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Time course of hemolysis in respiratory alkalosis. ✡

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Introduction

In 1958 Balke and associates observed hemolysis in blood samples from persons who were hyperventilating (2). In 1961 Bindslev and associates reported the results of a series of in vitro experiments showing the relationship between lowered PCO_2, increase in pH and hemolysis(3). In 1963 Bindslev concluded from in vivo experiments that the increase of pH in respiratory and metabolic alkalosis is the major cause of hemolysis (4).

In the above experiments the progress of hemolysis at different hours after the establishment of hyperventilation has not been reported. As hemolysis is one of the complications encountered during extracorporeal circulation the time required for the start and progress of lysis is important.

Methods

Twenty mongrel dogs of either sex, weighing 6-9 Kg, were used for this study. The animals were fasted for 24 hours with free access to water. They were anesthetized with sodium pentobarbital, 30 mg/Kg, i. v. After tracheotomy the animals were conducted to an intermittent positive pressure respirator with a frequency of 48 strokes/minute. The stroke volume was 25-30 ml/Kg, and the time of hyperventilation was 6 hours. The experiments were carried out in two groups:

Group 1 (control group). 14 dogs were hyperventilated with a mixture of air and oxygen containing 6% CO_2. CO_2 concentratoin was monitored in the inspiratory air with the aid of a calibrated Godart capnograph in a semiclosed circuit

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Group 2 (experimental group) 14 dogs were hyperventilated with a mixture of air and oxygen.

Blood samples were drawn from the femoral artery, before and 1, 2, .. and 6 hours after establishment of hyperventilation.

Serum hemoglobin concentration was measured spectrophotometrically using modifiedb enzidin method of Crosby et al except that a 9% NaCl was used instead of distilled water to prevent precipitation of plasma proteins (7) Blood pH was measured anaerobically with a glass electrode at 37° C.

Results

The changes of pH of 4 dogs is given in table 1. In three dogs it reached a value of 7.80_7.95 in the first hour and 7.92_7.90 at 4th hour. In two of these animal pH remained stable, but in the third dog it decrcased to 7.53 at sixth hour. In the fourth dog pH increased from 7.49 to 7.80 in the first hour and decreased thereafter reaching a value of 7.68 at the sixth hour. Hemolysis was marked in the first three dogs but was slight in the fourth dog.

Dog No	b.a	1	2	3	4	5	6
1	7.45	7.85	7.88	7.89	7.90	7.92	7.92
2	7.45	7 80	7.86	7.95	7.92	7.94	7.94
3	7.45	7.95	7.95	7.95	7.92	7 82	7.53
4	7.49	7.80	7 72	7.74	7.74	7.70	7.68

Table 1- pH changes in 4 dogs before and during 6 hours of hyperventilation with air and oxygen.

Plasma hemoglobin concentration increased gradually from prealkalotic value (6.0 mg ' 100 ml plasma) to 639 mg/100 ml at sixth hour. As can be seen in Table 2, hemolysis developed rapidly in one dog (No-10), was not observed in another (No 4) and was delayed until the 4th hour in two dogs (No 7 & 8).

In the control group blood pH and plasma hemoglobin concentration remained at the prehyperventilation value.

Dog No	b.a	1h	2h	3h	4h	5h	6h
1	0	3	12	33	160	273	—
2	4	14	58	152	248	328	—
3	3	7	60	400	625	940	1050
4	12	12	12	11	11	11	11
5	4	6	12	42	42	142	192
6	2	7	210	500	850	1960	1060
7	10	10	10	10	90	198	270
8	7	6	15	15	36	78	124
9	6	14	21	25	89	128	140
10	12	480	900	1390	1780	2140	2240
$\overline{X}$	6.0	55.9	131.0	257.9	392 8	622.8	639.6

Table 2_ Plasma hemoglobin concentration (mg/100 ml plasma) in alkalotic dogs bofore and at different hours, after the establishment of alkalosis

Discussion

The occurrence of hemolysis in the experimental group and its absence in the control group shows that hemolysis is not due to the mechanical action of hyperventilation and is related to the increase of blood pH. Our results are in accord with the observations of Balke and associates (2) in man and Bindslev in dog (4) and in addition shows that inspite of the fact that alkalosis is produced in the first minutes after establishment of hyperventilation, hemolysis needs at least one hour (and usually two hours) to be started and is progressive.

The literature survey of alkalosis shows that glucose metabolism is increased in different tissues' including perfused rat heart (8,22-24) cat brain slices (9), intact muscle, kidney and liver slices, and epididymal fat pad from the rat (14), red blood cells (6,19_21), white blood cells (16) frog muscle (18) cell free extract of rat muscle (25,26), heart lung preparation (1), isolated limb preparations (10), and in human subjects (5,11,17) and intact dog (12,15) we might speculate that observed lysis may be due to an inadequacy of energetic substances in red cells in alkalotic state.

Summary

Blood pH and plasma hemoglobin concentration were measured in dog undergoing hyperventilation with or without 6% CO 2. Blood pH rose in the first minutes in the alkalotic group and hemolysis appeared mostly during second hour after alkalosis was established. It increased gradually during the following hours of hyperventilation. No hemoly

sis was observed in the group undergoing hyperventilation with 6% CO_2. It is concluded thal hemolysis is unrelated to mechanical action of hyperventilatroin and in due to alkalosis. the possible cause of hemo lysis and related litrature is discussed.

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INTRAUTERINE INFECTION. *

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Pothways of fetal and early neonatal infection.

Review of the «AMNIOTIC INFECTION SYNDROME» in 150 autopsies of new and stillborn babies.

The risk of intrauterine infection occuring in the newborn infant was mentioned by KUSSNER (1877) and GEYL (1880) at the end of the last century (14).

SLEMONS (1915) reported the occurrence of bacteria in the sub-amniotic space near the attachment of the umbilical cord during prolo-nged labour (21).

DOUGLAS and STANDER (1943) have shown that mortality and morbidity of the newborn are directly related to the length of labour and this is on account of intrapartum infection (13-14).

In recent years obstetricians, paediatricians and Pathologists have recognized the problem of intrauterine infection and, among them WILIAM and KURT BENIRSCHKE have written much about this subject.

According to MULLER (1956) the foetus may be infected by the following routes:

1- Hematogenous spread via maternal blood.

2- Ascending amniotic infection in which vaginal bacteria reach the uterine cavity directly through the cervical canal.

3- Transdecidual spread, either on account of an exacerbation of an existing endometritis or because of an ascending infection occuring between the uterine wall and the memoranes.

4- Via the fallopian tubes.

From a practical view point the hematogenous spread which causes antenatal infection and the ascending amniotic infection which occurs during labour are the more important causes of intrauterine infection and early neonatal infective death (2-4-6-7-14-18-20).

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