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REFERENCE DATA ON THE ANATOMY AND SERUM BIOCHEMISTRY OF THE SILVER FOX

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SUMMARY

Clinically healthy silver foxes obtained from a closed colony were investigated for the purpose of establishing base-line data for this species. The anthropometry (body weight; body length; length and width of the head; width, depth, and circumference of the chest; length of the tail), anatomical measurements (weight; longitudinal and transverse length; thickness of the main organs) and serum biochemical assays (AST, ALT, ALP, LDH, CK, lipase, GGT, T-Cho, β-Lipo, TG, Phos-Lip, Tp, T-Bil, UA, BUN, Crea, Glu, Ca, IP, Mg, Fe, Na, K, Cl, LDH and CK isoenzymes) were carried out. The data were presented as mean values with standard deviations, and compared with those of the dog.

The coefficient of variation (CV) for each of the anthropometric parameters was low, except for that of female body weight for which the CV was 17%. The body size of the male was larger than the female, and the weights of the main organs, corresponding to body size, were greater than the female. The results were equivalent to those for a Beagle dog aged between 3 and 5 months. Significant differences between the sexes were detected in the following parameters: concentrations of BUN, β-Lipo and T-Bil ($p<0.01$); concentration of Mg and Glu ($p<0.05$); activity of LDH and lipase ($p<0.05$).

The biochemical data ware uniform with some exceptions. These were AST (142 IU/l) and ALP (122 IU/l) in a 5-year-old male fox, Glu (over 200 mg/dl) in four 2-year-old female foxes, CK (629 IU/l) in a 2-year-old female fox, and finally CK (366 IU/l) and lipase (428 IU/l) in an 8-year-old female fox, all of which were elevated. These data were similar to the reference values for the dog previously reported.

The reference values presented in this report for the silver fox will be valuable as a guide for clinical diagnosis and research.

Key words: silver fox, anthropometry, anatomy, biochemistry

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INTRODUCTION

Recently the animal protection movement has gained momentum in America and Europe and is spreading throughout the world. This movement has important implications for biological, medical and veterinary scientific studies using animals. As a result, as an alternative to the use of animals for such studies, the utilization of cell cultures and invertebrates has been developed and become common. However, there are still fields in which the use of animals is necessary. In these fields, it is preferable to use animals other than companion animals to which man has strong sentimental ties. There are still many problems in replacing companion animals as experimental animals with domestic ones such as cattle, pigs or sheep. The silver fox, a fur-bearing animal similar to the dog in many respects, may be a more appropriate experimental animal than others, since studies of its behavior, nutrition, genetics and reproduction have been carried out over a long period, and it can be raised easily under laboratory conditions [2, 6, 9, 13, 14, 17, 18]. However, since there is limited information available regarding the anthropometry, anatomy and biochemistry of the silver fox [15, 16], it is necessary to establish reference values. In this report, 45 clinically healthy silver foxes were investigated for the purpose of establishing base-line data for this species. These data will be useful for veterinary practitioners and scientific investigators in interpreting biochemical data determined by similar laboratory methods.

MATERIALS AND METHODS

Silver foxes were obtained from a fox farm in Hokkaido, Japan. Twenty-five clinically healthy females (2–8 years, mean 4.9 years) and 20 males (2–7 years, mean 5.4 years) were examined. The animals were housed in wire pens elevated 1m off the ground in order to prevent infection with Echinococcus and other parasites, and were raised in an isolated area. In order to preserve the quality of the fur, the animals were kept in a closed colony.

The anthropometric parameters measured were body weight ; length of the head, body and tail ; width of the head ; and depth and circumference of the chest, using the method described by Fukui et al for dogs[5]. That is, body length : from the manubrium sterni to the tuber ischium ; body height : from the spinous process of the first thoracic vertebra to the tip of the foot-pad ; length of the head : from the tip of the nose to the tuber of os occipitale ; length of the tail : from the first to the last coccygeal verterbra ; depth of the chest : from the spinous process of the first thoracic vertebra to the xiphoid process ; width of the head : between the two zygomatic processes ; circumference of the chest : circumference at the same site as for width of the chest measurement.

The main organs including the brain, lungs, heart, liver, spleen, pancreas, kid-

neys, prostate, testes and the uterus were weighed after autopsy, and the longitudinal and transverse length, and thickness were measured. These parameters were measured after the attachments were removed. The kidney was weighed with the capsule intact.

Blood samples for biochemical analysis were collected from the cephalic vein with the animals conscious. It was easier to draw 5ml blood samples while manually restraining the animals. The samples were collected in vacutainer tubes for serum and then the serum separated. The sera in ice were sent to the biochemical laboratories. A serum analyzer (Hitachi 736) was used to measure the following parameters: blood urea nitrogen (BUN), creatinine (Crea.), uric acid (UA), total cholesterol (T-Cho), total bilirubin (T-Bil), triglyceride (TG), phospholipid (Phos-Lip), β-lipoprotein (β-Lipo), alkaline phosphatase (ALP), lactate dehydrogenase (LDH), γ-glutamyl transferase (GGT), creatine kinase (CK), aspartate aminotransferase (AST), alanine aminotransferase (ALT), inorganic phosphorus (IP), calcium (Ca), magnesium (Mg), and iron (Fe). Glucose (Glu) was determined with Olympus ACA-800, lipase with Hitachi-7150, and sodium (Na), potassium (K) and chloride (Cl) with Fuji Drichem-800 (ion-specific electrodes). Serum protein concentration was evaluated using a hand refractometer with Atago SPR-N. CK and LDH isozyme analysis were performed using cellulose acetate plates and were read with a densitometer (Helena laboratories).

The mean, standard deviation (SD) and the coefficient of variation (CV) for the anthropometric and anatomical data were calculated. The statistical differences were determined by Student's t-test.

Results

The anthropometric data for each age group and the sexes are presented in Table 1. The body weight of the male and female were 8.7 ± 1.0 kg and 6.1 ± 1.1 kg, and the body length of the male and female were 55.8 ± 2.8 cm and 51.0 ± 3.5 cm respectively. CVs for each of the parameters were low, except that of 17% for the body weight of the female. The measurements for each parameter were larger in the male than in the female.

Anatomical measurements of the organs are shown in Table 2. The organs from 12 male and 10 female animals were weighed and measured and the mean, SD and CV were calculated without taking the age difference into account. The CV of spleen thickness and weight, measurements of the pancreas in both sexes, liver thickness, weight of the left kidney, measurements of the uterus in the female, weight of the lungs, and transverse length and weight of the prostate in the male are all over 20%. CV of other parameters were low.

The biochemical values are listed in the Table 3, 4 and 5. Two animals were excluded from the statistical test, because one had markedly higher activity of AST

TABLE 1. Measurements of anthropometric parameters from silver foxes aged from 2 to 8 years.

S		N	BW (kg)	BL (cm)	LH (cm)	WH (cm)	DC (cm)	WC (cm)	CC (cm)	LT (cm)
M	Mean	20	8.7	55.8	17.5	7.9	12.8	11.2	45.8	47.0
	SD		1.0	2.8	1.3	0.6	1.0	1.3	3.0	4.3
	CV(%)		11.5	5.0	5.0	7.5	8.0	11.0	6.6	9.0
F	Mean	25	6.1	51.0	16.9	7.1	11.0	9.2	36.9	41.4
	SD		1.1	3.5	0.7	0.4	1.0	1.1	3.5	4.2
	CV(%)		17.0	6.8	3.0	5.0	9.0	12.0	9.5	9.9

SD : standard deviation. CV : the coefficient of variation.
N : number of the animal. BW : body weight. BL : body length.
LH : length of the head. WH : width of the head. DC : depth of the chest.
WC : width of the chest. CC : circumference of the chest.
LT : length of the tail. S : sex. M : male. F : female.

(142 IU/l) and ALP (122 IU/l), and the other had higher activity of CK (366 IU/l) and lipase (428 IU/l) compared with other animals examined.

As shown in Table 3, the concentration of BUN (22.1±2.9 mg/dl in the 7-year-old male group and 18.9±3.4 mg/dl in all males) and Glu (171.0±35.6 mg/dl in the 7-year-old male group and 130±27.8 mg/dl in all males) were significantly higher than those for the other age groups. There were no statistical differences for each age group in the other parameters. Compared with the female, the concentration of BUN (18.9±3.4 mg/dl in the male and 14.9±2.4 mg/dl in the female) and T-Bil (0.9±0.3 mg/dl in the male and 0.7±0.2 mg/dl in the female) were significantly high ($p<0.01$), while the concentration of β-Lipo (27.0±8.3 mg/dl in the male and 41.4±18.4 mg/dl in the female) was significantly low ($p<0.01$) in the male. The concentration of Glu in the male (130.4±27.8 mg/dl) was also significantly lower ($p<0.05$) than in the female (157.4±50.1 mg/dl). No significant differences between the sexes were observed in other parameters.

From Table 4, the concentration of Fe (208.7±16.5 μg/dl) in the 5-year-old male group differs significantly from other male groups, and the concentration of Fe in all males was 175.1±38.2 μg/dl. Significant difference between the sexes ($p<0.05$) was found in the concentration of Mg, the values of which for the male and female were 1.7±0.3 mg/dl and 1.9±0.3 mg/dl, respectively. There were no significant differences within each age group or between the sexes in other parameters.

TABLE 2. Measurements of anatomic parameters from silver foxes aged from 2 to 8 years.

ORGAN	SEX	ORGAN WEIGHT(g)	LONGITUDINAL LENGTH (cm)	TRANSVERSE LENGTH (cm)	THICKNESS (cm)
Brain		48.9± 2.0	—	—	—
Heart	M	57.6± 5.3	58.8± 4.8	54.0± 2.6	34.7± 1.5
	F	50.4± 7.9	53.3± 3.2	51.0± 5.0	36.5± 5.4
Lungs	M	76.4±23.5#	—	—	—
	F	67.5±11.2	—	—	—
Liver	M	189.0±20.6	156.4±11.6	88.5± 8.3	22.3± 2.8
	F	165.2±17.0	142.1±10.4	79.3± 8.7	22.8± 5.2#
L. Kidney	M	20.9± 2.4	48.8± 2.2	30.7± 2.3	19.9± 1.9
	F	16.8± 3.4#	43.9± 5.7	33.1± 6.0	21.4± 4.0
R. Kidney	M	21.2± 2.2	49.9± 2.7	30.4± 2.3	20.4± 1.7
	F	17.0± 2.6	45.2± 2.7	30.0± 2.3	19.8± 1.7
Spleen	M	13.5± 6.4#	127.4±13.9	33.8± 4.3	6.4± 1.3#
	F	12.0± 5.4#	116.3± 8.8	31.2± 4.6	6.5± 1.4#
Pancreas	M	14.2± 6.8#	173.5±76.7#	23.8± 5.3#	5.9± 2.7#
	F	10.6± 3.6#	126.1±24.9#	26.2± 5.8#	6.7± 1.8#
Prostate	M	1.7± 0.5#	18.2± 3.2	12.7± 5.2#	10.1± 2.0
L. Testis	M	6.3± 0.9	33.9± 1.2	20.7± 2.1	15.9± 1.1
R. Testis	M	6.7± 0.7	33.1± 2.1	20.6± 2.1	15.7± 1.6
Uterus	F	4.1± 2.1#	93.0±35.2*#	105.7±41.2**#	

Mean ± SD.
* : the length of the corpus uteri.
** : the length of the cornu uteri.
# : parameters which CV are over 20%.

From Table 5, the activity of LDH (143.9±41.0 IU/l in the male and 124.3±35.1 IU/l in the female) differed significantly ($p<0.05$) between the sexes, that of the male being higher. The activity of LDH in the 6-year-old male group (175.3±39.8 IU/l) was significantly higher than any other male age group. Significant difference ($p<0.05$) between the sexes was also found in the activity of lipase (181.3±40.6 IU/l in the male and 143.5±51.1 IU/l in the female), that of the male being higher. No significant differences within each age group or between the sexes were observed in other parameters.

The LDH isoenzyme patterns are shown in Fig. 1. LDH1 and LDH5 were dominant. The patterns of the CK isoenzyme are shown in Fig. 2. MM was dominant.

TABLE 3. Reference values of serum biochemical parameters from silver foxes aged from 2 to 8 years.

PARAMETERS	SEX	AGE (year) 2 ((M:3. F:8)¥	4 (M:0. F:4)	5 (M:6. F:2)	6 (M:7. F:4)	7 (M:3. F:5)	8 (M:0. F:2)	TOTAL (M:20. F:25)
T-Bil**	M	0.9± 0.1		1.1± 0.1	0.9± 0.2	0.7± 0.2		0.9± 0.3
(mg/dl)	F	0.8± 0.3	0.7± 0.0	0.8± 0.0	0.7± 0.2	0.7± 0.1	0.8± 0.0	0.7± 0.2
T-Cho (mg/dl)	M	136.7±12.3		132.2±14.8	119.3± 7.4	136.0±19.4		128.7±15.1
	F	130.5±27.4	125.7±16.0	122.5± 3.5	121.5±14.9	131.0±17.1	142.5±13.5	129.7±20.7
β-Lipo**	M	31.3±11.5		21.5± 4.8	29.3± 8.0	28.3± 4.5		27.0± 8.3
(mg/dl)	F	44.5±27.1	47.7± 4.0	31.0± 1.0	28.8± 9.2	45.4±12.9	39.0± 1.0	41.4±18.4
TG(mg/dl)	M	24.3± 0.5		24.0± 2.9	26.1± 6.4	29.0± 4.3		25.6± 4.9
	F	28.4±11.5	27.3± 3.3	22.0± 1.0	21.6± 5.2	28.0± 5.7	31.0± 0.0	26.9± 7.9
Phos-Lipo	M	301.0±17.2		287.2±28.3	277.9±19.7	301.3±31.6		288.2±26.3
(mg/dl)	F	282.9±53.6	288.6±35.6	276.0± 1.0	275.3±33.2	300.4±43.1	298.0±22.0	287.7±42.1
UA (mg/dl)	M	0.6± 0.0		0.5± 0.1	0.6± 0.1	0.6± 0.0		0.6± 0.1
	F	0.7± 0.1	0.5± 0.0	0.6± 0.1	0.6± 0.1	0.6± 0.2	0.6± 0.1	0.6± 0.1
BUN (mg/dl)**	M	14.5± 1.3		18.3± 3.2	19.8± 2.1	22.1± 2.9#		18.9± 3.4
	F	15.0± 3.2	14.6± 1.0	13.4± 1.1	16.1± 1.6	14.9± 2.4	14.9± 0.3	14.9± 2.4
Crea (mg/dl)	M	0.7± 0.0		0.8± 0.1	0.8± 0.1	0.7± 0.0		0.8± 0.1
	F	0.8± 0.1	0.7± 0.0	0.8± 0.0	0.8± 0.1	0.7± 0.0	0.8± 0.1	0.8± 0.1
Glu (mg/dl)*	M	108.0±10.9		118.8±18.2	132.6±13.4	171.0±35.6#		130.4±27.8
	F	188.8±70.4	123.3±11.1	160.5± 6.5	133.0±18.1	154.4±34.0	131.0±14.0	157.4±50.1
Tp (g/dl)	M	7.0± 0.2		6.8± 0.5	7.2± 1.0	7.2± 0.9		7.1± 0.8
	F	6.8± 0.4	7.0± 0.5	7.4± 0.0	6.1± 0.8	5.8± 0.4	6.3± 0.7	6.5± 0.7

* ($P<0.05$). ** ($P<0.01$) : parameters for which significant differences are detected between the sexes.
# : parameters for which significant differences are detected within each age group.
¥ : numbers of animals for each age group and sex.
F : female. M : male.

TABLE 4. Reference values of Serum biochemical parameters from silver foxes aged from 2 to 8 years.

PARAMETERS	SEX	AGE (year) 2 ((M : 3. F : 8) ¥	4 (M : 0. F : 4)	5 (M : 6. F : 2)	6 (M : 7. F : 4)	7 (M : 3. F : 5)	8 (M : 0. F : 2)	TOTAL (M : 20. F : 25)
Ca (mg/dl)	M	4.8± 0.2		4.7± 0.2	4.7± 0.1	4.4± 0.3		4.7± 0.2
	F	4.7± 0.1	4.9± 0.2	4.8± 0.1	4.6± 0.2	4.8± 0.2	4.9± 0.1	4.7± 0.2
IP (mg/dl)	M	3.6± 0.8		3.2± 0.6	3.5± 0.4	3.1± 0.2		3.4± 0.6
	F	3.3± 0.7	3.5± 0.1	3.2± 0.4	3.7± 0.6	3.2± 0.7	3.7± 0.4	3.4± 0.6
Mg (mg/dl)*	M	2.0± 0.3		1.7± 0.1	1.8± 0.3	1.5± 0.2		1.7± 0.3
	F	2.2± 0.3	1.8± 0.1	1.9± 0.1	2.0± 0.1	1.6± 0.1	1.8± 0.2	1.9± 0.3
Fe (μg/dl)	M	180.3±11.1		208.7±16.5#	147.3±40.2	167.7±25.7		175.1±38.2
	F	144.9±21.9	130.0±21.4	143.0± 0.0	129.0±15.1	158.4±24.4	162.5±12.5	144.9±22.6
Ma (mEq/l)	M	124.7± 2.6		125.5± 1.6	123.3± 5.3	123.7± 1.7		124.2± 3.8
	F	126.4± 5.9	129.7± 1.2	130.0± 1.0	127.8± 0.8	126.0± 2.0	122.0± 1.0	126.9± 4.1
K (mEq/l)	M	4.3± 0.0		4.2± 0.2	4.3± 0.4	3.6± 0.1		4.2± 0.3
	F	4.4± 0.7	4.0± 0.1	4.6± 0.1	4.1± 0.2	3.9± 0.2	3.7± 0.3	4.1± 0.5
Cl (mEq/l)	M	93.0± 0.8		92.5± 2.4	89.8± 2.7	88.0± 0.8		90.8± 2.9
	F	91.8± 4.9	96.0± 0.8	94.5± 0.5	95.0± 1.0	92.8± 1.9	90.0± 1.0	93.4± 3.7

* ($P<0.05$) : parameters for which significant differences are detected between the sexes.
# : parameters for which significant differences are detected within each age group.
¥ : numbers of animals for each age group and sex.
F : female. M : male

TABLE 5. Reference values of Serum biochemical parameters from silver foxes aged from 2 to 8 years

PARAMETERS	SEX	AGE (year) 2 ((M : 3. F : 8) ¥	4 (M : 0. F : 4)	5 (M : 6. F : 2)	6 (M : 7. F : 4)	7 (M : 3. F : 5)	8 (M : 0. F : 2)	TOTAL (M : 20. F : 25)
AST (IU/l)	M	6.7± 2.6		40.5± 4.5	46.9± 8.0	41.0± 5.1		42.6± 7.2
	F	43.9±10.5	40.7±11.1	46.5± 0.5	41.7± 6.6	41.0± 1.8	46.0± 7.0	42.7± 8.1
ALT (IU/l)	M	55.0±17.0		70.7±32.5	55.3± 2.8	61.7± 6.2		61.1±20.9
	F	59.5±12.8	50.0±13.4	59.0± 8.0	49.5± 7.9	52.6± 9.4	62.5± 5.5	56.2±12.0
ALP (IU/l)	M	50.0±12.8		62.7±27.4	57.0±13.0	50.7± 8.3		56.7±19.0
	F	54.6±11.8	53.3± 9.8	56.5± 5.5	52.5± 9.1	53.2± 6.9	62.5± 5.5	54.2± 9.8
LDH (IU/l)*	M	119.0±13.6		130.0±27.4	175.3±39.8#	113.0±26.9		143.9±41.0
	F	121.8±30.1	106.3±10.9	101.0±16.0	113.5±10.2	152.2±41.7	138.0±58.0	124.3±35.1
GGT (IU/l)	M	1.3± 0.5		1.2± 0.4	1.6± 0.5	1.3± 0.5		1.4± 0.5
	F	1.5± 0.5	1.7± 0.9	1.0± 0.0	1.5± 0.5	1.0± 0.0	1.0± 0.0	1.4± 0.6
CK (IU/l)	M	71.3±16.0		80.5±22.4	101.9±38.4	62.0±17.8		84.9±32.4
	F	136.1±186.7	123.7±68.8	75.5±14.5	101.3±35.4	93.6±40.0	204.5±161.5	118.9±124.2
Lipase*	M	151.0±22.7		198.8±32.9	178.3±26.0	182.7±65.7		181.3±40.6
(IU/l)	F	164.0±43.2	119.0±15.9	149.5±27.5	126.0±23.2	160.0±56.4	300.5±127.5	143.5±51.1

* ($P<0.05$) : parameters for which significant differences are detected between the sexes.
# : parameters for which significant differences are detected within each age group.
¥ : numbers of animals for each age group and sex.
F : female. M : male

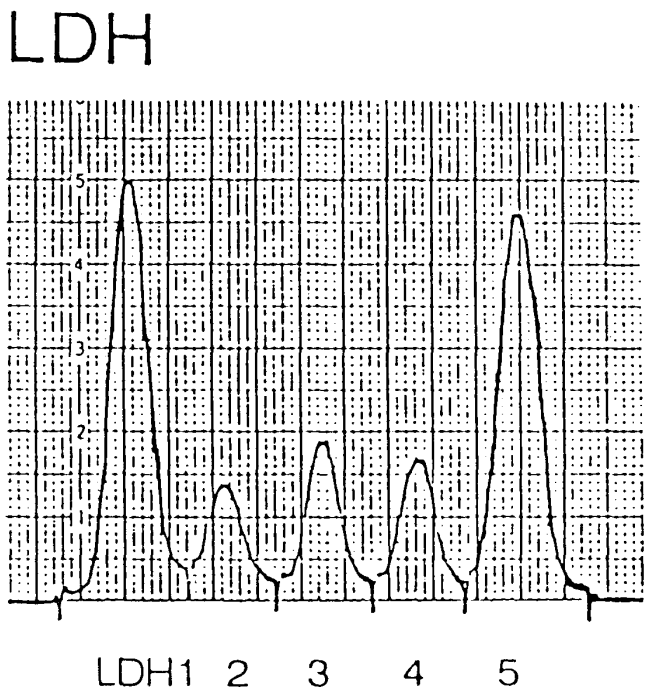

FIGURE 1. : Patterns of the serum LDH isoenzyme of the silver fox

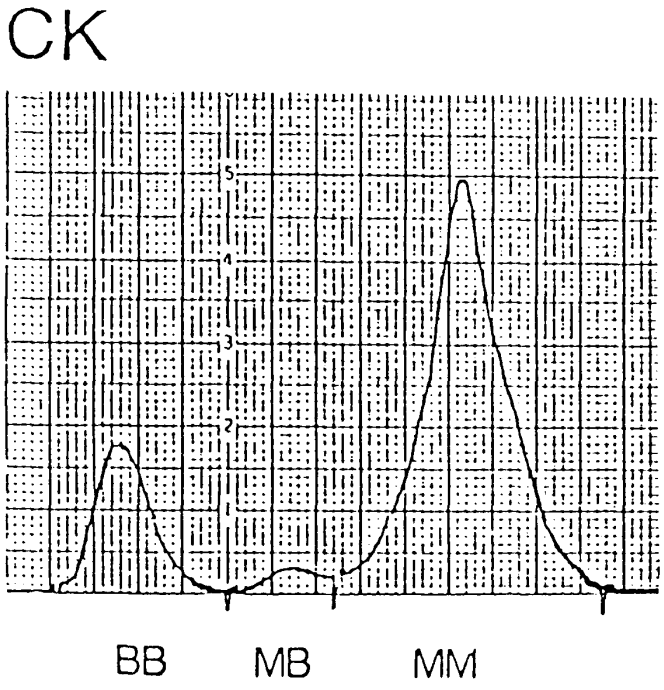

FIGURE 2. : Patterns of the serum CK isoenzyme of the silver fox

DISCUSSION

Since the silver fox is a fur-bearing animal, rather than hematological and biochemical studies, behavior, nutrition, genetics and reproduction have commonly been investigated in the past[2, 6, 9, 13, 14, 17, 18]. There is a necessity to establish base-line reference values for silver fox. Clinically healthy silver foxes were sampled and reference values for anthropometry, anatomical and serum biochemical parameters are presented in this report.

CV of the anthropometric measurements were low, with the exception of body weight of the female, indicating that the body size of the silver fox is uniform. This may be because the study was conducted using a closed colony. The body weight and body length obtained in this study were greater than those previously reported [16], but similar to the values for the red fox[10]. The body size of the male was larger than that of the female, as found in the reference literature[10, 16].

The pancreas, spleen and uterus being exceptions, the measurements for the main organs were uniform. These results were equivalent to those for beagle dogs aged between 3 and 5 months[5]. However, the weight of the liver from the silver fox (189.0 ± 20.6 g in the male and 165.2 ± 17.0 g in the female) was markedly lighter than those from a beagle of 3 months old (male 231 ± 19.5 g, female 286 ± 31.5 g)[5]. The measurements of the main organs from the male were larger than those from the female. This may be due to the larger body size of the male. The high CV of the pancreas may be derived from man-made errors, for instance, difficulties were met in removing attachments from the pancreas clearly. The high CV of the uterus may be due to individual differences.

The mean values of the biochemical parameters presented were statistically similar within each age group and for the sexes. However, the concentration of Fe from the 5-year-old male age group, BUN and Glu from the 7-year-old male group, and the activity of LDH from the 6-year-old male group were significantly higher than those of the other age groups. There have been no previous reports of the significance test being carried out between age groups for the silver fox. The results may be affected by the small sample size in each age group. Therefore, a larger sample size may have been needed to make the reference values more significant.

Significant differences between the sexes were detected in the following parameters: concentration of BUN, β-Lipo, and T-Bil ($p<0.01$), concentration of Mg and Glu ($p<0.05$), and the activity of LDH and lipase ($p<0.05$). A sex-linked increase in renal excretion of T-Bil was reported in male dogs[3, 4]. This appears to be opposite to the phenomenon observed here. A similar result for T-Bil has been observed previously by Benn[1]. However, in his report, no significant differences were seen between the sexes in the concentrations of BUN and Glu. These results differ from the present study.

Comparing the biochemical reference values for the silver fox presented in this report with that for the dog reported previously[11, 14, 19], the concentration of Glu was higher, while those of Cl and Na were lower. This differed from the results for the swift fox published by Mainka in which concentrations of Glu, Cl and Na were the same levels as in the dog[12]. Benn's studies on the mixed silver and red fox indicated that the concentration of Glu was higher, but those of Cl and Na were the same as the dog[1]. The activity of LDH was higher and the isozyme patterns were also different from the dog. Both LDH1 and LDH5 were dominant in silver fox, but only LDH5 was dominant in the dog[8]. The activity of CK was found to be the same as the dog. The isozyme patterns were similar to the dog for which MM was dominant, but with BB also present[7].

The reference values presented in this report for the silver fox will be valuable as a guide for clinical diagnosis and research.

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