Orotic Acid in Guinea Pig Milk: Changes in Concentration During Lactation

ABSTRACT
The orotic acid concentration in guinea pig milk decreased rectilinearly beginning on d 1 through a lactation duration of 21 d. Concentration was 29 μg/ml on d 1 and 5 μg/ml on d 17. On d 18 to 21 it was less than 1 μg/ml. The regression equation of orotic acid on day of lactation was: Y(μg/ml orotic acid per milliliter milk) equals 32.3 - 1.6X (day of lactation) with a correlation coefficient of .94. Total orotic acid on a daily basis peaked on d 6 at 950 μg and declined to less than 5 μg by d 21. The rectilinear regression equation from d 6 to 21 was as follows: Y(μg/ml orotic acid per milliliter milk) equals -1.64 + .68X (grams of milk production per day) with a correlation coefficient of .97. This may be used as a marker for persistency of lactation.

INTRODUCTION
Orotic acid (6-carboxy uracil) is an intermediate in the biosynthesis of pyrimidine nucleotides (11). Orotic acid exists in milk in concentrations from 0 to 200 μg/ml or more, depending upon species and stage of lactation. Cow's milk contains large but variable concentrations of orotate with an average of 80 μg/ml (10). Accordingly, cow's milk is the major source of orotic acid in the human diet, and it is of nutritional significance in that rats fed a diet supplemented with 1% orotic acid developed fatty livers (7). Robinson (12) and Durschlag and Robinson (8) reported that this response is specific to the rat among several species studied. There are few data on orotic acid content in milk of guinea pig, which is considered as an excellent animal to study milk production and milk composition. To provide more information on orotic composition of milk, samples of guinea pig milk were collected and analyzed on a daily milk production basis through the lactation period.

MATERIALS AND METHODS
Common albino guinea pigs (Camm Research Institute, Wayne, NJ) were used. One milk sample was obtained daily for 21 consecutive d postpartum according to the procedure outlined by Anderson et al. (4). Samples were frozen as soon as possible and stored at -20°C until assayed. Orotic acid was measured on all milk samples in duplicate using the colorimetric method of Larson and Hegarty (10), which was modified from that of Adachi et al. (1). In this procedure, orotic acid is converted to barbituric acid, which is complexed to form a color with para-dimethylaminobenzaldehyde. Daily values were plotted and found to be linear. Therefore, analysis of variance and regression analysis for linear and quadratic models were performed using SAS (13).

RESULTS AND DISCUSSION
The colorimetric procedure for assay of orotic acid was chosen because it is more specific than the microbiological procedures (2, 9). Sensitivity was 1 μg/ml of milk. Analysis of variance revealed that 3.7% of the total was due to variation in duplicates and 96.3% was due to variation among days. Figure 1 shows the orotic acid concentrations on a daily basis throughout the 21-d lactation of the guinea pig. Orotic acid concentrations decreased (P<.01) through the lactation period beginning on d 1 with a concentration of 29 μg/ml and declining to a detectable low of 5 μg/ml on d 17. For the rest of lactation (d 18 to 21), orotic acid was not
detectable (<1 μg/ml). The regression equation calculated from 21 daily means was: Y(micrograms orotic acid per milliliter milk) equals 32.3 - 1.6X (day of lactation) with a correlation coefficient (r) of .94. Previously Anderson et al. (4) showed that the lactation curve in guinea pigs peaked at d 6 or 7 and decreased precipitously afterwards. From d 6 to d 21 of lactation a rectilinear regression resulted between orotic acid concentration and milk production: Y(micrograms orotic acid per milliliter milk) equals -1.64 + .68X (grams of milk/d) with a r of .97. The theoretical lactation curve of the guinea pig, using the equations of Anderson et al. (4), and the actual total orotic acid concentrations are represented in Figure 2. Three equations were found to represent best the model to predict total orotic acid in guinea pig milk: 1) from d 1 to d 6, Y = 242.88 + 135.9D, r = .96; 2) from d 7 to d 11, Y = 1655.3 - 103.4D, r = .97; 3) from d 12 to d 17, Y = 980.8 - 51.3D, r = .93, where D is equal to the day of lactation.

The decrease in orotic acid along the lactation period was also researched by Larson and Hegarty (10); they tested two different samples of guinea pig milk at d 8 and d 15 of lactation and found orotic acid concentrations were 18 and 1 μg/ml, respectively.

The significance of orotic acid concentrations in guinea pig milk may be of a dual nature, one reflecting mammary gland activity and the other reflecting needs of the offspring. Because orotic acid is an intermediate in pyrimidine nucleotide synthesis (6, 11), it may reflect the rate of RNA synthesis, and therefore, protein synthetic activity of mammary secretory cells. As such, it may be a marker to measure the beginning of declining lactation and subsequent rate of persistency of lactation. The physiological significance of the daily decline in orotic acid concentrations may impact upon changes in guinea pig milk ingredients (3, 5) as well as upon mammary gland components (14). It may be useful as an estimator of persistency of lactation in the guinea pig and perhaps some other milk-producing animals.

The alternative explanation for the presence of orotic acid in guinea pig milk relates to its significance as a vitamin (B13) for microorganisms. Guinea pigs are cecal animals. The pigs are born in a very mature condition. The concentrations of orotic acid in guinea pig milk
Figure 2. Theoretical lactation curve (grams per day) of the guinea pig (Δ——Δ); total orotic acid in milk (micrograms) per day (○——○).

are less than those in ruminants and higher than those in monogastric animals. Survival rates are high for guinea pigs allowed to suckle for no more than 7 d. Perhaps this is because the vitamin B₁₃ is sufficient to stimulate growth of flora in the digestive tract, especially in the cecum, to enable the animals to function like an adult when they are no more than a week old.

REFERENCES