Serum Biochemical and Hematological Parameters in Crossbred Swine from Birth Through Eight Weeks of Age

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SUMMARY

Nineteen serum biochemical and seven hematological parameters were determined for crossbred swine from birth through eight weeks of age. From birth (before nursing) to eight hours (after ingestion of colostrum) of age, there was an increase in concentrations of serum total protein, blood urea nitrogen and total bilirubin and increased activities of glutamic-oxaloacetic transaminase, alkaline phosphatase and lactic dehydrogenase. There was a decrease in serum sodium, chloride and potassium concentrations, hemoglobin concentration and packed cell volume during the same period. There was an increase in both serum potassium concentration and erythrocyte count from five (weaning) to six weeks of age. At the same time, there was a decrease in serum sodium and chloride concentrations. The mean concentration of serum cholesterol did not change during the first 24 hours of neonatal life; however, it increased during the 24 to 72 hour period with a linear decrease to six weeks of age.

RÉSUMÉ

Les auteurs ont déterminé 19 paramètres biochimiques sérologiques et sept paramètres sanguins chez des porcelets croisés, à compter de leur naissance et jusqu'à l'âge de huit semaines. Ils notèrent, de la naissance (avant l'allaitement) jusqu'à huit heures (après l'ingestion du colostrum), une augmentation de la concentration des protéines sériques totales, de l'azote uréique sanguin et de la bilirubine totale, ainsi qu'une activité accrue de la transaminase glutamique-oxaloacétique, de la phosphatase alcaline et de la déshydrogénase lactique. Ils observèrent aussi une diminution de la teneur du sérum en sodium, en chlorure et en potassium, coïncidant avec une baisse du taux d'hémoglobine et de l'hématocrite.

De la cinquième (sevrage) à la sixième semaine, la teneur du sérum en potassium et le nombre d'hématies étaient plus élevés; en même temps, la teneur du sérum en sodium et en chlorure subissait une baisse.

La concentration moyenne du cholestérol sérique demeura stable au cours des 24 heures qui suivirent la naissance. Elle augmenta cependant de la 24e à la 72e heure après la naissance, pour ensuite décroître de façon linéaire jusqu'à la sixième semaine.

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INTRODUCTION

Numerous studies have been conducted to determine baseline values for serum biochemical and hematological parameters; however, few experiments have been designed to evaluate the interrelationships of blood constituent changes. The use of swine in pediatric research studies (5) has necessitated multivariate analyses of blood and serum from neonatal pigs. Definition of normal changes in blood parameters will enable scientists to determine the feasibility of using swine as biomedical research subjects for evaluating the mechanisms of disease processes. This study was designed to assess changes in 19 serum biochemical and seven hematological parameters, in crossbred swine, from birth through 56 days of age.

MATERIALS AND METHODS

The crossbred (Hampshire x Duroc) pigs used in this study were born in February. They were housed in a concrete building with an earth floor. Each pig was given a single intramuscular injection of 30 mg of iron, as ferric ammonium citrate, at seven days of age. Males were castrated at three days of age. All pigs were given access to a 19% protein diet at ten days of age and when weaned at five weeks of age, were given a 16% protein diet.

Each pig was weighed, placed in a dorsal recumbent position (2) and blood was collected from the jugular vein. Pigs from seven through 56 days of age were sampled between six and seven a.m., after a 12 hour fast. At birth, at eight hours and at one and three days of age, different pigs were used for each sample. From seven through 56 days of age, samples were collected from the same 40 pigs; however, they were different from those used for the first three days. The numbers of pigs sampled at birth, at eight hours and at one and three days of age were 17, 16, 28 and 21, respectively.

Of the 13 ml blood sample, 2 ml, with EDTA-K as the anticoagulant, were used for hematological determinations. The remaining 11 ml were allowed to clot at room temperature for four hours prior to separation of the serum by centrifugation at 12,500 X g and 2°C for ten minutes. Each serum sample was frozen and stored in a glass vial at -10°C until analyzed.

Serum total protein (TP), glutamic-oxaloacetic transaminase (OT), alkaline phosphatase (AP), lactic dehydrogenase (LDH), calcium (Ca), inorganic phosphorus (IP), chloride (Cl), blood urea nitrogen (BUN), glucose (Glu), total bilirubin (TB) and cholesterol (Chol) were determined with a modified survey model Sequential Multiple AutoAnalyzer (SMA-12/30) (20). Calcium/phosphorus ratios were calculated. Sodium (Na) and potassium (K) concentrations were determined by flame photometry[1]. The protein fractions were separated into albumin (Alb), *alpha*-globulin (α-Glob), *beta*-globulin (β-Glob) and *gamma*-globulin (γ-Glob) on cellulose polyacetate strips, stained with Ponceau S and quantitated with a densitometer[2]. Total globulin (TG) concentrations were calculated.

Erythrocyte (RBC) and leukocyte (WBC) counts were made with an electronic cell counter[3]. Packed cell volumes (PCV) were determined by the microhematocrit method and hemoglobin concentrations (Hgb) were determined using the cyanmethemoglobin method[4]. Mean corpuscular volumes (MCV), mean corpuscular hemoglobins (MCH) and mean corpuscular hemoglobin concentrations (MCHC) were calculated. Mathematical calculations and statistical interpretations were accomplished according to Steel and Torrie (19).

RESULTS

Mean concentrations, or activities, of serum biochemical constituents and hemotological parameters, at birth and at one and eight weeks of age, are depicted in Table I. Mean body weights at birth and at 0.33, one, three, seven, 14, 21, 28, 35, 42, 49 and 56 days of age were 1.2, 1.4, 1.6, 2.1, 2.8, 3.5, 4.7, 6.1, 7.1, 8.5 and 10.3 kg, respectively.

The mean concentration of serum TP (Fig. 1) more than doubled from birth to eight hours of age. From seven through 56 days of age, the mean concentration of serum TP was 5.9 g/100 ml. The significant ($P<0.01$) increase in TP, from birth to eight hours of age, was a result of the significant ($P<0.01$) increase in γ-Glob concentration from birth to eight hours of

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[1]Model 105; Beckman Instruments, Fullerton, California.

[2]Gelscan, Gelman Instrument Co., Ann Arbor, Michigan.

[3]Model B; Coulter Electronics, Hialeah, Florida.

[4]Unopettes; Becton-Dickinson, Rutherford, New Jersey.

TABLE I. Means, Standard Errors of Means and Ranges of Serum Biochemical and Hematological Values for Crossbred Swine from Birth through Eight Weeks of Age

Parameter	Age								
	At Birth			1 Week			8 Weeks		
	Mean	S.E.	Range	Mean	S.E.	Range	Mean	S.E.	Range
Total protein, g/100 ml	2.98	0.12	2.3 - 4.2	6.31	0.08	4.8 - 8.0	5.66	0.10	4.4 - 7.6
Albumin, g/100 ml	0.59	0.04	0.39 - 0.94	1.81	0.44	0.90 - 2.37	2.49	0.04	1.94 - 3.45
Alpha-Globulin, g/100 ml	1.25	0.14	0.32 - 2.08	1.61	0.05	0.88 - 2.72	1.43	0.03	0.99 - 1.73
Beta-Globulin, g/100 ml	0.74	0.10	0.40 - 1.57	1.33	0.03	0.59 - 2.00	1.07	0.03	0.77 - 1.43
Gamma-Globulin, g/100 ml	0.45	0.04	0.16 - 0.75	1.54	0.06	0.52 - 2.50	0.68	0.04	0.37 - 2.08
Glutamic-oxaloacetic transaminase, K.U.	23.7	2.4	14 - 42	26.5	0.8	15 - 43	42.8	1.5	25 - 67
Alkaline phosphatase, K.-A.U.	92.3	6.7	58 - 155	61.4	2.9	22 - 122	26.9	0.6	20 - 37
Lactic dehydrogenase, W.U.	140.1	9.5	87 - 230	306.6	6.2	210 - 428	320.2	7.3	222 - 415
Calcium, mg/100 ml	11.84	0.22	10.2 - 14.0	12.32	0.12	10.7 - 14.8	11.60	0.10	9.4 - 12.5
Inorganic phosphorus, mg/100 ml	5.49	0.20	4.1 - 6.9	8.65	0.14	6.2 - 11.1	8.69	0.14	6.4 - 10.1
Sodium, mEq/l	142.0	1.8	133 - 156	138.4	0.8	127 - 155	140.3	0.9	133 - 165
Chloride, mEq/l	108.5	0.6	106 - 112	107.4	0.5	93 - 113	104.6	0.4	93 - 109
Potassium, mEq/l	6.17	0.33	4.3 - 8.6	5.45	0.08	4.1 - 7.2	5.75	0.10	4.9 - 7.2
Blood urea nitrogen, mg/100 ml	16.3	1.2	10 - 22	14.0	0.6	6 - 23	16.9	0.6	10 - 24
Glucose, mg/100 ml	113.9	6.9	74 - 169	130.6	3.5	59 - 171	99.1	2.2	82 - 130
Total bilirubin, mg/100 ml	0.25	0.02	0.2 - 0.5	0.53	0.02	0.2 - 0.7	0.19	0.01	0.1 - 0.3
Cholesterol, mg/100 ml	84.9	7.4	40 - 147	179.8	6.9	102 - 320	117.3	2.2	92 - 145
Leukocyte count X 10^3, cells/mm^3	9.62	0.66	6.29 - 15.75	12.66	0.56	6.25 - 25.50	19.64	0.63	13.34 - 28.26
Hemoglobin, g/100 ml	12.18	0.33	9.7 - 14.4	8.32	0.21	4.9 - 11.5	9.01	0.11	8.2 - 10.8
Packed cell volume, %	42.0	1.5	26.8 - 50.5	28.1	0.6	17.0 - 38.2	30.0	0.3	25.8 - 36.0
Erythrocyte count X 10^6, cells/mm^3	5.29	0.20	4.18 - 7.09	4.65	0.10	3.55 - 7.41	4.60	0.08	3.16 - 5.65
Mean corpuscular volume, μ^3	79.6	1.9	58.1 - 90.8	60.8	1.6	38.2 - 94.3	66.0	1.4	54.6 - 97.5
Mean corpuscular hemoglobin, $\mu\mu g$	23.2	0.7	19.3 - 31.2	17.9	0.5	10.7 - 23.7	19.8	0.4	16.2 - 29.4
Mean corpuscular hemoglobin concentration, %	29.4	0.7	26.7 - 36.9	29.8	0.8	17.0 - 39.9	30.0	0.2	27.5 - 32.0

age (Fig. 2). However, the mean concentration of serum Alb did not change markedly during this same period of time. Serum concentrations of α-Glob and β-Glob averaged 1.47 and 1.11 g/100 ml, respectively, from one through eight weeks of age.

Mean serum OT, AP and LDH activities (Fig. 3) increased significantly ($P<0.01$) between birth and eight hours of age. The mean activities of OT and LDH were 33 Karmen Units (K.U.) and 281 Wacker Units (W.U.), respectively, from one through eight weeks of age. Mean serum AP activity decreased significantly ($P<0.01$) from eight hours to three weeks of age and averaged 25 King-Armstrong Units (K.-A.U.) from three through eight weeks of age.

The mean concentration of serum Ca increased significantly ($P<0.01$) from 24 hours to three days of age (Fig. 4). The mean concentration of serum IP increased significantly ($P<0.01$) from three days to one week of age. The average concentrations of serum Ca and IP were 11.9 and 8.3 mg/100 ml, respectively, from one through eight weeks of age. The mean Ca/P ratio, from one through eight weeks of age, was 1.43.

Mean serum Na, K and Cl concentrations decreased significantly ($P<0.01$) from birth to eight hours of age (Fig. 5). When the pigs were weaned, the mean concentrations of serum Na and Cl decreased significantly ($P<0.01$) whereas the mean concentration of serum K increased significantly ($P<0.01$). The mean concentration of serum BUN increased significantly ($P<0.01$) from birth to 24 hours of age and then decreased significantly ($P<0.05$) by three days of age.

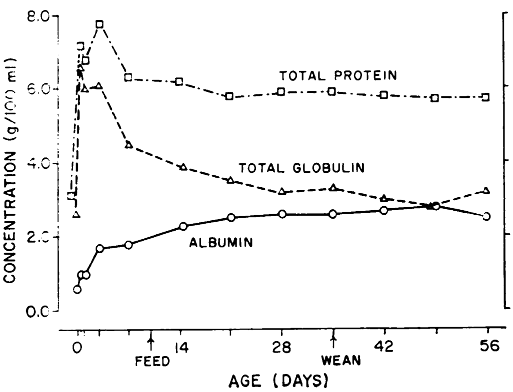

Fig. 1. Mean concentrations of total protein, total globulin and albumin in serum from crossbred swine from birth through eight weeks of age.

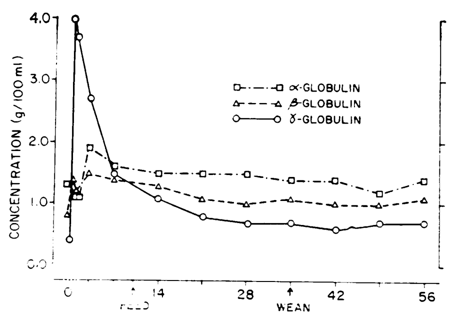

Fig. 2. Mean concentrations of α-globulin, β-globulin and γ-globulin in serum from crossbred swine from birth through eight weeks of age.

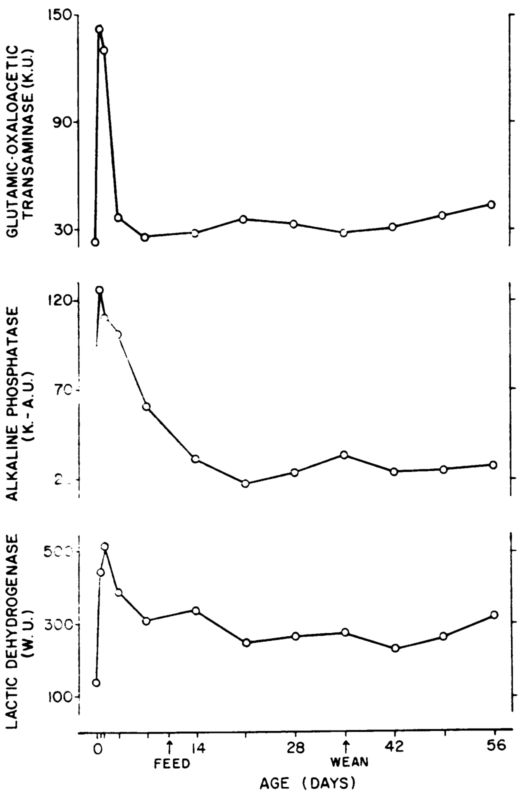

Fig. 3. Mean activities of glutamic-oxaloacetic transaminase (Karmen Units), alkaline phosphatase (King-Armstrong Units) and lactic dehydrogenase (Wacker Units) in serum from crossbred swine from birth through eight weeks of age.

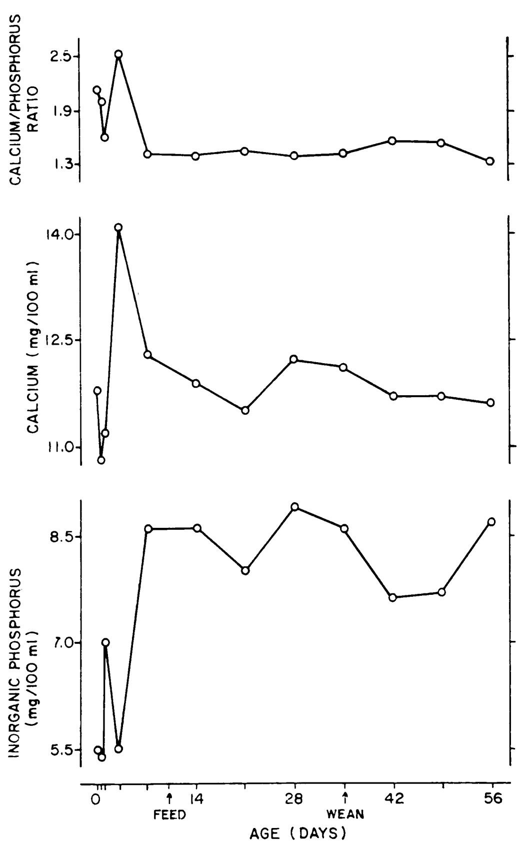

Fig. 4. Mean concentrations of inorganic phosphorus and calcium, as well as mean calcium/phosphorus ratios, in serum from crossbred swine from birth through eight weeks of age.

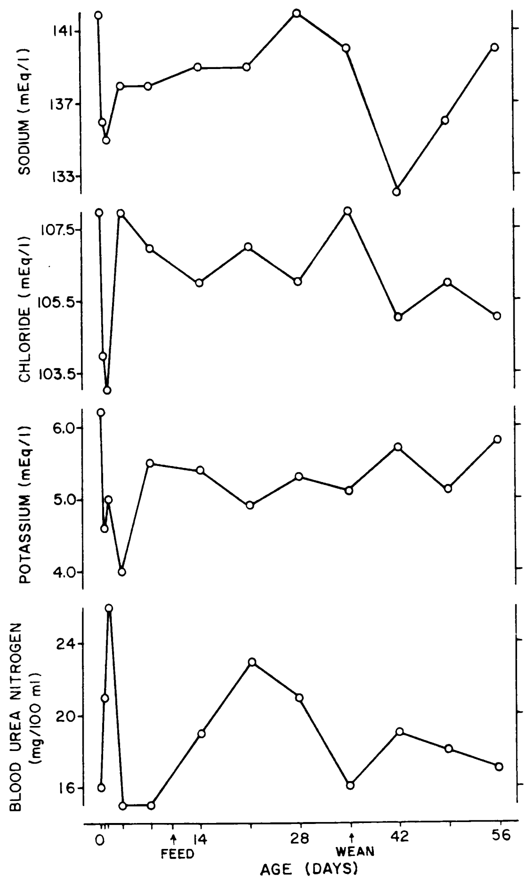

Fig. 5. Mean concentrations of sodium, chloride, potassium and blood urea nitrogen in serum from crossbred swine from birth through eight weeks of age.

The mean concentration of serum Chol was 84 mg/100 ml during the first 24 hours of age; there was a significant ($P<0.01$) increase to 180 mg/100 ml at three days of age (Fig. 6). There was a significant ($P<0.01$) linear decrease from 180 mg/100 ml at three days to 83 mg/100 ml at six weeks of age. The mean concentration of serum TB increased significantly ($P<0.01$) from birth to 24 hours of age; subsequent to that time, there was a significant ($P<0.05$) curvilinear decrease to five weeks of age. The mean concentration of serum Glu was 116 mg/100 ml for the overall study.

Mean Hgb and PCV decreased significantly ($P<0.01$) from birth to eight hours of age (Fig. 7). Mean RBC decreased less rapidly than did Hgb or PCV. However, mean RBC decreased significantly ($P<0.01$) from birth to 72 hours of age. Mean RBC increased significantly ($P<0.01$) from weaning (five weeks) to six weeks of age. MCV, MCH and MCHC values (Fig. 8) were variable throughout the course of the study. As a result of the increase in RBC between five and six weeks of age, MCV and MCH decreased significantly ($P<0.01$) from five to six weeks of age. Mean WBC increased gradually throughout the duration of the experiment (Fig. 7).

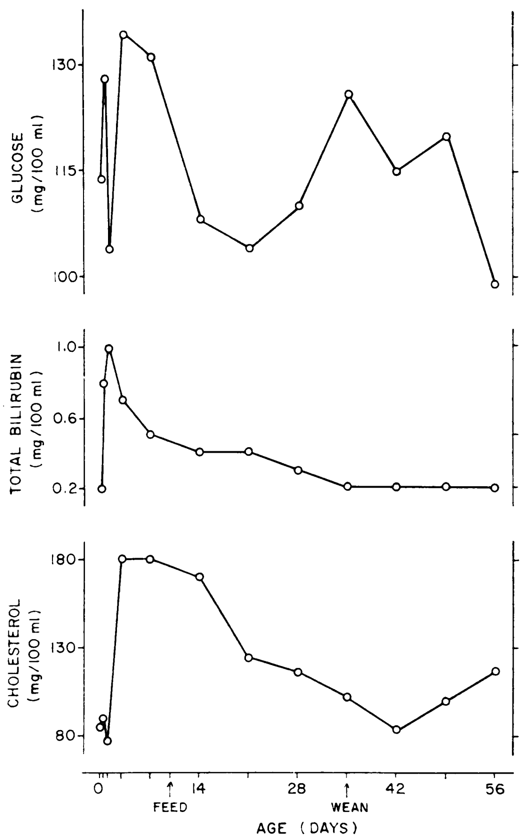

Fig. 6. Mean concentrations of cholesterol, total bilirubin and glucose in serum from crossbred swine from birth through eight weeks of age.

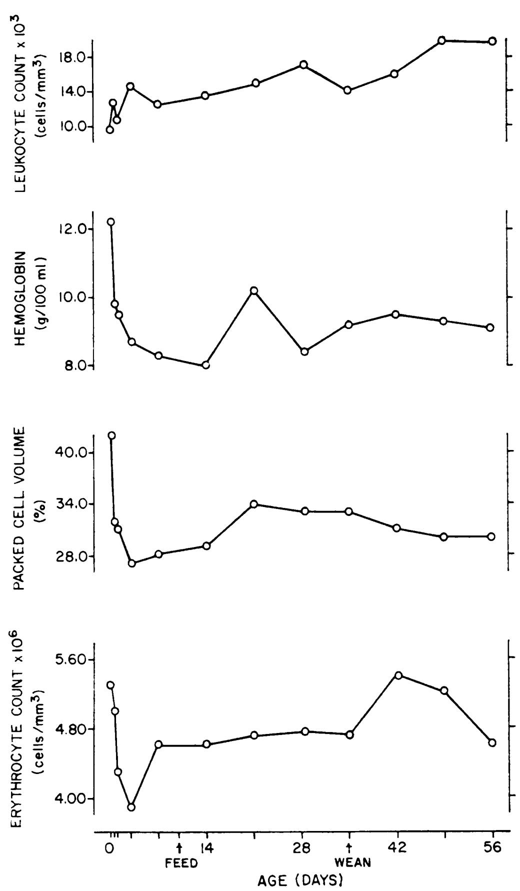

Fig. 7. Mean erythrocyte counts, packed cell volumes, hemoglobin concentrations and leukocyte counts in blood from crossbred swine from birth through eight weeks of age.

DISCUSSION

The initial increase in serum TP concentration, as a result of the increase in TG concentration with no change in Alb concentration, was consistent with previous reports on pigs. From birth to 24 hours of age, the increase in TG concentration was 94, 154, 156, 158 and 285% for our study and the studies of Ramarez *et al* (15), Miller *et al* (10), Tumbleson *et al* (21) and McCance and Widdowson (8), respectively. The decreases in RBC, PCV and Hgb during the first three days of neonatal life were similar to those reported by Miller *et al* (11); however, mean PCV and Hgb were greater than reported (3) for fetuses at 110 to 113 days of gestation. Mean RBC, PCV and Hgb at two weeks of age were similar to previous reports (4, 11). There was a 25% decrease in mean PCV from birth to 24 hours of age which was comparable to decreases of 15% (15), 21% (14) and 35% (8) observed in other laboratories. It would appear that changes were associated with neonatal development rather than a pathological condition.

During the first 24 hours of life, the decreases in mean serum concentrations of Na, K and Cl with concomitant increases in TP, γ-Glob, BUN and TB concentrations and OT, LDH and AP activities while mean concentrations of serum Alb, $\underline{\alpha}$-Glob, β-Glob, Ca, IP and Chol did not change, suggests the presence of specific mechanisms for the absorption of different molecules. The reason for rapid intestinal absorption of large molecules during only the first 24

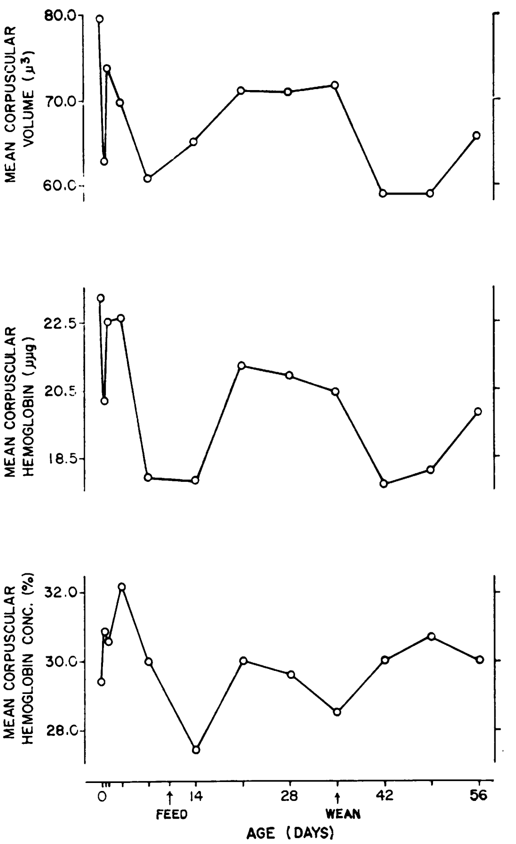

Fig. 8. Mean corpuscular volumes, mean corpuscular hemoglobins and mean corpuscular hemoglobin concentrations in blood from crossbred swine from birth through eight weeks of age.

to 36 hours of neonatal life (1, 6, 7, 9, 13, 17, 18) is still undefined. Contributive factors which have been suggested include changes in the intestinal absorptive cells (13, 17, 18), location of the Golgi apparatus (18), development of metabolic systems in the absorptive cells (21, 23) and loss of available surface membrane for invagination (6). Cessation of absorption of large molecules has been shown to be not dependent upon absorption of large molecules *per se* (7), but occurred after ingestion of 300 mEq of glucose (6) which was independent of concentration or volume of glucose solution. The high activity of alkaline phosphatase in serum of the newborn, with an increase to an even higher level, was consistent with previous reports for swine serum (21, 23) and intestine (12, 16). Perhaps the increase in enzyme activity results in, or is a result of, a change in absorptive capacity.

The delay in serum Chol concentration increase until after 24 hours of neonatal life may have been the result of a lack of development of a particular intestinal absorption mechanism, the immediate uptake of Chol by other tissues or a need for the hepatic tissue to develop metabolic systems to metabolize Chol. The increase in serum K concentration, from five to six weeks of age, was similar to that reported by Ullrey *et al* (22). The changes in serum Na, K and Cl concentrations, subsequent to weaning, were probably due to changes in dietary intake as well as stress involved with removal from the dam.

Due to the changes in γ-Glob, AP and Chol and the variability in Na, BUN, Glu, MCV, MCH and MCHC, from one through eight weeks of age, it is recommended that multivariate analyses, assessing only those parameters relevant to the experimental design, be conducted to adequately evaluate particular disease or nutritional conditions. The relative changes in blood constituent concentrations reported in this paper may be used to determine the numbers of experimental units necessary for demonstrating statistically significant differences, at the desired level of probability, for a particular experiment. The mean values, relative changes, variability and ranges for the serum biochemical and hematological parameters evaluated in this study may be most valuable for investigators designing research protocols using swine as biomedical research subjects.

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