Macrominerals in Guinea Pig Milk During 21 Days of Lactation[1]

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ABSTRACT

Milk samples were obtained daily from English short-hair albino guinea pigs for 21 d. Analyses included six macrominerals: Ca, P, K, chloride, Na, and Mg (in order of decreasing concentration). All minerals except K gradually increased in concentration from the beginning to the end of lactation. Calcium concentration began at 38 m*M* on d 1 and was 78 m*M* on d 21. The pattern of increase was quadratic: Y (m*M*) = 39 −.48X (day of lactation) + .11 X^2. Phosphorus concentration was 38 m*M* on d 1 and highest at 51 m*M* on d 21. Chloride was 19 m*M* on d 1 and 68 m*M* on d 21. Sodium was 13 m*M* on d 1 and highest at 42 m*M* on d 21. Magnesium was 11 m*M* on d 1 and was highest on d 18 (13 m*M*). However, K was 31 m*M* on d 1, reached a high of 33 m*M* on d 3, and was lowest on d 19 (12 m*M*). These changes in concentration and previously reported volume changes suggest alterations in functional capacities of ionic transport mechanisms of secretory cell membranes in this species.

INTRODUCTION

Macrominerals in milk provide the primary mineral requirements of the neonate during the critical period after birth when it cannot forage for itself. These minerals are also needed in adult humans in quantities sufficiently high that cow milk provides these in a convenient package and in a highly utilizable form. In an effort to enhance understanding of the physiological mechanisms that control quantity and quality of milk in the cow, the guinea pig was chosen as a model.

Declines in quantity of milk during a lactation have been observed in many species (13, 14). Reasons for production losses (lack of persistency) over the lactation period are not documented clearly but are thought to be related to gradual losses in secretory cells and to decreases in hormonal stimuli. The milk production of the guinea pig has been determined (2). Lactation curves show very rapid changes in the quantity of milk over 3 wk. Typically, milk yield declines from a peak of 40 g/d on d 7 to 20 g/d by d 14 and only 3 g/d by d 21. Only part of the decline is attributable to loss in cell members (12). Quality of milk changes rapidly during this period of rapid change in milk volume. Percentage protein, fat, and ash increase as milk production declines, whereas lactose concentration appears to control to some extent the rapid volume decline (1).

Of considerable interest are the macrominerals in milk as they relate to dietary needs of neonatal mammals. Among these are Ca, P, and Mg. Because of the dramatic changes in guinea pig milk volume (2) and composition of protein, fat, lactose, and ash (1), the guinea pig may be a valuable model in which to study mechanisms regulating milk composition. As part of such studies, detailed information on the changes in mineral composition of guinea pig milk is needed. Therefore, the objective of this study was to determine changes in macrominerals (i.e., Ca, P, K, Na, Cl, and Mg) in guinea pig milk throughout a normal lactation. The purpose of the present study was to determine the extent of changes in concentrations of macrominerals in a species known to have rapid quantitative and qualitative changes in milk production over 21 d. As an outcome of the study, hypotheses were developed to

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suggest reasons for the inordinate vacillations in concentrations of minerals.

MATERIALS AND METHODS

Common albino guinea pigs (Camm Research Institute, Wayne, NJ) served as the foundation stock. Guinea pigs were fed Purina (St. Louis, MO) pellets and were provided with fresh water at all times. Samples were obtained from lactating guinea pigs according to procedures reported previously using a milking apparatus designed in our laboratory (9). Samples were placed in acid-washed glass vials and frozen until mineral analyses could be made.

Calcium, P, K. Na, and Mg were measured in duplicate at the University of Missouri Agricultural Experiment Station Chemical Laboratory using methods published by the Association of Official Analytical Chemists (3). Chloride was measured in our laboratory using an Orion chloride electrode (6).

Volumes of milk required for analyses varied with the procedure, 2 ml being the most common amount used. Numbers of individual samples analyzed varied from 3 to 12/d with totals over the 21-d period of Ca 185; P 184, K 191, Na 180, Mg 191, and chloride 176. Each observation represented the mean of duplicate samples from single animals on a single day; none was the result of pooled samples from several guinea pigs or from several days production pooled from the same guinea pig. Results were expressed either as percent, Milligrams per milliliter or as millimoles.

Upon completion of the chemical analyses and tabulation, data were subjected to statistical tests. Each mineral was regressed on day of lactation. Statistical analyses were by SAS release 79.6 using an Amdahl 470 V/7 Digital Computer (11). Procedures for SAS included ANOVA, systems regression, and correlation. Regression equations examined included linear, quadratic, and cubic models. Equations in which the highest order term was significant were selected to describe changes in milk mineral content during lactation.

RESULTS

Based upon 185 observations for Ca concentration in individual guinea pig milk samples taken daily for 21 consecutive d, regression equations for best fit were developed. Of the three models tested, the quadratic model resulted in the best fit with a coefficient of determination (R^2) of .72 ($P<.01$). With Y as the percent Ca and X as the day of lactation, the following equation was generated: $Y = .154 -.0019X + .0045X^2$. Expressed on a molar basis, the equation was $Y = 39 -.48X$ (day of lactation) $+ .11\ X^2$. Calcium concentration was 39 m*M* (1.52 mg/ml) on d 1, with a low of 36 m*M* (1.44 mg/ml) on d 3, and a high of 78 m*M* (3.12 mg/ml) on d 21. It averaged 58 m*M* (2.3 mg/ml) for the 21 d (Figure 1).

Phosphorus concentrations in guinea pig milk were more variable than concentrations of Ca as evidenced by the R^2 of .40 (Figure 2). However, this quadratic model was significant ($P<.05$). Phosphorus concentration was 38 m*M* (1.17 mg/ml) on d 1, dropped to 36 m*M* (1.12 mg/ml) on d 6, and rose gradually to a high of 51 m*M* (1.57 mg/ml) on d 21 with an average of 40 m*M* (1.24 mg/ml) for the 21-d period. The magnitude of change for P was only 34.8% (from a low of 36 m*M* to a high of 51 m*M*), whereas the change in Ca was 117% (from a low of 36 m*M* to 78 m*M*).

The best arithmetic fit of the data for K was a cubic equation. Potassium was 31 m*M* (1.22 mg/ml) on d 1. It reached a high of 33 m*M* (1.28 mg/ml) on d 3 and d 4, after which it declined slowly to a low of 12 m*M* (.48 mg/ml) on d 19 (Figure 3). The mean concentration for 21 d was 23 m*M* (.89 mg/ml). The change from the maximum to the minimum concentration represented a decrease of 64% (33 to 12 m*M*).

Chloride in guinea pig milk showed a cubic response in a positive direction (Figure 4); that is, it increased through the lactation, but K decreased. The R^2 was .72 ($P<.01$). Chloride concentration was 19 m*M* (.67 mg/ml) on d 1, dropped to a low of 18 m*M* (.64 mg/ml) on d 2, and reached a peak of 68 m*M* (2.42 mg/ml) on d 21. There was an increase of 279% during the lactation (18 to 68 m*M*). The mean concentration of chloride over the 21-d lactation was 43 m*M* (1.52 mg/ml).

Sodium concentration increased in a manner similar to chloride. The best equation was a cubic expression (Figure 5) with a R^2 of .73 ($P<.01$). On d 1, the Na concentration was 13 m*M* (.30 mg/ml), but it reached a low point on d 4 (8.5 m*M* or .20 mg/ml). The high was reached on d 21 at 42 m*M* (.97 mg/ml). This repre-

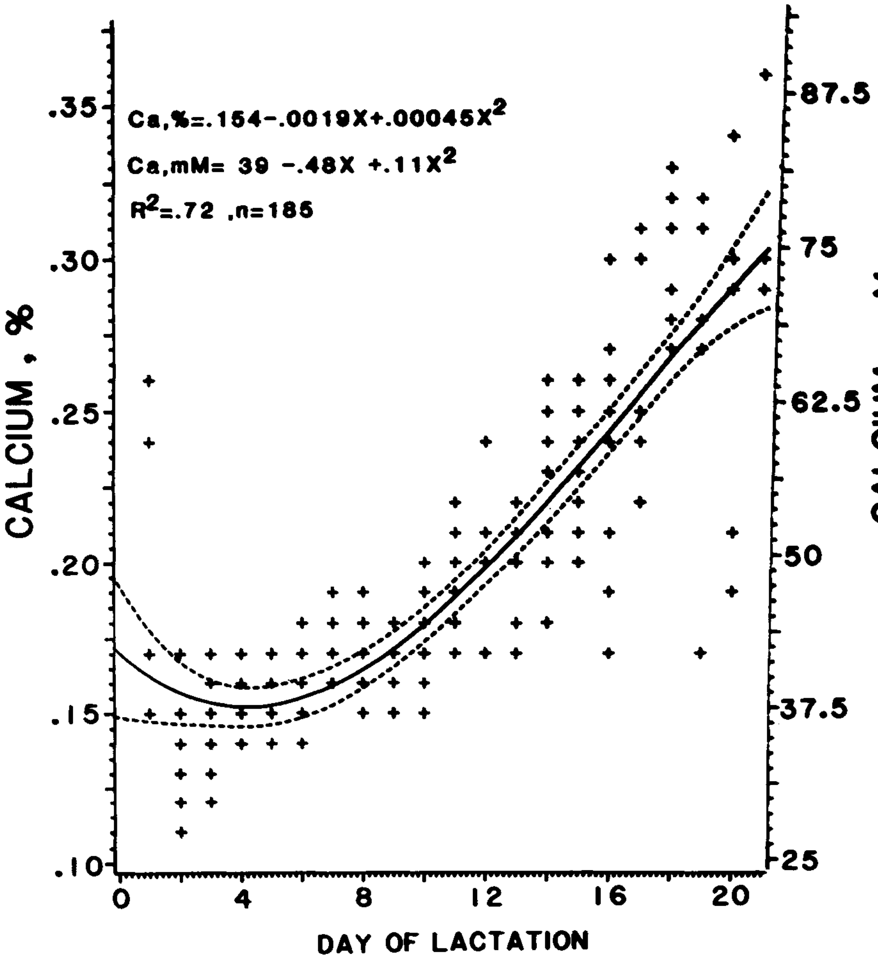

Figure 1. Concentration of Ca in guinea pig milk on a daily basis for 21 d (X is day of lactation). Individual data points are indicated by crosses, solid line indicates least squares regression, and dashed lines indicate 95% confidence limits.

sented a 397% increase (8 to 42 m*M*). Mean concentration of Na during the lactation was 20 m*M* (.47 mg/ml).

Magnesium concentration increased according to a cubic mathematical model (Figure 6). The coefficient of determination (R^2) was only .24, indicating a poor fit of the data points. Nonetheless, it was significant at 5% probability. Magnesium concentration was 11 m*M* (.28 mg/ml) on d 1. It was reduced to a low of 8.8 m*M* (.22 mg/ml) on d 6 and reached a high of 13 m*M* (.32 mg/ml) on d 18. The magnitude of increase was 47.4% (8.8 to 13 m*M*). The mean for 21 d was 11 m*M* (.27 mg/ml).

DISCUSSION

Concentrations of macrominerals in guinea pig milk may be compared with those in milk of the dairy cow (8). Potassium is 35 m*M* (1.38 mg/ml) in milk of cows and only 23 m*M* in the guinea pig when averaged over 21 d. If the highest daily concentration in guinea pig milk is used, it is 33 m*M*, a concentration not too different from that in milk of the cow. Calcium ranks second in concentration in milk of dairy cows at 30 m*M* (1.20 mg/ml). This compares with 51 m*M* in the guinea pig when averaged over 21 d. At the low point on d 3, Ca in guinea pig milk is 36 m*M* which is only slightly higher

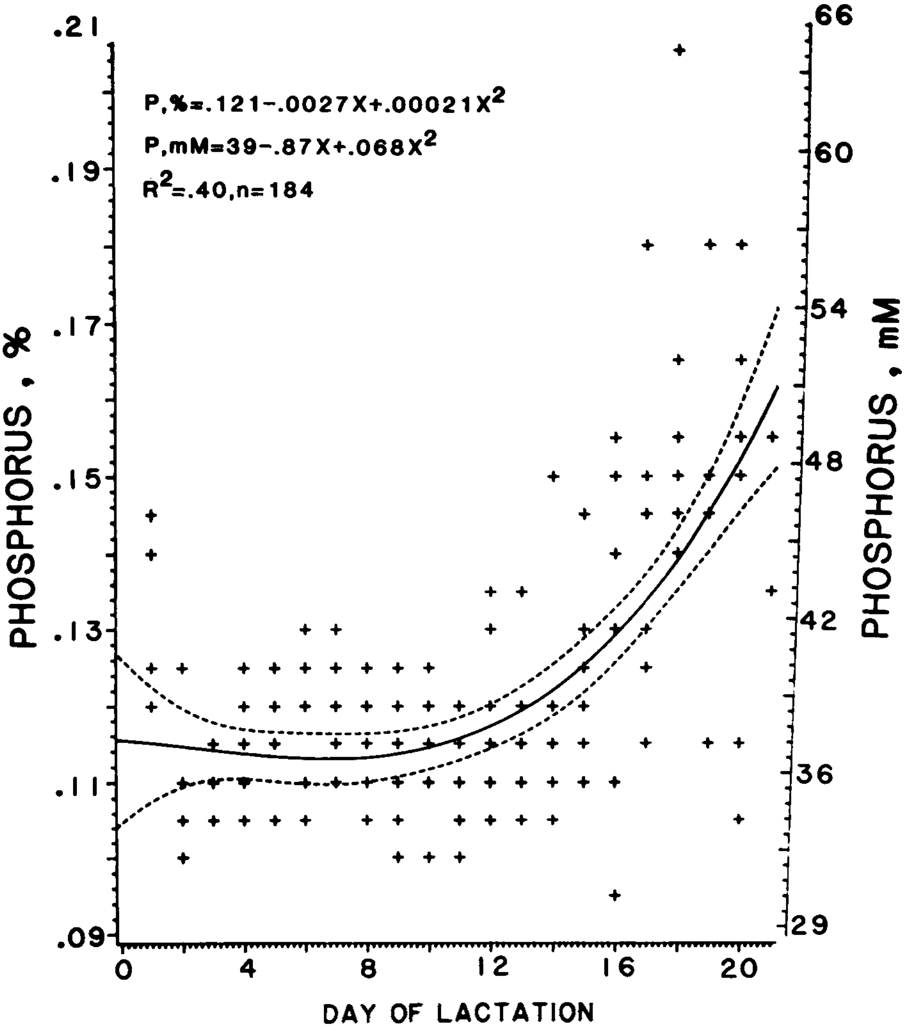

Figure 2. Concentration of P in guinea pig milk on a daily basis for 21 d (X is day of lactation). Individual data points are indicated by crosses, solid line indicates least squares regression, and dashed lines indicate 95% confidence limits.

than Ca in cow milk. Because guinea pig milk contains an average of 8.9% protein (ranging from 6.2% on d 1 of lactation to 11.7% on d 21) (1), versus 3.1 to 3.6% for cow milks, and because milk Ca is complexed to milk proteins, guinea pig milk could be expected to have more Ca than does cow milk. Unfortunately, although the Ca-binding properties of cow milk proteins have been researched extensively, very little is known about the interactions between Ca and milk proteins in guinea pig milk. Therefore, quantitative analysis of the relative changes in milk protein and milk Ca in guinea pigs must await further research into the chemistry of guinea pig milk proteins. Chloride is third in concentration in cow milk at 25 m*M* (.874 mg/ml). This is approximately 60% of the mean concentration of 43 m*M* in guinea pig milk. Phosphorus ranks fourth in cows milk at 26 m*M* (.80 mg/ml), approximately two-thirds of the 40 m*M* found in guinea pig milk. In cow milk, P is present as protein bound phosphate.

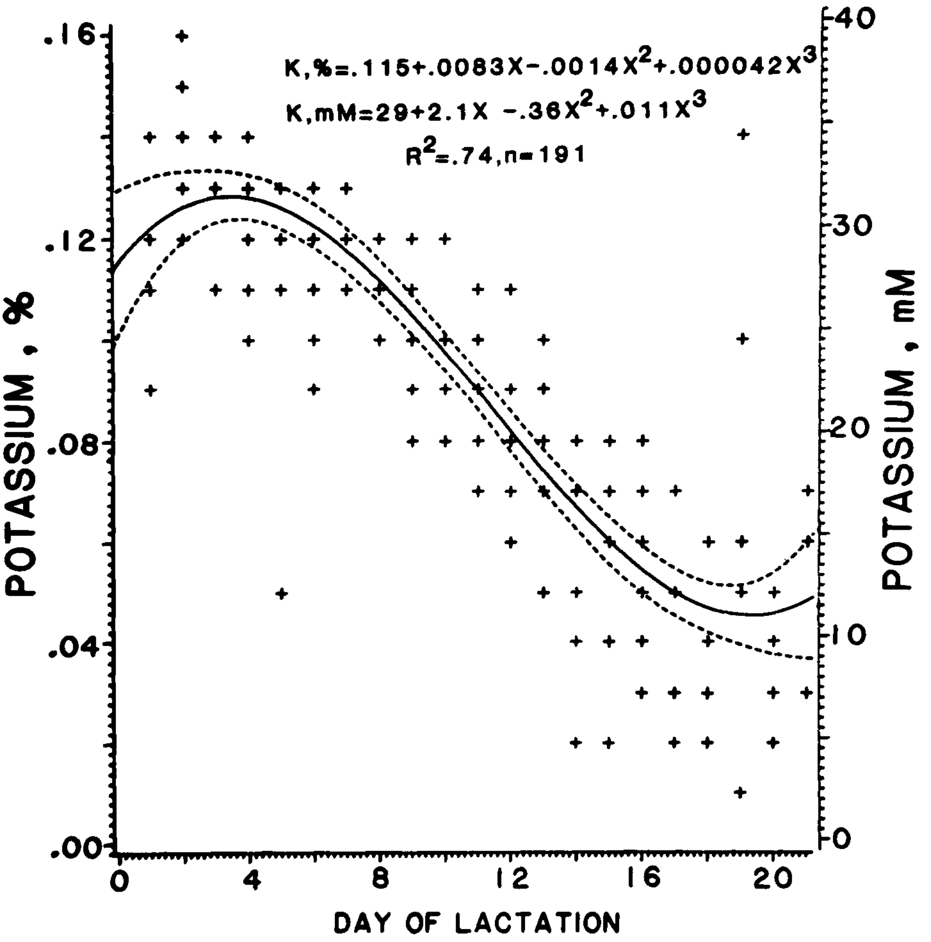

Figure 3. Concentration of K in guinea pig milk on a daily basis for 21 d (X is day of lactation). Individual data points are indicated by crosses, solid line indicates least squares regression, and dashed lines indicate 95% confidence limits.

If this is also true in guinea pig milk, guinea pig milk might be expected to contain substantially more P than does cow milk. However, specific phosphoproteins have not been studied in guinea pigs as they have been in cows. Therefore, quantitative comparisons of phosphate-protein interrelations in cow and guinea pig milk must await further research in the chemistry of guinea pig milk proteins. Sodium ranks fifth in the order of mineral concentration in both cow and guinea pig milk. It is slightly higher in the former at 25 m*M* (.58 mg/ml) than in the latter at 20 m*M* (.47 mg/ml). Sixth and last among the macrominerals measured in this study is Mg. Magnesium averages 5 m*M* (.12 mg/ml) in cow milk and more than double that in guinea pig milk at 11 m*M* (.27 mg/ml).

Curvilinear changes in mineral concentrations during the short period of lactation in the guinea pig may aid the understanding of mineral transfer via the mammary epithelial cell. Much has been researched in this area (7) and more may be gleaned from the present study. Only K declined in concentration as the lactation progressed over time. It reached a high concentration in milk of the guinea pig on d 3 and 4. The decline on d 5 and thereafter may have been a signal for later events, i.e., peak

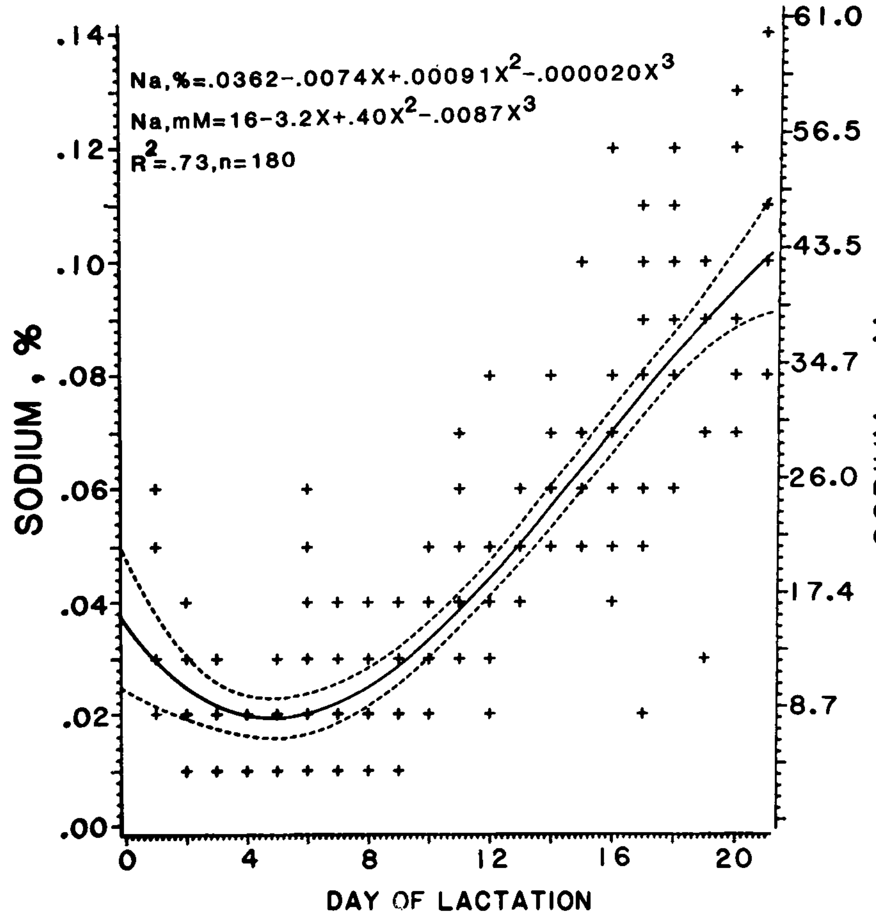

Figure 4. Concentration of Na in guinea pig milk on a daily basis for 21 d (X is day of lactation). Individual data points are indicated by crosses, solid line indicates least squares regression, and dashed lines indicate 95% confidence limits.

milk production was reached on d 7, followed by a rapid decline to d 18 (2). At the same time that K reached a high concentration, Na concentration dropped to a low of 8.5 m*M* on d 4. The concomitant changes in these minerals may have reflected a deficiency at the basal cell membrane in most secretory cells to maintain the Na-K pump at maximum. Thereafter, the changes in Na and K in milk suggested a gradual and continual deterioration in the Na-K pump. An in vitro study using mouse mammary epithelial cells indicates that prolactin increases transepithelial Na transport, tending to force Na out of the milk (4). Although periparturient prolactin concentrations have not been studied in the guinea pig, this hormone may decline rapidly in early lactation in this species. Inability of the pump to function properly early in lactation may partly explain the reason for a premature decline in the lactose concentraton of guinea pig milk reported in an earlier study (1). Alternatively, these changes in milk Na and K concentrations may reflect an increase in paracellular as opposed to transcellular transport of ions during late lactation. Peaker (10) reviewed ion transport in the mammary gland. One indication of paracellular ion transport is high milk Na and chloride coupled with low milk K and lactose. Such changes in milk composition are quite striking during late lactation

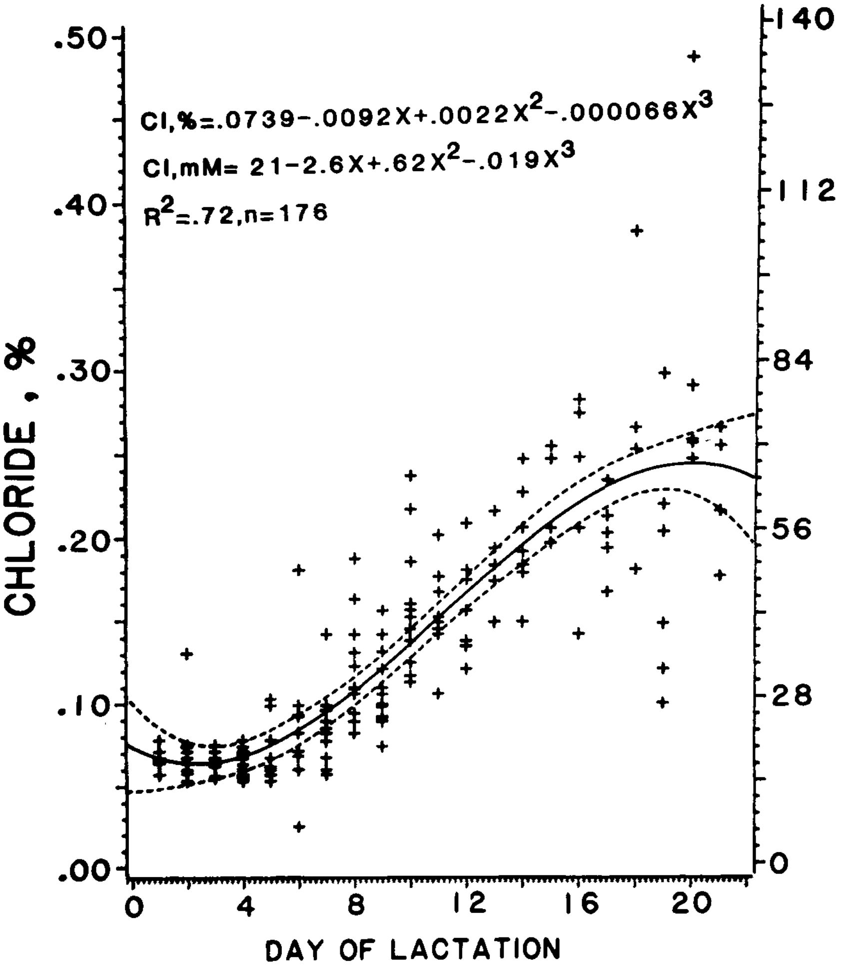

Figure 5. Concentration of ClO_3 in guinea pig milk on a daily basis for 21 d (X is day of lactation). Individual data points are indicated by crosses, solid line indicates least squares regression, and dashed lines indicate 95% confidence limits.

in the guinea pig. These findings may lead to answering why milk production cannot be maintained at a peak with 100% persistency. The changes shown in guinea pig milk are much more subtle in cows milk, just as the decline in the lactation curve is much more gradual in the cow than in the guinea pig.

Changes in chloride concentrations of guinea pig milk were similar to changes in Na concentration, but were slightly smaller in magnitude (397% increase in Na vs. 300% increase in chloride. These changes may represent increased paracellular ion transport, as discussed previously. Alternatively, chloride concentration may also be regulated by ionic effects or by a chloride pump. Although much less is known about chloride pumps, Peaker (10) proposed that a chloride pump in the apical mammary membrane may reabsorb chloride from milk. However, there is little experimental evidence for such a mechanism controlling milk chloride.

Calcium concentrations of guinea pig milk increase as lactation progresses, but the mechanisms controlling this mineral appear to be quite different than those discussed. Milk contains very little free Ca. Most milk Ca is complexed with protein or citrate. Although the existence of Ca pumps has been clearly

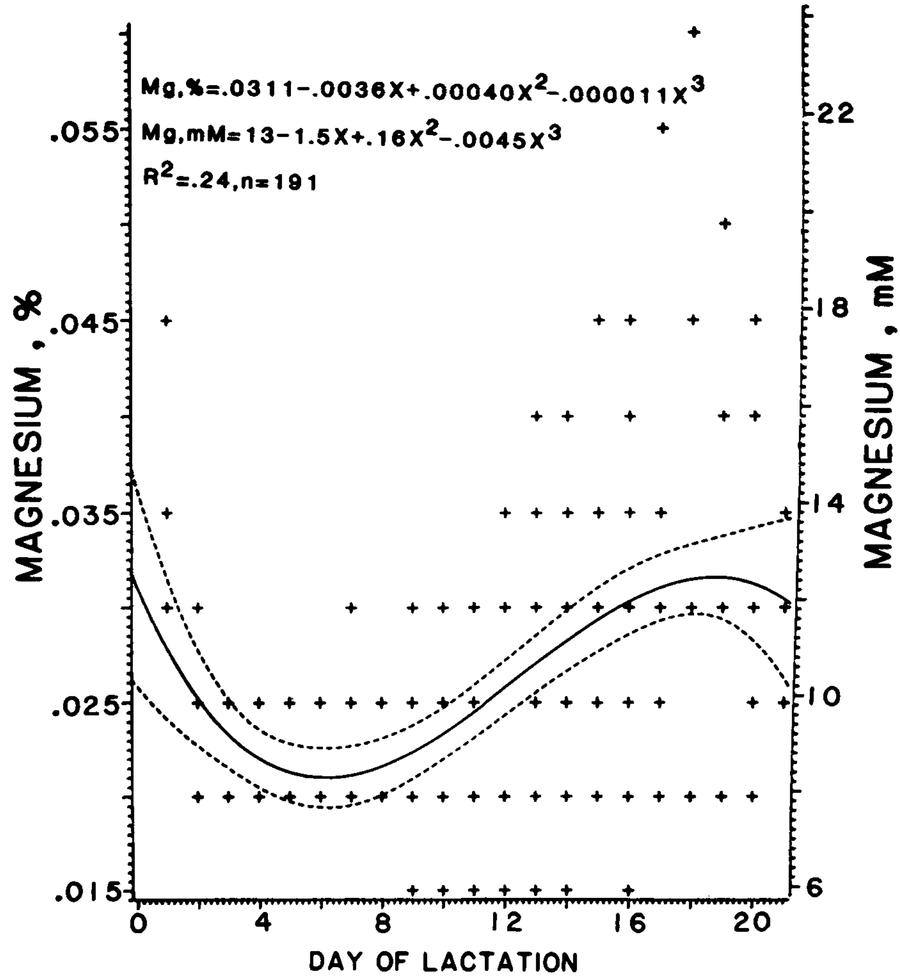

Figure 6. Concentration of Mg in guinea pig milk on a daily basis for 21 d (X is day of lactation). Individual data points are indicated by crosses, solid line indicates least squares regression, and dashed lines indicate 95% confidence limits.

demonstrated (5, 10), changes in the activity of such pumps may not be responsible for increased concentration of Ca in guinea pig milk during late lactation. Instead, the Ca may be transported along with milk proteins. Because the protein concentration of guinea pig milk increases as lactation progresses, this may account for the increase in milk Ca that occurs during lactation. Similar considerations also appear to apply to milk P and Mg. As discussed, much more research on the chemistry of guinea pig milk proteins is needed before quantitative explanations of these processes are possible.

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