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MARES' MILK COMPOSITION AS RELATED
TO "FOAL HEAT" SCOURS

BY

ROBERT H. JOHNSTON

A thesis submitted
in partial fulfillment of the requirements for the
degree Master of Science, Major in
Animal Science, South Dakota
State University

1969

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**MARES' MILK COMPOSITION AS RELATED
TO "FOAL HEAT" SCOURS**

This thesis is approved as a creditable and independent investigation by a candidate for the degree, Master of Science, and is acceptable as meeting the thesis requirements for this degree, but without implying that the conclusions reached by the candidate are necessarily the conclusions of the major department.

Thesis Adviser

Date

Thesis Adviser

Date

Head, Animal Science Department

Date

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INTRODUCTION

A condition known as "foal heat" scours often develops in young foals during the mare's first estrum, which usually commences at nine days post partum. Although death due to this affliction is rare, the results such as loss of hair around the anus, unthriftiness of the foals, foul odor in the stable area, and unsightliness of the foals causes more than minor concern among horsemen.

Several causes for "foal heat" scours have been proposed by those familiar with the condition. Among these theories are (1) changes in the composition of the milk, (2) overloading of the foal's digestive tract, (3) ingestion of genital discharge from the mare, and (4) others among which include contamination of the milk in the udder by bacteria.

To date there have been no studies directed toward finding the cause or causes of "foal heat" scours. While some investigations of mare's milk have been conducted, none were designed to study a possible relationship between the mare's nine day estrum and changes which might occur in the milk. This research was initiated to study possible changes in mares' milk composition at the nine day period and perhaps establish a milk composition change with respect to the "foal heat" scours condition.

REVIEW OF LITERATURE

In surveying the literature on mares' milk composition and its possible relationship with "foal heat" scours, an attempt was made to determine (1) what is an "average" composition of this substance, (2) what factors bring about real or apparent changes from the "average," and (3) what component changes or other causes would be able to precipitate the condition known as "foal heat" scours.

The data available as to the composition of mares' milk are somewhat limited. Variations in sampling technique, breeds, stage of lactation, rations, season of the year, and methods of analysis all tend to cloud an already unclear picture and make it difficult to establish what is an "average" composition of mares' milk. The paucity of information in this fairly general subject makes it even more difficult to set up a casual relationship with an even less widely studied topic--"foal heat" scours.

"Foal Heat" and "Foal Heat" Scours

The "foal heat" follows soon after parturition and the mean interval has been given by Asdell (1964) as 9.1, 9.7, 11.3, and 11.4 days for mixed breeds, heavy mares, Percherons, and Belgians, respectively. Nishikawa (1959) has cited an average of 8.6 days from parturition to first estrum from observations on 10 mares of oriental breeding.

The occurrence of "foal heat" scours is fairly common and has been noted as such by Linton (1931), Udall (1943), Doll (1956),

Siegmund (1961), and Stowe (1968). Williams (1926) stated that "50 to 75% of all foals have diarrhea when nine days old." Doll (1956) in describing the condition explained that it often causes worry among horsemen. However, he has observed that the diarrhea developed some 8 to 12 days after birth, was usually mild and persisted for only a few days.

Average Composition of Mares' Milk

In one of the older and relatively more comprehensive works on mares' milk done by Vieth (1885), milk was studied from 15 Russian mares. The mares used were from 5 to 6 years old and were milked every other hour from 8 a.m. to 6 p.m., the foals not being allowed to suck during this time. Milk samples of the 15 mares were composited for 11 different milkings each, and the average values obtained were specific gravity = 1.0349, water = 90.06%, fat = 1.09%, protein = 1.89%, sugar = 6.65%, and ash = 0.31%. The pH of the fresh milk varied from neutral to slightly alkaline. Vieth also found that there was little difference in component parts either between mares or between individual samples drawn at different times, i.e., between the 10 a.m. sample and the 12 p.m. sample.

Linton (1931) gave a good review of the work on the composition of mares' milk up to that time which was done mainly by German, French, and Russian researchers. Realizing that no control was practiced with regard to the types of mares used, stage of lactation, rations, or methods of sampling and analysis, he summarized their independent works as follows:

	<u>Total Solids, %</u>	<u>Protein, %</u>	<u>Fat, %</u>	<u>Sugar, %</u>	<u>Ash, %</u>
Av.	9.78	2.18	1.29	5.93	0.38
Max.	11.20	3.34	2.50	7.26	1.20
Min.	7.47	1.33	0.12	3.28	0.26

Linton's first research of mares' milk was initiated in 1926-27 by requests from veterinarians and horsemen to analyze samples of milk from various animals to determine if the milk was abnormal in any of its constituents. The mares included in Linton's study represented eight breeds and a wide range of age and stage of lactation. Again, the methods of sample collection varied between mares. The results are as follows:

	<u>Total Solids, %</u>	<u>Protein, %</u>	<u>Fat, %</u>	<u>Sugar, %</u>	<u>Ash, %</u>
Av.	10.96	2.69	1.59	6.14	0.51
Max.	18.74	7.60	7.88	8.78	0.95
Min.	5.93	0.55	0.09	1.65	0.28

Masek (1938) analyzed milk from the wild mare, Przewalski, at the zoological gardens in Prague. He concluded that the milk of this mare being 89.52% water, 2.22% fat, 6.09% lactose, and 1.43% protein was not significantly different from the milk of tame mares as indicated by previous research.

Frenzel (1937) in his milk utilization and metabolism experiments with sucking foals concluded that the quality of mares' milk was extremely high. His results showed that the digestibility of milk nutrients for the first week was approximately 90%. Kleinert (1941) found the digestibility of all mares' milk nutrients to be over 95%.

Holmes et al. (1947) drawing "usually" the foremilk (that stripped out before the foal was allowed to suck but not stripping the teat of its entire contents) of three Percheron mares reported a range in average fat values for each individual from 1.0% to 1.6% and a range in total solids from 10.06% to 10.40%. The pH values varied between mares and between days for each individual. The range observed in pH was from 6.66 to 6.98.

Morrison's (1959) 22nd edition of Feeds and Feeding gives the following values for mares' milk composition: 9.4% dry matter, 1.1% fat, 2.0% protein, and 5.9% NFE (NFE would correspond to lactose). Kon and Cowie (1961) listed the composition of mares' milk as 10.1% total solids, 1.6% fat, 2.2% protein, 0.4% ash, and 6.0% lactose. Neither reference listed the number or types of mares involved, stage of lactation, or number of analyses; however, their values correspond favorably with those obtained from more completely documented sources.

Causes for Change in the Composition of Mares' Milk

Several different factors have been demonstrated to cause changes or variance in the composition of mares' milk. Listed among the factors are (1) daily variation and stage of lactation, (2) first, middle, or last drawn milk from the teat in a given milking, (3) diet, (4) stasis or pressure in the udder, and (5) estrum.

Daily Variation and Stage of Lactation. In a study by Linton (1937) with Shetland ponies and a Clydesdale mare, milk samples were drawn daily for the first 3 weeks post partum and then weekly

throughout the remainder of the lactation. Protein and ash values showed a tendency to decrease as the lactation progressed, while total solids, fat, and lactose values increased. Many isolated exceptions were noted which did not conform to the above mentioned trends; however, Linton ascribes this variation as being "due to the method of sampling by which milk samples were drawn from udders containing varying quantities of milk."

Papp (1939), while studying the effects of summer and winter feeding regimes, observed that under either system of feeding practiced protein and ash values decreased throughout the whole of lactation.

Holmes et al. (1947) milked three Percheron mares on the same calendar dates though the stage of lactation varied from 9 to 129 days post partum. Values for protein, ash, and moisture varied quite markedly from day to day for each mare.

Holmes and Lindquist (1947) conducted a study of milk from three Percheron mares and one lighter weight saddle mare for the first 21 days post partum. The average fat values were highest on the second day at 2.7% and gradually decreased to 1.3% on day 21 post partum. However, variations occurred. Especially noticeable were increases around the ninth day (time of first estrum) and the 16th day of lactation. Total solids in this study showed a similar tendency, dropping sharply from 16.5% shortly after parturition to 12.1% one day later and then gradually reaching an average of 10.3% on day 21 post partum. Again, minor daily variation was found. Hydrogen ion concentration was lowest for the first 4 days post partum with average values

being at a pH of 6.53. Then from the 5th through the 21st days, values varied between 6.73 and 6.94.

The milk of 15 mares, foaling between March and July, was studied by Krasnova (1962) for a 6 month lactation each. Mares were milked every 2 hours for a 12 hour period daily the first 2 months of lactation and then every 3 hours for the remainder of the test period. The highest protein levels occurred during the first month of lactation, while lactose values were at their highest level during the third month. At no time was there any great difference in fat content noticed.

Smanenkov and Sanygina (1964) studied the crude protein and amino acid make-up of milk from three Russian draft mares on the 3rd and 13th days of lactation. Mean total protein contents were 3.76% and 2.41%, respectively, for the two sampling days. The ratios in which the amino acids occurred varied widely.

Ullrey et al. (1966) investigated the milk of three Arabian and seven Quarter Horse mares at partum and 12 hours, 24 hours, 2 days, 5 days, 8 days, 3 weeks, 5 weeks, 2 months, 3 months, and 4 months post partum. Gross energy, specific gravity, total solids, crude protein, and ash values were all extremely high at parturition and then dropped sharply at 12 hours post partum. From this point on, the values dropped steadily to the end of lactation. Lipid values were lowest at parturition, rose to a peak at 24 to 48 hours, and then began a steady decline through the remainder of lactation. Lactose values appeared to rise steadily from their lowest point at parturition to a high at 3 months.

From the evidence submitted, it would appear that daily variation and stage of lactation do affect milk quality.

First, Middle or Last Milk Drawn From the Teat. A second factor which would appear to alter the composition of mares' milk is the manner of sample collection. Work in this area indicated that the percentages of constituent parts will vary, depending upon the fraction of sample tested--that is to say, first, middle, or last milk drawn from the milk cisterns will vary in composition.

In the 18th edition of Feeds and Feeding, Henry and Morrison (1923) stated that relative to mares' milk "the last portion carries the most fat." Dittrich (1938) analyzed 28 milk samples from four mares drawn either just prior to or just after suckling by the respective foals. Fat values were greatest for those samples drawn after suckling. The range in fat values was quite extensive, reaching from a low of 0.10% to a high of 3.74%. Kleinert's (1941) study of suckling foal metabolism for two foals and lasting 93 and 73 days, respectively, showed similar results. He found that mare milk obtained near the end of a suckling was richer in fat but did not vary greatly with respect to the other constituents.

Diet. Diet is a third factor which can change the composition of mares' milk. Work in this particular area is limited and therefore results are not conclusive. Papp (1939) studied milk produced by mares under winter and summer feeding practices. The results showed an increase in dry matter, lactose, fat, protein, and ash for summer milk

as compared to winter milk. An investigation by Lebeder (1961) involving 12 mares receiving equal grain rations and either grass or oat and vetch silage yielded the following conclusions. Mares receiving the better quality roughage (oats and vetches) not only had a 32% greater production but also the milk itself contained more protein, fat, and vitamin C. Tankuanpaa and Knudsen (1965) studied milk fat from 20 mares on pasture and 9 mares housed in a stable receiving 1 to 5 kg. of an oats, barley, and wheat bran mixture along with hay fed free choice. In this investigation, mares on pasture had less total fat with an average of 2.19% as compared to mares inside with an average of 2.30% fat. There was a significantly higher percentage of polyunsaturated fatty acids for mares on pasture and also a greater percentage of fatty acids with fewer than 14 carbon atoms.

Stasis or Pressure in the Udder. Linton (1937) used the milk from five Shetland ponies to study the effects of stasis or pressure in the udder on the composition of mares' milk. By allowing the milk to accumulate in the udder for varying lengths of time prior to milking, he obtained the following results. Fat percentages varied somewhat but were usually low for intervals under 12 hours; however, they rose sharply when the milking interval was increased to 24 hours or more. Protein levels remained relatively normal for intervals under 24 hours; but, similar to fat values, the protein content rose to extremely high levels after the 24 hour period. Lactose values of milk remained constant for intervals up to 12 hours, after which time they began a

rapid decline. Percentages of ash increased steadily the longer milk was retained in the udder.

Estrum. The effect of estrum on the composition of cows' milk has been studied much more extensively than has the effect of estrum on mares' milk. This phenomenon, however, was important enough for Linton (1931) to devote special attention to the subject and to give a discussion of the analysis of seven samples of mares' estral milk. His results showed a similarity to the observations made on dairy cattle up to that time. The fat content showed the greatest fluctuation of all components and was either extremely high or extremely low, depending upon the individual from which the sample had been obtained. Holmes and Lindquist (1947), as mentioned previously, also noted a deviation from the normal downward trend of the fat percentage at the ninth day estrum.

Possible Causes of "Foal Heat" Scours

A number of possible causes of "foal heat" scours have been proposed by various authors; and, in fact, most authors suggest several theoretical causes which may act independently or in conjunction with each other to produce the indisposition of the foal. The possible causes may be listed in four general areas: (1) changes in the milk, (2) overloading of the digestive tract, (3) ingestion of genital discharge, and (4) other proposed causes.

Changes in the Milk. Doll (1956) noted that the cause of "foal heat" scours may be a change in the dam's milk which comes about

simultaneously with her ninth day heat period. Henry and Morrison (1923) stated that, when milk is too rich, the resulting ingestion may terminate in diarrhea. They felt that fat was usually the disturbing element. Linton (1931) in comparing samples of estral milk from mares, some of whose foals were suffering from diarrhea, found that much the same changes take place in mares' milk as in cows' milk during estrum with fat being the most variable constituent. Holmes and Lindquist (1947) in their study of Percheron mares showed the fat percentage dropped steadily from the 5th to the 21st day post partum, with an exception at the time of first estrus when a temporary rise in the percent fat was observed.

Overloading of the Digestive Tract. A second theory also has support from several authors. According to Udall (1943), the removal of the dam for service on the ninth day results in irregular suckling, engorgement of the udder with milk, and consequent overeating. Henry and Morrison (1923) noted that the foal actually suffers from an "oversupply" of nourishment, while Siegmund (1961) also mentioned the engorgement of the udder and felt that foals may overload the digestive tract.

Ingestion of Genital Discharge. Siegmund (1961), in observing how diarrhea often develops around the time of the dam's first estrus, suggested that ingestion of genital discharge which has gravitated to the udder and teats may be a common cause. Sohnle (1910), Williams (1926), and Udall (1943) also mentioned the above postulation with

Sohnle quite decisively attributing it to the "fulmination of the intra-uterine infection of the mare at her first estrum when there is an increased virulence with copious discharges...and the foal inevitably takes the virus with its milk."

Other Proposed Causes. There are, of course, other theories as to the cause of "foal heat" scours. Krafft and Bernhart were cited by Sohnle (1910) as having demonstrated that mares excrete cocci in their milk. Udall (1943) felt, besides the oversupply of milk and the possible ingestion of genital discharges, one must remember that a foal may develop diarrhea from the eating of feces, straw, grass, and other foreign material. Stowe (1968) raised eight orphan foals on a calf milk replacer but found "in some foals a self-limiting diarrhea appears at the 'foal heat' period despite the foal's having no contact with the mare."

Summary of the Literature

The literature cited is a compilation of the investigations and opinions of many authors with respect to mares' milk composition and "foal heat" scours. Before one can draw any definite conclusions about either subject--mares' milk or "foal heat" scours--or the possible interrelationship of the two, all information presented must be studied and evaluated in its proper perspective. Several factors concerning the information available may influence conclusions as to the relative merits of the different sets of data and opinions which have thus far been presented.

Since the advent of agricultural mechanization and the subsequent decline of the horse as a source of farm power, scientific interest in this animal has also waned until fairly recent times when the pleasure horse began his rise in popularity. Because of this lapse in interest, much of the literature on horses is not only old but also has been done with the larger draft horses and not the lighter weight saddle horses which are in vogue today.

Another item which should be noted is that a significant amount of the research reported was done by foreign workers and is available in abstract form only. This fact in itself does not render the information less reliable; however, it is more difficult to make a fair, critical evaluation.

The opinions expressed by the several authors in this review were based upon varying degrees of supportive evidence. The manner in which experiments were conducted varied often not only between experiments but also within experiments with respect to one or more of the following: breed, age of mare, stage of lactation, diet, method of sample collection, and methods of analysis. Rather than state rigid values for mares' milk composition it would be more reasonable to list the ranges which one might expect to find, keeping in mind that the problem of collecting a truly representative sample of mares' milk has yet to be solved. The following composition ranges could be expected.

<u>Total Solids, %</u>	<u>Protein, %</u>	<u>Fat, %</u>	<u>Sugar, %</u>	<u>Ash, %</u>
9.5 - 13.0	2.5 - 4.0	1.2 - 2.5	5.5 - 8.0	0.35 - 0.55

Nowhere in the literature could be found any investigations which dealt solely with the problem of "foal heat" scours. That this problem does exist is not in question, but the extent of the problem and the possible causes are as yet still in the realm of speculation.

EXPERIMENTAL PROCEDURE

Since very little work has related mares' milk composition to the incidence of "foal heat" scours, it was decided to study the composition of mares' milk for the first 14 days post partum and determine whether or not a gross change could be noted for any components in association with the mares' ninth day heat period. The six Quarter Horse mares used in this study were arbitrarily assigned individual numbers, between one and six, inclusive, for identification. The ages of these mares were (in order) three, six, five, seven, seven, five; and these mares had previously had zero, zero, one, three, two, and one lactations, respectively. Mares were housed in box stalls during the night and turned into exercise lots during the day (weather permitting) both before and after foaling. Mare six had to be taken to Iowa State University at Ames for the last 7 days of her test period to be bred. Housing, feed and sample collection at the new location were handled in the same manner, however.

The grain mixture fed to the mares in this test consisted of 64.0% rolled oats, 19.2% cracked corn, 9.6% soybean oil meal (44.0%), 6.4% bran, 0.48% trace mineral salt, and 0.32% steamed bone meal. The grain portion of the diet was offered at a rate of 3.5 to 5.5 kg. daily, depending on the individual mare's size and condition. A mixture of alfalfa-brome hay of good quality along with mineral supplement, salt, and water were available ad libitum. The mares foaled between March 26 and May 28, 1968.

Milk samples were collected every 24 hours following parturition for 14 days. At the same time each day, the mares were separated from their foals 15 to 30 minutes by tying the dams outside their stalls while the foals remained inside where they were unable to nurse. Upon returning to the stalls, the mares were very willing to "let down their milk" and allow the foals to nurse--at which time all the milk was drawn by hand from the teat which the foal was not suckling. Samples were taken immediately to the laboratory in a closed container. Volume and pH were determined when samples reached room temperature and the casein from a 10 ml. sample was removed by precipitation and then stored on the filter paper at -20° C. until analyzed. Of the remaining sample, 10 ml. aliquots (20 ml. where volume of milk sample permitted) were taken and then stored at -20° C. until analyzed. At the time of milking each day, the first stream of milk drawn from the udder was collected directly in a sterile petrie dish and taken immediately to the veterinary laboratory for a bacterial culture study.

Ash was determined by burning a 10 gram sample (after preliminary drying) in a muffle furnace at 760° C. Crude protein was determined by multiplying the Kjeldahl nitrogen value obtained from an approximately 10 ml. sample by 6.38. Casein was analyzed for by adding 1.5 ml. of 10% acetic acid to 10 ml. of sample mixed with 30 ml. of water at 40 to 42° C. The precipitate formed was then collected by filtration and the protein content determined with the use of the macro-Kjeldahl method.

Total solids were determined by the Mojonnier method. A 2 gram sample was placed in a sample dish and the dish was then placed on a hot

plate (180° C.) until the free moisture had boiled off. The sample was then put in a vacuum oven at 100° C. and a vacuum of 22 inches of mercury for 10 minutes. After being transferred to a special circulation type cooling desiccator for 5 minutes, the solids were then weighed.

Fat was determined by the Mojonnier method also, which utilized specially adapted equipment consisting of an electric hot plate, vacuum oven, and a cooling chamber. This method is a modification of the Rose-Gottlieb method for fat determination. A 10 gram sample was placed in an extraction flask and to it were added in order 1.5 ml. of concentrated ammonium hydroxide, 10 ml. of 95% ethyl alcohol, 25 ml. of ethyl ether, and 25 ml. of petroleum ether. It was then centrifuged at 600 rpm for a one-half minute period and the ether layer poured off into a previously weighed fat dish. The sample was extracted again, this time using 5 ml. of alcohol and 15 ml. each of the ethers and shaken for 30 seconds after each addition. Centrifuging and separation were repeated. The ether was evaporated on the Mojonnier fat electric hot plate at 135° C. and 20 inches of mercury of vacuum, placed in the specially designed Mojonnier cooling chamber for 7 minutes, and weighed.

A bio-assay for estrogenic activity was made by injecting fresh 1 ml. milk samples obtained on days 5, 7, 9, 11, and 13 post partum into the neck of a castrated female mouse. The live weight of the mouse was taken exactly 48 hours post injection and the animal was then slaughtered. The uterus was removed immediately, weighed, and expressed as a percent of live weight.

RESULTS AND DISCUSSION

Introduction

An analysis of variance of the results obtained in this study (table 1) showed that the differences between days for ash, total solids, and crude protein values were significant, while the differences between mares for volume, pH, and crude protein were also statistically significant. Fat and lactose percentages were not significantly different either between days or mares. The mares were observed carefully for signs of estrus, which routinely commenced at 9 days following parturition. All mares accepted the stallion at least once daily on both days 9 and 10 p.p. (post partum). Since the foals of mares one through six (in order) began to scour on days 15, 21, 14, 17, and 9 p.p., respectively, it can readily be seen that only one foal developed the scouring condition by day 9 p.p. Profuse scouring accompanied by irritability and lasting approximately 3 days were the symptoms observed in all cases.

Volume

Volume varied significantly at the $P < .01$ level between mares but not between days. This finding would suggest that each mare varied from the others regarding the ability to produce milk in quantity. However, since no significant differences were noted between days for the individual mares or all of them as a group, it could be assumed that the variations which did occur were due to chance.

TABLE 1. ANALYSES OF VARIANCE

	d.f.	S.S.	M.S.
<u>pH</u>			
Days ^a	13	0.2231	0.0171
Mares	5	0.3609	0.0722**
Error	63	0.6415	0.0102
<u>Volume</u>			
Days	13	21,873.08	1,682.54
Mares	5	93,752.43	18,750.49**
Error	63	241,284.01	3,829.90
<u>Fat</u>			
Days	13	4.002	0.308
Mares	5	1.475	0.295
Error	63	16.248	0.258
<u>Total Solids</u>			
Days	13	40.597	3.123**
Mares	5	4.751	0.950
Error	63	44.205	0.702
<u>Ash</u>			
Days	13	0.0833	0.0064*
Mares	5	0.0112	0.0022
Error	63	0.1720	0.0027
<u>Crude Protein</u>			
Days	13	8.558	0.658**
Mares	5	1.419	0.284**
Error	63	5.199	0.083
<u>Lactose</u>			
Days	13	7.181	0.552
Mares	5	5.491	1.098
Error	63	33.506	0.532

* Differ significantly at $P < .05$ level.

** Differ significantly at $P < .01$ level.

^a No samples were obtained on day 8 p.p. for mare 2 or day 1 p.p. for mare 6.

Henry and Morrison (1923), Udall (1943), and Siegmund (1961) all attributed at least a portion of the cause for "foal heat" scours to an oversupply of milk at the ninth day--especially when the dam was separated from the foal for breeding. However, by studying figure 1 it can readily be seen that in only one instance (mare 4) was the collected amount highest for the mare's ninth day, while in two cases (mares 5 and 6) the amounts collected per individual mare were actually lowest at 9 days p.p.

The time for which the mares were separated from their foals for breeding was about the same as the length of time which they were normally separated prior to collecting the milk samples through the entire test period. The breeding practices are about the same at the South Dakota State University Horse Barn as for most other horse breeding establishments. The mare is teased and, if found to be in heat, tied up away from the colt only long enough to place a tail wrap on the mare and allow the stud one mounting. The whole process very seldom takes longer than 15 to 30 minutes. Theories which propose an over engorgement of the udder during the separation for breeding have no basis in fact because the time interval would appear to be a minor factor. Even when draft horses were used years ago, the mares were away from their foals for longer periods of time while working than while being bred. The mares' milk producing capacity or even an accurate method for estimating this capacity has not yet been established, so final judgment should be reserved until that time.

Figure 1. Volume of milk collected daily per individual mare.

pH

The differences in pH of the milk, like volume, were also found to be highly significant between mares but not between days. Even though the differences between days were not statistically significant with respect to hydrogen ion concentration, figure 2 demonstrates that for all mares the pH was always higher on day 14 p.p. than day 1 p.p.; and in only one instance was the highest pH for an individual mare recorded on the last day of the test period. Most of the readings began low at day 1 p.p., became most alkaline between days 1 and 14 p.p., and then gradually declined but to a reading which was still higher than the day 1 p.p. value.

Holmes and Lindquist (1947) obtained very similar results to those found in this investigation. While the initial pH values on day 1 p.p. for three Percheron mares were somewhat higher (6.48, 6.45, and 6.65) than most of the pH values recorded for this study, they did show a tendency to hit a peak during the first 13 days of lactation and then began a gradual decline. In the study by Holmes and Lindquist, the values obtained the first 3 days were the lowest.

Fat

The mean percentage of fat in the milk from the six mares in this study was highest at 2.7% on day 1 p.p., lowered considerably by day 2 p.p., and followed an irregular downward trend to day 8 p.p., at which time the percentage of fat was 1.7% (figure 3). At day 9 p.p. the fat value rose by some 0.3% and declined again until the end of the test period. A rise in the fat percentage of the milk occurred between the

Figure 2. pH of fresh mare milk.

Figure 3. Lactose, crude protein, fat, and ash as a percent of whole milk.

day previous to and the day on which each of three foals scoured. No milk samples were being collected after 14 days p.p., the time during which the remaining three foals scoured, so the characteristics of the milk from this period are unknown. The changes noted for the early scouring foals, however, amounted to less than 0.5% actual increase, and there were numerous other daily increases of greater magnitude which occurred for the milk of individual mares that did not precipitate a scouring condition in the foals. The fat of the mares' milk appeared to be the least constant individual component.

The results obtained by Ullrey et al. (1966) differed slightly from those obtained in this study in that lipid concentrations were lowest at parturition, reached their highest concentration between 24 to 48 hours, and then declined through the remainder of the lactation. In the study by Ullrey et al. (1966) no samples were taken between day 8 and day 21 p.p., so possible changes which might have occurred at day 9 p.p. could not be detected. Holmes and Lindquist (1947) showed results very similar to those found in this investigation. Initial fat percentages averaged 2.2% on the first day after parturition and had declined to 1.7% on day 5 p.p. and then further declined to 1.3% on day 21 p.p. These researchers also noted a temporary increase in the average fat percentage beginning on day 9 p.p., at the time of the dam's first estrus.

Lactose

The lactose concentrations in this study were at their peak on day 1 p.p. and very low on day 2 p.p., from which time they rose slowly until the end of the test period (figure 3). However, it should be

pointed out that the lactose concentration for the milk of one individual was extremely high on day 1 p.p. and not in accord with the values obtained from the other mares. No statistical differences could be determined for lactose concentrations of the milk, either between days or mares. All the values ranged between 4.7% and 9.1% with the exception of the one first day value of 11.1% for mare three. Daily variations in the percentage of lactose were small and no patterns of fluctuation could be associated with the scouring conditions noted for the foals.

Linton (1937), Krasnova (1962), and Ullrey et al. (1966) substantiated this study's results. These researchers also found that the concentrations of lactose in mares' milk increase as the lactation progresses.

Ash

The mean percentage of ash, as indicated by this investigation, in the mares' milk appeared to decline slowly if somewhat irregularly after day 6 p.p. (figure 3). The highest single value recorded was 0.67% and the lowest was 0.35%. The differences between days were significant at the 5% level, although the differences in ash content of the milk between mares were not.

The tendency for the ash content of mares' milk to decrease with advancing stages of lactation was first noted by Linton (1937) with 104 milk samples from Thoroughbred mares and also with 197 milk samples drawn from Shetland ponies. A sharp drop in ash content was reported by Ullrey et al. (1966) for samples collected between 0 to 12 hours

following parturition, but then a very slight rise occurred until day 8 p.p., after which time the percentage of ash steadily declined.

Crude Protein

Crude protein was the only component of the mares' milk in this study which was found to be significantly different both between days and mares. For both days and mares, these differences were highly significant ($P < .01$). The mares (one through six) had mean crude protein percentages of 3.13, 3.08, 2.83, 2.89, 2.87, and 2.75, respectively. The mares consistently produced milk of higher protein concentrations immediately following parturition which then declined steadily throughout the test period of 14 days (figure 3).

The results obtained in the South Dakota State University study compare quite favorably with those of Linton (1937) and Ullrey et al. (1966) who also noted the rapid initial decline in protein content which gradually tapered off. Papp (1938), too, found a percentage decrease in mares' milk as the lactation progressed.

Total Solids

The range of total solids in this study varied from a high value of 18.29% for a sample collected immediately following parturition to a low value of 10.51%. No values other than those for day 1 p.p. samples exceeded 13.0% (figure 4). Total solids were found to differ significantly at the 1% level for samples collected on different days; however, no significant differences in total milk solids were found between mares. The basic tendency noted was for total solids to have higher values

Figure 4. Total solids as a percent of whole milk.

Figure 5. Crude protein, lactose, fat, and ash as a percent of the total solids.

closely following parturition, drop off quickly at day 2 p.p., and then begin a gradual decline throughout the remainder of the test period.

Ullrey et al. (1966) found a decrease in total solids with values dropping from 25.2% to 11.5% for samples collected at 15 minutes and 12 hours p.p. Holmes and Lindquist (1947) recorded average values for total solids of 16.5% for samples collected shortly after parturition, while milk samples collected 24 hours later averaged only 12.1%. From this time until 21 days p.p. the average total solids declined slowly to 10.3%. In contrast, however, to the findings in this study and those by Ullrey et al. (1966) and Holmes and Lindquist (1947), an increase in total solids was noted by Linton (1937) as the lactation of mares on pasture progressed.

In order to establish the relationship of the individual components of the total solids fraction to the gradual decline in total solids as the lactation progressed, the values of lactose, crude protein, fat, and ash were calculated as percentages of the total solids. Figure 5 shows the contributions of the individual constituents to the total milk solids and also the tendency of the solids as they gradually decreased through the test period.

Both crude protein and ash contribute directly to the decline of total solids since these values decline as lactations continue. They also decline as a percent of the total solids as can be seen in figure 5. While the differences in lactose were not significant between days, they do show a trend toward increasing gradually, both as a percentage of the milk and thus as an increasingly greater proportion of the total

solids. The percentage of fat, like lactose, did not differ from day to day enough to be of significance statistically. However, fat did decline irregularly to day 8 p.p., rose again on day 9 p.p., and then began to decline again. The fat as a percentage of the whole milk and its contribution to the total solids both showed this tendency.

Casein

When the volume of milk collected per sample was large enough to permit the analyses for the components previously discussed and an analysis for casein, such a determination was made. The lowest casein value found of 40 samples tested was 1.09% while the highest was 3.27%. Casein represented between 34.4 to 70.8% of the total crude protein. Realizing that insufficient evidence prevents the stating of any proven generalities, observations from this study would tend to indicate that, as the lactation progresses, the casein drops slightly as a percentage of the total crude protein fraction. In one sample of mare's milk containing 1.43% crude protein, Masek (1938) found that 47.5% of this fraction was casein.

Bio-assay

In the bio-assay for estrogenic activity of the milk, five female mice were left with their ovaries intact while five were castrated and received no hormonal stimulation from any source. This was done so as to determine what uterus/live weight ratios could be expected for normally stimulated and nonstimulated uterine development, respectively. The mean value for the five intact females was 0.53%, while for the

castrated mice the mean value of uterus/live weight ratio was 0.19%. The values which were obtained from the individuals treated with 1 cc. of fresh mares' milk ranged from 0.10% to 0.68%; however, no trends could be noted and none of the differences were shown to be significant. Even though varying degrees of stimulation appeared to exist, a more sensitive and exacting test will have to be designed before drawing any definite conclusions.

Bacterial Cultures

The bacteriological examinations of the fresh milk samples showed no consistent patterns of contamination. Most of the organisms identified could be explained as skin or air contaminants which were not secreted in the milk. In over half of the milk samples, no bacterial growth of any kind was found.

CONCLUSIONS

Our study of the milk from six Quarter Horse mares obtained on the first 14 consecutive days post partum showed highly significant ($P < .01$) differences between mares for volume, pH, and crude protein, and highly significant ($P < .01$) differences between days for total solids and crude protein. The differences in ash content of the milk between days were significant. No definite trends could be noted with respect to milk volume differences between days, however, the mares did appear to differ from each other with respect to the quantities produced daily per individual mare. Though differences in pH for days 1 to 14 were not significant statistically, values recorded in most cases were lowest shortly after parturition, reached a high value during the middle of the test period and had declined on day 14 p.p. to a point in between the highest and lowest values previously recorded. Total solids, crude protein, fat, and ash all showed a basic tendency to decline as the lactation progressed and in all cases the differences between days were significant with the exception of fat. The mean fat percentages also varied from the other components due to a slight increase in concentration noted at day 9 p.p. Lactose was the only component of the milk in this study which showed an inclination to increase daily though this tendency was not of any significance statistically.

The mouse-uterine assay for estrogenic activity of the milk indicated isolated positive results, but the test used was not able to point out any definite trends. A study for detecting bacterial

contamination of the milk led to the conclusion that the mares' milk in this investigation was free of microbial infestation.

The foals in this study began to demonstrate the symptoms of the "foal heat" scours condition on days 15, 21, 14, 13, 17, and 9 p.p., respectively, with the last case being the only one to occur in conjunction with the mare's first estrum. No significant correlations were found with respect to the mares' first estral periods and composition changes in the milk. Also, the initiation of the scouring condition occurred at a time other than during the respective dam's heat period with the exception of one foal.

Some information pertinent to "foal heat" scours was obtained from other researchers after the initiation of this study. Dr. Howard D. Stowe of the University of Kentucky observed the "foal heat" scours condition at 9 days p.p. on several foals which were being raised on a milk replacer. The foals had absolutely no contact with the respective mares or their milk. Dr. William J. Monson of the Borden Chemical Company, through personal correspondence relative to his company's three years of study of a mares' milk replacer, also reported that they have frequently noted a marked scouring condition shortly after one week of age. These foals likewise were completely separated from their dams and had no access to mares' milk.

Several changes in the approach to further research in this area of "foal heat" scours could prove enlightening, as follows:

- (1) Devise a method for the collecting of more representative samples of mares' milk.
- (2) Compare the performance of foals suckling their respective dams with foals on milk replacer diets.
- (3) Utilize a larger number of experimental animals.

Even though the mean fat percentage of the milk increased at the ninth day post partum, and slight milk fat increases were also noted just prior to the scouring of three foals, the actual differences were found to be nonsignificant and would therefore make questionable the possibility that milk fat is the causative agent of "foal heat" scours. The foals in this trial did not all scour during the dam's first estrus and other research has shown that this condition can develop in foals raised away from their dams. These facts would tend to challenge the supposition that "foal heat" scours is attributable to changes brought about by the dam's estral period.

SUMMARY

Under the conditions of this study, highly significant differences ($P < .01$) between the composition of mares' milk were noted for volume, pH and crude protein of milk samples collected on the first 14 days post partum for six Quarter Horse mares. Differences between days were significant ($P < .01$) for total solids and crude protein, while differences for ash were significantly ($P < .05$) different to a lesser extent. Differences in fat and lactose percentages were not significant either between mares or days. The average volume of milk collected did not show any trends, while pH values were lowest at the beginning of the test period, reached a high before 14 days and then declined. Total solids, crude protein, fat, and ash generally declined as a percentage of the milk with increasing stages of lactation, except for a brief rise in the fat concentration at 9 days post partum. Lactose showed an irregularly increasing pattern positively associated with days after parturition. A bio-assay for estrogenic activity indicated possible positive results while a study to detect bacterial contamination of the milk turned up negative results.

All the foals in this study developed the "foal heat" scours symptoms; however, only one of these conditions developed during the dam's estrus. Since evidence has been presented by Stowe of the University of Kentucky and Monson of the Borden Chemical Company that the scouring condition under investigation in this study can develop in foals having no association with their dams or their dam's milk,

further study in the area of "foal heat" scours is warranted. Perhaps the problem of "foal heat" scours could be approached by using better methods of sampling mares' milk, comparing foals suckling their dams with those on milk replacer rations, and utilizing larger numbers of individuals. Such an approach should provide additional evidence for establishing causative agents of "foal heat" scours.

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APPENDIX

TABLE 1. VOLUME OF MILK COLLECTED PER SAMPLE (ML.)

Days post partum	Mare number					
	1	2	3	4	5	6
1	40	240	430	60	72	--
2	86	220	220	100	120	104
3	112	280	92	185	80	120
4	108	120	176	185	152	170
5	108	38	170	220	164	106
6	134	50	185	230	158	166
7	122	60	173	158	60	68
8	94	--	215	150	84	44
9	122	160	195	250	25	37
10	120	110	99	185	160	64
11	90	100	158	180	112	39
12	120	190	115	170	195	58
13	98	200	148	182	128	72
14	140	230	143	180	210	80

TABLE 2. pH OF FRESH MILK SAMPLES

Days post partum	Mare number					
	1	2	3	4	5	6
1	6.30	6.45	6.40	6.30	6.45	--
2	6.29	6.60	6.50	7.00	6.50	6.40
3	6.29	6.55	6.55	6.40	6.52	6.38
4	6.32	6.60	6.50	6.40	6.50	6.45
5	6.35	6.65	6.71	6.45	6.45	6.60
6	6.40	6.70	6.55	6.48	6.46	6.52
7	6.45	6.60	6.55	6.65	6.57	6.65
8	6.48	--	6.72	6.50	6.50	6.57
9	6.40	6.70	6.73	6.49	6.50	6.53
10	6.45	6.60	6.65	6.40	6.60	6.68
11	6.40	6.72	6.60	6.40	6.58	6.39
12	6.40	6.48	6.49	6.45	6.52	6.54
13	6.35	6.50	6.45	6.55	6.70	6.53
14	6.55	6.60	6.45	6.50	6.68	6.51

TABLE 3. FAT AS A PERCENT OF WHOLE MILK

Days post partum	Mare number					
	1	2	3	4	5	6
1	2.648	2.444	1.846	2.292	4.071	--
2	2.498	1.310	2.356	1.622	2.328	1.831
3	2.153	1.505	2.260	2.255	2.153	2.065
4	2.220	1.442	2.142	2.222	1.976	2.269
5	2.060	1.806	1.118	1.875	2.059	2.156
6	1.587	2.961	2.026	1.663	1.687	1.725
7	1.912	1.984	1.828	1.428	1.204	2.102
8	1.965	--	1.641	1.757	1.381	1.683
9	1.941	1.858	1.554	1.380	3.273	1.794
10	1.787	1.323	2.405	2.807	1.381	1.458
11	2.336	1.235	2.578	1.716	1.736	1.497
12	1.541	2.350	3.079	1.756	1.687	1.202
13	2.615	1.304	1.581	2.021	1.716	1.035
14	2.078	1.090	2.051	1.817	1.418	1.663

TABLE 4. LACTOSE AS A PERCENT OF WHOLE MILK

Days post partum	Mare number					
	1	2	3	4	5	6
1	5.8547	5.7144	11.1462	6.5693	8.2183	--
2	5.8680	5.3324	5.6975	6.1922	7.1171	5.8075
3	6.6490	5.6036	6.5652	6.4153	7.0225	6.9908
4	4.7133	6.2563	9.1060	6.8142	6.5089	6.9982
5	6.4763	6.1671	5.9637	6.9331	6.6131	6.8938
6	6.4203	6.7975	7.0568	6.9367	6.5464	6.6767
7	6.6358	6.5047	6.7893	6.4832	6.0755	7.4789
8	6.6213	--	6.9354	6.3979	6.6584	6.7788
9	6.7312	6.7791	7.0194	6.9496	6.4044	6.2577
10	7.0446	6.1518	6.8389	5.9542	6.4414	6.6077
11	6.9801	6.7685	6.5929	6.9050	6.5592	7.0866
12	7.1496	6.4731	6.4321	6.6696	6.8434	6.7046
13	6.6912	6.7784	6.8030	6.8670	6.7820	7.0685
14	6.0999	6.8167	6.1893	6.5549	7.0795	6.9106

TABLE 5. ASH AS A PERCENT OF WHOLE MILK

Days post partum	Mare number					
	1	2	3	4	5	6
1	.4143	.4516	.6748	.4237	.4677	--
2	.5010	.5066	.4685	.5268	.4789	.5425
3	.4990	.5454	.4848	.4137	.4665	.5212
4	.4367	.5607	.5148	.4108	.4821	.5428
5	.5337	.5329	.4783	.5839	.4909	.6022
6	.4867	.4795	.4652	.3993	.4576	.5503
7	.4262	.4733	.5027	.4528	.5275	.3611
8	.4897	--	.5016	.4901	.5206	.4312
9	.5218	.4539	.4696	.5284	.4756	.4993
10	.4394	.3862	.4431	.3778	.5206	.4143
11	.4199	.4735	.4061	.4290	.4738	.4664
12	.4414	.4719	.4839	.5114	.4306	.5274
13	.4248	.5206	.3770	.4210	.3600	.3545
14	.4161	.4893	.5167	.4461	.4205	.4024

TABLE 6. CRUDE PROTEIN AS A PERCENT OF WHOLE MILK

Days post partum	Mare number					
	1	2	3	4	5	6
1	3.629	3.684	4.618	4.211	2.921	--
2	3.616	3.406	2.792	2.773	2.841	3.692
3	3.198	3.364	3.324	3.293	2.846	3.212
4	3.221	3.412	2.860	3.141	3.302	2.980
5	3.398	3.115	2.948	3.255	3.141	2.638
6	3.332	2.828	2.723	3.169	2.890	3.310
7	3.448	2.706	2.928	3.065	3.186	2.740
8	3.213	--	2.441	2.608	2.785	2.367
9	2.851	3.135	2.460	2.583	2.976	2.274
10	2.729	3.082	2.593	2.365	2.782	2.293
11	2.831	2.849	2.389	2.441	2.760	2.606
12	2.875	3.189	2.365	2.663	2.606	2.262
13	2.627	2.690	2.415	2.429	2.681	2.317
14	2.798	2.736	2.763	2.482	2.507	2.235

TABLE 7. TOTAL SOLIDS AS A PERCENT OF WHOLE MILK

Days post partum	Mare number					
	1	2	3	4	5	6
1	12.546	12.294	18.285	13.496	15.678	--
2	12.483	10.555	11.314	11.114	12.765	11.873
3	12.499	11.018	12.634	12.377	12.488	12.789
4	12.591	11.671	12.695	12.588	12.269	12.790
5	12.468	11.621	10.508	12.647	12.304	12.290
6	11.826	13.066	12.271	12.168	11.581	12.262
7	12.422	11.668	12.048	11.429	10.993	12.682
8	12.289	--	11.519	11.253	11.345	11.260
9	12.045	12.226	11.503	11.441	13.129	10.825
10	12.000	10.943	12.280	11.504	11.125	10.773
11	12.567	11.326	11.966	11.491	11.529	11.656
12	12.007	12.484	12.360	11.600	11.567	10.696
13	12.358	11.293	11.176	11.738	11.175	10.775
14	11.392	11.132	11.520	11.300	11.425	11.211