COMPARISON OF ARTERIAL BLOOD GASES AND ACID-BASE BALANCE IN YOUNG AND AGED BEAGLE DOGS, WITH REGARD TO POSTPRANDIAL ALKALINE TIDE

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(Received August 9, 1999 ; Accepted May 31, 2000)

ABSTRACT — Arterial blood pH and blood gas tensions were measured before and after feeding in seven young (1-year-old) and 6 aged (10 to 12 year-old) conscious beagle dogs with a portable “i-STAT” blood gas analyzer to obtain the fundamental physiological data on the laboratory dogs.

Before feeding, arterial blood pH, HCO_3^- and base excess (BE) were 7.395 ± 0.022 , 20 ± 1.0 mmol/L and -4 ± 1.1 mmol/L in young dogs, and 7.408 ± 0.031 , 23 ± 1.5 mmol/L and -2 ± 1.8 mmol/L in aged ones. After feeding, the above values changed to 7.449 ± 0.021 , 25 ± 1.1 and 1 ± 1.3 in young dogs, and 7.456 ± 0.022 , 28 ± 1.7 and 4 ± 1.9 in aged ones.

After feeding, each of the values for pH, HCO_3^- and BE was increased in comparison with those before feeding in both young and aged dogs, and alkalization of blood, the alkaline tide, was observed in all healthy dogs. The values for PCO_2 also rose from 33.6 ± 1.91 to 36.7 ± 1.95 (mmHg) in young dogs, and 36.4 ± 2.97 to 40.3 ± 3.06 (mmHg) in aged ones, and in 3 of 6 aged dogs slight hypoxia ($\text{PO}_2 < 80$ mmHg) was noted. These changes in PCO_2 and PO_2 were considered to be due to compensatory hypoventilation for the alkalization of blood. To confirm the cause of the alkaline tide phenomenon, we investigated the association of the alkaline tide with gastric acid secretion by cimetidine, an H_2 -blocker; and found that it was effective to inhibit postprandial alkaline tide in dogs.

After feeding, ionized Ca concentration (iCa) was decreased inversely with an arterial blood pH increase in both young and aged dogs. The mean values of iCa in aged dogs after feeding was lower than those in young dogs.

In the present experiments, it was demonstrated that in beagle dogs the acid-base balance and blood gas values were largely affected by feeding, especially in aged dogs. In case blood gases would be evaluated in toxicity studies, we should consider the influence of feeding and aging.

KEY WORDS: Arterial blood gases, Acid-base balance, Alkaline tide, Beagle dog, Ionized calcium

INTRODUCTION

Blood gas analysis is a useful and powerful method for knowing the acid-base status in the body. In recent years, the importance of functional examinations has been stressed in pre-clinical toxicity studies for candidates of pharmaceuticals. Blood gas analysis in acute and chronic toxicity studies can give useful information to doctors in clinical trials, when they evaluate the toxicological data for new chemical compounds. However, blood gas analysis has not yet become general as a part

of toxicity studies.

One of the reasons that the blood gas analysis has not been performed generally in toxicity studies may be that the treatment and maintenance for a conventional gas analyzer is complicated and difficult. Recently a portable blood gas analyzer which uses a disposable cartridge which can measure blood gases rapidly and precisely has come into wide use in emergency medical and ICU, etc. (Nakamura *et al.*, 1997). We thought that it would be possible to conduct blood gas analysis quickly and easily in animal rooms and laboratories

with the analyzer.

As to blood gas analysis in dogs, Langbroek *et al.* (1990) investigated the influence of feeding on blood gases and acid-base balance in mongrel dogs. The change of arterial blood gases which accompanies aging has been reported in humans and in various kinds of dogs (Gunnarsson *et al.*, 1996; Cerveri *et al.*, 1995; King *et al.*, 1992; Aguilera-Tejero *et al.*, 1997). However, on the basal values of blood gases in beagle dogs, there are insufficient data despite the species used frequently in the toxicity studies.

The purpose of the present experiment was to obtain the fundamental values of blood gases with a portable analyzer in beagle dogs. The influence of aging and feeding on blood gases was also investigated.

MATERIALS AND METHODS

Animals

Seven young (1-year-old) and six aged (10 to 12 year-old) beagle dogs were used. The animals were confirmed to be healthy by blood examination and to complete feeding within 30 min after feeding. The animal room was maintained at $24 \pm 2^\circ\text{C}$ and $55 \pm 15\%$ humidity with a 12 hr lighting cycle (light on: 6:30~18:30). Each animal was individually housed in a stainless cage and given 300 g/day of a diet (DS: Oriental Yeast Co., Ltd.) once a day, and tap water freely.

Blood gas analyzer

The blood gas was analyzed with a portable blood gas analyzer 'i-STAT' (i-STAT Corporation, USA) using a disposable cartridge (EG7+) which contained a micro-fabricated bio-sensor array.

Arterial partial pressures of oxygen (PO_2 : Clark electrode), carbon dioxide (PCO_2 : Severinghaus electrode), pH (ion-selective electrode), hematocrit (Hct: conductometrical), sodium (Na: ion-selective electrode), potassium (K: ion-selective electrode), and ionized calcium (iCa : ion-selective electrode) were measured directly on whole blood at 37°C , and calculated values for bicarbonate (HCO_3^-), total CO_2 (TCO_2), base excess (BE), oxygen saturation (sO_2), and hemoglobin (Hgb) were recorded automatically.

The unit for PO_2 and PCO_2 was presented by mmHg printed out from the machine in this paper, instead of kPa as defined (recommended) by the International System of Units: (1 mmHg = 0.133 kPa).

Procedure

Every dog was restrained in the lateral recumbent

position, and 0.1 mL of arterial blood was withdrawn with a non-heparinized syringe from the femoral artery before (fasting) and 2 hr after feeding. Blood samples were immediately analyzed with an 'i-STAT'.

The blood gas was analyzed for each of three dogs out of seven young and six aged dogs on the following day to check the data reliability. In addition, two young and two aged dogs were injected intravenously with 5 mg/kg of cimetidine, an H_2 blocker, to inhibit gastric acid secretion at 15 min before feeding, and were observed for changes in blood gas values before and after feeding.

Statistical analysis

Data are presented as mean and standard deviation. Statistical significance was determined by the use of Student's paired (influence of feeding) or unpaired (influence of aging) *t*-test (Gad and Weil, 1982). Linear correlation and linear regression analyses were used to evaluate the relation ship between the artery blood pH and ionized calcium concentration.

RESULTS

Table 1 shows the mean values of measurement parameters by portable blood gas analyzer 'i-STAT' before and after feeding in healthy beagle dogs. Fig. 1 shows individual data of base excess and Fig. 2 shows the values of the measurement parameters of a typical aged dog on a radial pattern graph.

Before feeding, there were no differences in arterial PO_2 , PCO_2 , and pH values between young and aged dogs. However, HCO_3^- and BE values in aged dogs were significantly higher than those in young dogs.

After feeding, PCO_2 , pH, and BE values rose significantly in all animals when compared with those before feeding. PO_2 values after feeding for aged dogs were significantly lower than those before feeding and those for young dogs. Slight hypoxia in 3 of 6 aged dogs were also noted ($\text{PO}_2 < 80$ mmHg).

Table 2 shows changes in artery blood gas values and acid-base balance after feeding with and without iv cimetidine treatment. In four dogs treated with cimetidine before feeding, a rise in BE and HCO_3^- after feeding was reduced in all dogs, and PCO_2 value did not increase in three of four dogs.

In oxygen saturation there was no difference for all comparisons. Hct and Hgb values for aged dogs before feeding were significantly lower than those in young dogs, and those values after feeding in both groups of dogs were higher than those before feeding.

Table 1. Blood gas values obtained from 7 young and 6 aged healthy beagle dogs before and 2 hr after feeding.

Parameter (Unit)	Young dogs (1 yr, n=7)		Aged dogs (10–12 yr, n=6)	
	Before feeding	2 hr after feeding	Before feeding	2 hr after feeding
PO ₂ mmHg	95 ± 9.0	88 ± 4.4 ##	94 ± 7.6 **	80 ± 8.0 ¹⁾ ** (p=0.0356) ## (p=0.0369)
PCO ₂ mmHg	33.6 ± 1.91 *	36.7 ± 1.95 * (p=0.0483) ##	36.4 ± 2.97 **	40.3 ± 3.06 ** (p=0.0020) ## (p=0.0269)
pH	7.395 ± 0.0219 *	7.449 ± 0.0213 * (p=0.0057)	7.408 ± 0.0311 **	7.456 ± 0.0223 ** (p=0.0132)
SO ₂ %	97 ± 0.8	97 ± 0.7	97 ± 0.8	96 ± 0.8
BE mmol/L	–4 ± 1.1 *	1 ± 1.3 * (p=0.0002) ##	–2 ± 1.8 **	4 ± 1.9 ** (p=0.0024) ## (p=0.0067)
HCO ₃ mmol/L	20 ± 1.0 *	25 ± 1.1 * (p=0.0003) ##	23 ± 1.5 **	28 ± 1.7 ** (p=0.0013) ## (p=0.0039)
TCO ₂ mmol/L	21 ± 1.0 *	27 ± 1.3 * (p=0.0002) ##	24 ± 1.5 **	30 ± 1.6 ** (p=0.0009) ## (p=0.0040)
Hct %PCV	46 ± 2.9 *	48 ± 3.7 * (p=0.0134)	41 ± 3.9 **	44 ± 4.7 ** (p=0.0372)
Hgb g/dL	15.7 ± 1.11 *	16.3 ± 1.25 * (p=0.0300)	14.2 ± 1.17 #	15.0 ± 1.67 # (p=0.0326)
Na mmol/L	144 ± 1.1	145 ± 1.3	144 ± 1.9	144 ± 1.5
K mmol/L	3.9 ± 0.24	3.6 ± 0.24	3.9 ± 0.25	3.9 ± 0.42
iCa mmol/L	1.39 ± 0.068	1.35 ± 0.046 ##	1.33 ± 0.059	1.28 ± 0.025 ## (p=0.0093)

Values: Mean ± standard deviation (S.D.). n = number of dogs. 1 mmHg = 0.133 kPa

¹⁾: 3 of 6 aged dogs fell into slight hypoxia (PO₂<80 mmHg).

*: P<0.05, the paired Student's *t*-test, Before feeding vs. 2hr after feeding (young dogs).

**: P<0.05, the paired Student's *t*-test, Before feeding vs. 2hr after feeding (aged dogs).

#: P<0.05, the independent Student's *t*-test, Young dogs vs. Aged dogs (before feeding).

##: P<0.05, the independent Student's *t*-test, Young dogs vs. Aged dogs (2 hr after feeding).

Fig. 1. Individual data of base excess in young and aged dogs before and 2 hr after feeding.
 : Top 7 young beagle dogs
 : Bottom 6 aged beagle dogs
 In 3 dogs each of young and aged dogs, blood gases were measured for two consecutive days.

Fig. 2. Values of the measurement parameters of a typical aged dog before and 2 hr after feeding.
 Fine line: Normal range, upper and lower limits.
 Thick line: Measured values.

Fig. 3. Correlations between the artery blood pH and ionized calcium concentration (log iCa).
 These data plotted are all values measured before and after feeding in 7 young dogs and 6 aged dogs.

Arterial blood gas in young and aged beagle dogs.

The iCa concentration was inversely related to arterial blood pH (Fig. 3). In Fig. 3 the linearity for pooled data between log iCa and pH is expressed by the equation: $\log iCa = 2.798 - 0.36 \cdot pH$ ($p = 0.0005$). The coefficient of correlation was good (0.631). The mean iCa values in aged dogs after feeding were lower than those in young dogs. There was no difference in the concentrations of Na and K in all comparisons.

DISCUSSION

The PCO₂ value (Mean $\pm$ S.D.) in young dogs before feeding was 33.6 ± 1.91 mmHg. This value was below the range of the human normal values (35-45 mmHg; Shapiro *et al.*, 1994).

The arterial pH value was 7.395, within the human normal range (7.35-7.45), and BE was -4 , near the lower limit of the human normal value (± 5). On the other hand, each value in aged dogs corresponded also to the human normal range (PCO₂, 36.4 mmHg, pH; 7.408, BE; -2). The lower value of PCO₂ in young dogs in fasting was indicated to be caused by respiratory

compensation toward acid in acid-base balance of BE values. It is supposed that this incidence is not due to acute hyperventilation, because the mean PO₂ was within the normal range (95 mmHg). There were no significant differences in PO₂ on fasting between young and aged dogs. It has been reported that PO₂ tends to decrease with aging in humans (Gunnarsson *et al.*, 1996; Cerveri *et al.*, 1995). However, PO₂ has not necessarily decreased in various kinds of aged dogs (King *et al.*, 1992; Aguilera-Tejero *et al.*, 1997). Furthermore, there was no report to describe the differences in acid-base balance between young and aged, such as our results. It is supposed that this discrepancy is due to the differences in 1) various kinds of dogs used vs. the same colony of beagle dogs in our experiment, and 2) housing in a different environment vs. housing under room conditions.

After feeding, alkaline tide, alkalization of blood, was observed in every dog. The values of pH, HCO₃⁻ and BE were elevated in comparison with those before feeding. Slight hypercapnia in young and aged dogs and slight hypoxia (PO₂ < 80 mmHg) in 3 of 6 aged

Table 2. Changes in artery blood gas values and acid-base balance after feeding, with and without cimetidine in young and aged healthy dogs.

Animal No.		No administration		Cimetidine 5 mg/kg, iv	
		Before feeding	After feeding	Before feeding	After feeding
Young 1	PO ₂	88	90 (Δ 2)	99	100 (Δ 1)
	PCO ₂	31.4	35.7 (Δ 4.3)	35.0	31.4 (▲ − 3.6)
	pH	7.434	7.447 (Δ 0.013)	7.391	7.458 (Δ 0.067)
	BE	− 3	1 (Δ 4)	− 4	− 2 (Δ 2)
	HCO ₃	21	25 (Δ 4)	21	22 (Δ 1)
Young 2	PO ₂	110	90 (▲ − 20)	99	99 (Δ 0)
	PCO ₂	31.6	37.5 (Δ 5.9)	33.4	31.8 (▲ − 1.6)
	pH	7.414	7.480 (Δ 0.066)	7.398	7.469 (Δ 0.071)
	BE	− 4	4 (Δ 8)	− 4	− 1 (Δ 3)
	HCO ₃	20	28 (Δ 8)	21	23 (Δ 2)
Aged 1	PO ₂	94	91 (▲ − 3)	97	90 (▲ − 7)
	PCO ₂	37.9	41.2 (Δ 3.3)	37.5	38.7 (Δ 1.2)
	pH	7.363	7.479 (Δ 0.116)	7.406	7.428 (Δ 0.022)
	BE	− 4	7 (Δ 11)	− 1	1 (Δ 2)
	HCO ₃	22	31 (Δ 9)	24	26 (Δ 2)
Aged 2	PO ₂	94	89 (▲ − 5)	98	92 (▲ − 6)
	PCO ₂	36.4	39.2 (Δ 2.8)	39.7	37.5 (▲ − 2.2)
	pH	7.414	7.447 (Δ 0.033)	7.397	7.434 (Δ 0.037)
	BE	− 1	3 (Δ 4)	0	1 (Δ 1)
	HCO ₃	23	27 (Δ 4)	24	25 (Δ 1)

Δ : increase. $\blacktriangle$: decrease.

dogs were noted after feeding. These changes in PCO₂ and PO₂ were considered to be due to compensatory hypoventilation for the alkalinization of blood. These results are very interesting, as there were no reports concerning the investigation of postprandial alkaline tide in aged animals or geriatric men.

To confirm an association between alkaline tide and gastric acid secretion, 5 mg/kg cimetidine, an H₂ blocker, was given intravenously at 15 min before feeding to two dogs in the young and aged groups. An inhibition of postprandial alkaline tide was demonstrated in every dog.

Johnson *et al.* (1995) reported that it was not possible to detect a postprandial alkaline tide phenomenon in physiological respiratory functions. Niv *et al.* (1993), however, have shown that the alkaline tide measurement was utilized as a new and convenient method for gastric acid output rate estimation. Our results supported the report by Niv *et al.* (1995) on the point of BE rising and an increase of PCO₂ for respiratory compensation after feeding.

From the above results, it seems reasonable to suppose that a slight base deficit in fasting for the young laboratory beagle dogs still had a normal value on acid base balance. The alkaline tide phenomenon may be used for the evaluation of a function of the digestive system in toxicological studies.

The iCa concentration in arterial blood was inversely related to arterial blood pH in both young and aged dogs. The mean iCa value in aged dogs after feeding was lower than that in young dogs. Gaiter *et al.* (1997) have reported the same correlation between pH and iCa, and that the same change in pH has caused greater change in iCa after the acute change in metabolic-acid balance than after respiratory imbalance. It has also been reported that a patient with hypocalcemia was led to severe metabolic alkalosis which resulted in tetany. (Kaye *et al.*, 1997). Therefore, it is speculated that aged dogs have a high risk of hypocalcemia. Additionally, the iCa concentration and pH value in arterial blood might be useful information for considering the cause of tetany in the toxicity studies of new entities.

In conclusion, the present experiment has shown that it was possible to conduct blood gas analysis quickly and easily in beagle dogs with a portable analyzer. It was also demonstrated that in beagle dogs the acid-base balance and blood gas values were affected by feeding, especially in aged dogs. In case blood gases would be evaluated in acute and chronic toxicity studies, we should consider the influence of feeding and

aging.

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