mineral intakes were recorded; however, they were not used in the statistical analyses because of a lack of replication.

Cattle were examined daily to detect morbidity and received antibiotic treatment as required for pinkeye (n = 6) and mastitis (n = 1). Heifers were given a pinkeye vaccine on day 1 and were treated with a pour-on for ectoparasites (Standguard, Elanco, Greenfield, Indiana) on days 1, 27, 84, 112, and 252. Heifers were treated for endo- and ectoparasites on day 196 (Cydectin Pour-on, Bayer Livestock, Shawnee Mission, Kansas).

As the breeding season approached, heifers were allotted to treatments in a concurrent research project investigating the use of sexed semen in a short-term fixed-timed artificial insemination protocol. This project had a 2 × 2 × 2 factorial arrangement of treatments, and heifers on this pre-existing nutrition project were stratified across these new experimental treatments to be bred by artificial insemination. In brief, on day 151, half the heifers were administered 5 mL of prostaglandin$_{2\alpha}$ ($PGF_{2\alpha}$); 7 days later, controlled internal drug release (CIDR) intravaginal progesterone inserts and 2-mL gonadotropin release hormone (GnRH) were administered to all heifers. After 7 days, all CIDRs were removed, and all heifers were administered 5 mL $PGF_{2\alpha}$. Heifers were inseminated at either 54 or 72 hours after CIDR removal (days 167 and 168) with either sexed or conventional semen. When inseminated, the heifers also received 2 mL of GnRH. On day 179, heifers were exposed to fertile bulls (1 bull/treatment, bulls had passed a breeding soundness exam within 21 days of use), bulls were rotated between groups on day 196. On day 214, a bull was found to be injured and was replaced with a third fertile bull. Bulls were removed on day 224.

Collection Periods and Description

Cattle were grazed on 8 ha mixed bermudagrass and fescue pastures throughout the summer months and supplemented at 0.5% of their body weight with soybean hulls, a low phosphorus feed product. This diet met or exceeded protein and energy requirements. Soil samples were taken in February 2020 and were analyzed at the University of Arkansas System Division of Agriculture's Marianna Soil Test and Research Laboratory. Two soil samples were taken per pasture on a transect to a depth of 4 inches. Soil phosphorus concentrations were extracted with Mehlich-3 and determined by inductively coupled argon plasma (ICAP). Concentrations ranged from 130 to 259 ppm. Forage samples were taken on day 0 and approximately every 28 days thereafter for a total of 6 dates. Samples were collected by walking pastures and taking grab samples at random points throughout the paddock. Forages were stored in a freezer at -20 °C until analyzed (Table 2). In order to measure concentrations of minerals in the diet, samples were taken from free choice minerals as well as the pelleted soybean hulls.

Reproductive Tract Scoring and Pelvic Area Measurements

After day 84, any heifers that weighed greater than 273 kg began monthly ultrasound evaluations. Heifers were rectally palpated and evaluated using real-time B-mode ultrasonography to determine the uterine horn and ovary size. Reproductive tract scores (RTS) were given on a scale of 1 to 5. A score of 1 was given if uterine horns were <20 mm and no palpable follicles were on the ovaries, while a score of 5 was assigned when the uterine horns were ≥30 mm and >10 mm follicles present as well as a visible

Table 2. Forage composition of pastures (dry matter basis).

Date	NDF[a]	ADF	CP	Ash
	%	%	%	%
June, day 0	67.23	35.15	14.94	8.41
July, day 27	66.71	30.65	12.31	7.51
August, day 56	69.47	32.81	12.81	7.36
September, day 84	68.23	30.09	14.06	7.67
October, day 112	68.06	31.38	15.38	7.93
November, day 140	72.67	34.23	11.31	6.19
Hay	68.99	31.43	13.13	6.85
Soyhull pellets	67.99	48.83	10.69	5.13

[a] NDF = neutral detergent fiber; ADF = acid detergent fiber; CP = crude protein.

corpus luteum (Pence et al., 2000). Heifers weighing >273 kg initially were given a score, while on day 112, a second data collection was completed to obtain data on any heifers that did not meet the weight requirements on day 84 and on those heifers that had an RTS of <4 on day 84. On day 112, pelvic area measurements were taken using a Rice pelvimeter. This device was used to measure the internal area of the pelvis, and area was determined by multiplying the height by the width of the pelvic opening. Height was measured using the linear distance from the middle of the pubic bone to the bottom of the mid sacrum, while width was measured using the linear distance between the ilia (Deutscher, 1987). These data allow producers to detect heifers that could potentially experience dystocia due to small pelvic area.

Statistical Analysis

Mineral intakes were analyzed using the MIXED procedure of SAS (SAS Institute, Inc., Cary, N.C.). Replicate was a random effect, and group was the subject. Treatment, period, and their interaction were fixed effects. Body weights, average daily gains, and reproductive tract scores were analyzed using the MIXED procedure. Pregnancy data were analyzed using the GLIMMIX procedure. Binary distribution and the compound symmetry covariance structure were specified. In all analyses, replicate was a random effect, and group was the subject. Treatment was the fixed effect. For the purpose of this study, $P < 0.1$ are considered significant.

Results and Discussion

The supplemental phosphorus group consistently had a greater daily mineral intake compared to the control group (Table 3; $P = 0.06$). It is important to note that during breeding season (occurring over two periods from days 166 to 224), bulls and heifers were combined into one replicate per treatment. During this time, the control group experienced a higher mineral intake. This is potentially due to decreasing the number of groups from eight to two.

Forage samples were taken and analyzed to determine neutral detergent fiber (NDF), acid detergent fiber (ADF), crude protein (CP), and ash. Table 2 illustrates a consistent NDF forage value until day 140, where it was greatest at 72.67%. Compared to other dietary sources, soyhull pellets had a significantly greater percentage of ADF. Percent ash values varied during the study, with the largest percentage coming from the initial data collection on day 0.

With soils rich in phosphorus concentrations, forages consequently take up the mineral and have large concentrations available for grazing animals. In addition to pasture grass, heifers were given soyhull pellets at 0.5% of their body weight to supplement dietary needs. It can also be noted that with the phosphorus concentration of the soyhull pellets combined with forages, heifers were well over their specific requirements. During the winter months, heifers were fed hay and continued to receive soyhull pellets. It is worth noting that the hay consisted of 0.39% phosphorus, a value greater than any concentrations heifers had grazed earlier in the season. While the concentration of phosphorus in the soyhull pellets was 0.10%, heifers were receiving a small portion of their body weight. In order to achieve maximum efficiency and performance, growing beef cattle need approximately 0.25% of their diet to consist of phosphorus. Table 4 demonstrates that the phosphorus concentrations of the forages alone were above the minimum requirement for growing heifers.

Table 3. Mineral intake of heifers (g/day).[a]

				P-value		
Date	**Control**	**Supplemental P**	**SE**	**Treatment**	**Period**	**Treatment × Period**
Days 0 to 27	76.35	91.98	7.34	0.06	< 0.001	0.41
Days 28 to 56	72.3	84.64				
Days 57 to 84	55.89	64.76				
Days 85 to 112	54.95	66.11				
Days 113 to 140	62.19	69.75				
Days 141 to 165	74.54	88.1				
Days 225 to 252	56.52	74.84				
Days 253 to 263	82	123.27				
Overall	66.84	82.93				

[a] During 2 periods when with bulls, heifers were housed in 1 replicate/treatment, consumption was as follows: days 166 to 196 = 105.79 and 60.26 g/day; days 197 to 224 = 80.6 and 76.84 g/day for control and supplemental P, respectfully. These data were not included in the above statistical analysis. SE = standard error.

Soil analysis showed the concentrations of phosphorus to range from 130 ppm to 259 ppm. Soils with phosphorus concentrations between 36 to 50 ppm are considered ideal for maintaining optimal forage growth, while those above 50 ppm are considered above optimum. Grasses in this area are excellent consumers of phosphorus. Plant tissue phosphorus will increase if soil concentrations are high in the mineral. Because of this, forages in this area have larger phosphorus concentrations compared to other pastures that do not have a history of livestock manure application.

Heifers in both groups were consistent in their average daily gains (Fig. 1 and Table 5), with the exception of days 84 to 112 where both groups experienced a decrease in weight gain, but the control group gained more than the supplemental phosphorus heifers ($P = 0.04$). This overall decrease is most likely due to heat stress from summer conditions. During days 141 to 168, heifers in the supplemental phosphorus group tended to have a greater daily gain ($P = 0.08$) compared to those in the control group; however from days 169 to 196, heifers in the control group tended to experience a greater rate of gain compared to the supplemental phosphorus group ($P = 0.07$).

On day 84, all heifers weighing >273 kg were given an ultrasound to determine size of their uterine horns and ovaries, and to check for presence of a corpus luteum (Table 6). There was not a difference ($P = 0.65$) between the groups, with the supplemental phosphorus group having an average score of 3.07 compared to the control group's

Table 4. Feed mineral composition.

Date	P	K	Ca	Mg	S	Fe	Mn	Zn	Cu
	%	%	%	%	%	mg/kg	mg/kg	mg/kg	mg/kg
June, day 0	0.36	2.32	0.39	0.18	0.23	269	95	63	9
July, day 27	0.36	2.20	0.40	0.20	0.23	97	118	56	7
August, day 56	0.37	1.90	0.43	0.20	0.24	175	98	91	12
September, day 84	0.34	2.10	0.44	0.20	0.25	194	67	63	8
October, day 112	0.37	1.91	0.47	0.20	0.26	237	96	206	16
November, day 140	0.28	1.26	0.39	0.15	0.19	171	103	99	8
Hay	0.39	1.52	0.49	0.36	0.25	123	97	94	9
Soyhull pellets	0.10	1.17	0.64	0.23	0.09	393	26	44	7

P = phosphorus; K = potassium; Ca = calcium; Mg = magnesium; S = sulfur; Fe = Iron; Mn = manganese; Zn = zinc; and Cu = copper.

Table 5. Average daily gain of heifers.

Date	Control	Supplemental P	SE[a]	*P*-value
Days 0 to 27	0.71	0.70	0.040	0.76
Days 28 to 56	0.62	0.59	0.049	0.65
Days 57 to 84	0.29	0.39	0.042	0.13
Days 84 to 112	0.17	0.01	0.041	0.04
Days 113 to 140	0.50	0.51	0.042	0.84
Days 141 to 168	0.41	0.62	0.069	0.08
Days 169 to 196	1.11	0.95	0.059	0.07
Days 197 to 224	0.65	0.68	0.041	0.58
Days 225 to 252	0.97	1.01	0.067	0.76
Days 253 to 264	0.48	0.58	0.174	0.70
Days 0 to 264	0.60	0.61	0.015	0.70

[a] SE = standard error.

value of 2.89. On day 112, there was still no difference ($P = 0.35$) in RTS. From these data, it can be determined that there was little statistical evidence that phosphorus played a role in the growth and development of heifer reproductive tracts. In addition to ultrasonography, pelvic area measurements were taken on day 112. Between the control and supplemented phosphorus groups, there was little variation ($P = 0.51$).

The control group had 35% of heifers bred to the supplemental phosphorus group's 31% rate ($P = 0.73$). A blood sample to determine whether heifers were bred early in the natural mating season found that 74% of open heifers in the control group versus 52% of the open heifers in the supplemental phosphorus group were bred ($P = 0.09$) early in the natural breeding season. After two months, bulls were removed from the groups, and breeding season concluded. A blood sample was taken from any heifers open from the last blood draw and was tested again to determine pregnancy status to the bull via the entire natural service period. The results from this collection determined a final 89% and 78% pregnancy rate ($P = 0.19$) for the control and supplemental phosphorus groups, respectively.

Conclusions

Throughout this study, there were no negative effects of removing phosphorus from the free choice mineral; however, it still remains important to have adequate phosphorus concentrations in the total diet. Heifers in the control group performed as well, if not better, in several areas of this study, particularly in regard to pregnancy rates. Compared to the control group, the supplemental treatment had an 11% lower end of season pregnancy

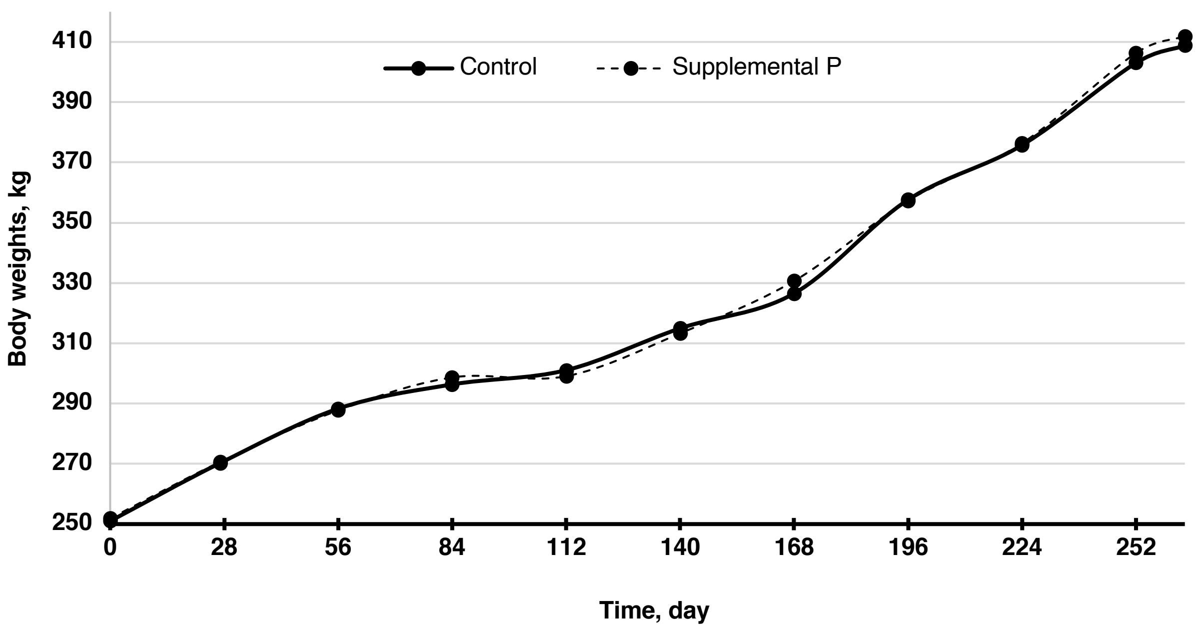

Fig. 1. Body weights of heifers.

Table 6. Reproductive data of heifers.

Evaluation	Day	Control	Supplemental P	SE	*P*-value
Reproductive tract score	84	2.89	3.07	0.27	0.65
	112	3.48	3.24	0.18	0.35
Pelvic area, cm^2	112	169	165	4.3	0.51
Pregnancy rate to synchronized breeding, %	196	35	31	10	0.73
Pregnancy rate for early bull bred, %	224	74	52	8.5	0.09
Pregnancy rate at end of breeding season, %	259	89	78	5.5	0.19

rate. When looking at other reproductive data, there was little variation between the two treatments. However, in the first attempt at breeding via artificial insemination, heifers in the control group had a higher rate of conception, and that trend continued during natural service. Producers in this situation, where the land had a history of manure application and forage concentration was 0.35% or greater, could either purchase mineral with or without phosphorus in it.

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