

## Variations of Plasma Thyroid Hormones Concentrations and Their Percentages During Different Ages of Sarabi Calves

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**Abstract:** The thyroid is a main endocrine gland that secretes its hormones such as thyroxine ( $T_4$ ) and free-triiodothyronine ( $T_3$ ). Many factors (i.e., gender, age and other physiological status) have complex effect on their levels in blood of animals. But, there is a little information about the changes of plasma thyroid hormones concentrations during different ages of domestic animals. In this study, variations in the plasma  $T_4$ ,  $T_3$ , free thyroxine ( $fT_4$ ), free triiodothyronine ( $fT_3$ ) concentrations and their percentages,  $T_4:T_3$  and  $fT_4:fT_3$  ratio during different ages and of different sexes in Iranian Sarabi calves were investigated. Blood samples were taken from the jugular vein of 42 clinically healthy animals in three age groups (1-14 days, 1-2 and 3-6 months) in autumn. The plasma was analyzed to determine thyroid hormones (i.e.  $T_4$ ,  $T_3$ ,  $fT_4$  and  $fT_3$ ) concentrations using chemiluminescence immunoassay (CLIA) method. Results indicated a significant difference in the  $T_4$  and  $fT_4$  concentrations among age groups ( $P<0.05$ ) and the highest levels of thyroid hormone were seen during the first two weeks after birth. No gender-dependent significant differences and correlation in the studied parameters were found. It was concluded that age (significantly), season and gender and their related physiological and endocrinological changes have the highest effect on the plasma thyroid hormones concentrations in Sarabi calves.

**Key words:** Thyroid hormones • Plasma • Chemiluminescence immunoassay • Calve • Age effect

### INTRODUCTION

The thyroid is an endocrine gland that secretes mainly thyroxine hormone. Thyroid hormones are involved in the regulation of metabolic events of different components in various body tissues, growth, reproductive performance, development and adaptation in ruminant and other farm animals [1-3]. Many factors such as age, breed, nutrition, gender, season, climate and geographical traits of the region, the rearing technology, some physiological conditions, method of measurement, drugs and disease effects on the plasma concentrations of thyroid hormones [3-10]. These hormones circulate in blood bound to proteins, but small amounts of thyroid hormones are found in free form and unbound to protein ( $fT_4$  and  $fT_3$ ). Only the free

hormone is responsible for the biological activity of thyroid hormones and available to tissues [3, 11]. The effect of age on plasma thyroid hormone levels was investigated in some farm animals and human. In most studies, the highest levels of thyroid hormones were reported during the first period of life, an age advance results in decreases of these hormone levels [3, 4, 12]. In most species, the percentage of  $T_4$  is higher than the other thyroid hormones in any period of life; and the free forms of these hormones have a little percentage. There is little information about the changes of plasma thyroid hormones concentrations during different ages of calves. So, the main objective of this study was to determine the effect of age on the plasma thyroid hormones concentrations and their percentages in Iranian Sarabi Calves.

**MATERIALS AND METHODS**

This study was performed on blood samples collected from forty-two Sarabi calves at the Improvement Center of Sarabi cow, located in East Azarbaijan province, Iran. Calves were divided into three age groups (1-14 days, 1-2 months and 3-6 months) and by gender. Animals were clinically healthy, free from internal and external parasites. Blood samples were taken before feeding, from the jugular vein, using disposable syringes containing lithium heparin (1,000 IU/ml). All samples were taken in autumn when the mean ambient temperature was 15°C. Plasma was separated by centrifugation at 750×g for 15 min and stored at -20°C until used. The levels of plasma T<sub>4</sub>, T<sub>3</sub>, fT<sub>4</sub> and fT<sub>3</sub> were measured by DiaSorin CLIA kits (Stradaper Crescentino-13040 Saluggia (Vercelli)-Italy) with the LIAISON analyzer. The validation for these hormones assays assessed the limits of detection, precision of standard curve following sample dilution and intra- and inter-assay coefficient of variation of the

results. The data were analyzed by one way ANOVA and Pearson's correlation method using SPSS/ver.17 software. Duncan's multiple range tests was used to detect significant differences between the means. All values shown as mean ± standard error (SE).

**RESULTS**

The mean and standard error (SE) of the plasma concentrations of thyroid hormones (T<sub>4</sub>, T<sub>3</sub>, fT<sub>4</sub> and fT<sub>3</sub>), T<sub>4</sub>: T<sub>3</sub> and fT<sub>4</sub>: fT<sub>3</sub> ratio, also the percentage of these hormones of Iranian Sarabi calves in different age groups are presented in Tables 1 and 2, respectively. Also, the values of the studied parameters in each age group according to gender are presented in Tables 3 and 4.

Additionally, correlation coefficients between the measured parameters are presented in Table 5. There were no significant correlations between age groups for any of the parameters. Also, gender had no significant effect on all of the studied parameters.

Table 1: The plasma concentrations of thyroid hormones, T<sub>4</sub>, T<sub>3</sub> and fT<sub>4</sub>: fT<sub>3</sub> ratio of Iranian Sarabi calves in different age groups

Age	N		T <sub>4</sub> (nmol/l)	T <sub>3</sub> (nmol/l)	fT <sub>4</sub> (pmol/l)	fT <sub>3</sub> (pmol/l)	T <sub>4</sub> : T <sub>3</sub>	fT <sub>4</sub> : fT <sub>3</sub>
1-14days	14	Mean	77.43 <sup>a</sup>	2.04	24.88 <sup>a</sup>	11.50	39.00	2.27
		SE	13.06	0.41	2.68	1.28	4.01	0.50
1-2months	14	Mean	57.86 <sup>b</sup>	1.56	19.57 <sup>b</sup>	9.29	53.32	2.23
		SE	4.64	0.33	1.62	1.17	14.41	0.20
3-6months	14	Mean	76.83 <sup>a</sup>	1.53	24.54 <sup>a</sup>	10.56	60.55	2.33
		SE	3.14	0.25	0.98	0.40	9.29	0.08
Total	42	Mean	68.94	1.62	22.50	10.17	54.10	2.28
		SE	3.61	0.18	1.03	0.56	7.17	0.11

\* There are significant differences among groups with different codes in a column (a, b) in each parameters (P<0.05)

\*\* SE: standard error

Table 2: The percentages of plasma thyroid hormones of Iranian Sarabi calves in different age groups

Age	N		%T <sub>4</sub>	%T <sub>3</sub>	%fT <sub>4</sub>	%fT <sub>3</sub>
1-14days	14	Mean	97.40	1.730	0.031	0.0150
		SE	0.24	0.175	0.005	0.0031
1-2months	14	Mean	97.36	1.700	0.033	0.0160
		SE	0.50	0.290	0.003	0.0015
3-6months	14	Mean	98.04	1.320	0.031	0.0140
		SE	0.28	0.190	0.001	0.0006
Total	42	Mean	97.65	1.540	0.030	0.0150
		SE	0.25	0.150	0.001	0.0008

\* There is no a significant difference among groups concerning the percentages of plasma thyroid hormones

\*\* SE: standard error

Table 3: The plasma concentrations of thyroid hormones, T<sub>4</sub>:T<sub>3</sub> and fT<sub>4</sub>: fT<sub>3</sub> ratio of Iranian Sarabi calves according to gender

Age	N	Gender	T <sub>4</sub> (nmol/l)	T <sub>3</sub> (nmol/l)	fT <sub>4</sub> (pmol/l)	fT <sub>3</sub> (pmol/l)	T <sub>4</sub> : T <sub>3</sub>	fT <sub>4</sub> : fT <sub>3</sub>	
1-14 days	6	Male	Mean	77.73	2.24	24.98	11.61	39.12	2.37
			SE	12.96	0.45	2.76	1.31	3.91	0.58
	8	Female	Mean	77.43	2.04	24.75	11.51	39.00	2.27
			SE	13.06	0.42	2.68	1.28	4.01	0.50
1-2 months	7	Male	Mean	47.02	0.79	17.42	6.91	83.84	2.69
			SE	2.60	0.23	0.21	1.34	32.67	0.42
	7	Female	Mean	64.37	2.05	20.87	10.72	35.01	1.96
			SE	5.48	0.37	2.50	1.40	6.17	0.09

Table 3: Continued

Age	N	Gender		T <sub>4</sub> (nmol/l)	T <sub>3</sub> (nmol/l)	fT <sub>4</sub> (pmol/l)	fT <sub>3</sub> (pmol/l)	T <sub>4</sub> : T <sub>3</sub>	fT <sub>4</sub> : fT <sub>3</sub>
3-6 months	7	Male	Mean	72.49	1.58	25.42	10.70	62.10	2.37
			SE	3.35	0.49	1.53	0.35	17.71	0.09
	7	Female	Mean	81.18	1.50	23.68	10.43	59.00	2.30
			SE	4.71	0.26	1.31	0.80	9.38	0.16
Total	20	Male	Mean	61.57	1.24	21.99	9.08	71.42	2.51
			SE	5.54	0.32	1.81	0.94	16.17	0.18
	22	Female	Mean	73.24	1.86	22.81	10.82	44.00	2.15
			SE	4.43	0.20	1.30	0.67	4.50	0.13

\* SE: standard error

Table 4: Correlation coefficients and statistical analysis between the measured parameters for total samples (n=42)

	T <sub>4</sub>	T <sub>3</sub>	fT <sub>4</sub>	fT <sub>3</sub>	T <sub>4</sub> : T <sub>3</sub>	fT <sub>4</sub> : fT <sub>3</sub>	%T <sub>4</sub>	%T <sub>3</sub>	%fT <sub>4</sub>	%fT <sub>3</sub>
T <sub>4</sub> Pearson's correlation	1	-0.189	0.155	-0.027	0.235	0.069	0.269	-0.271	-0.089	-0.224
Sig.(two-tailed)		0.439	0.527	0.912	0.334	0.780	0.266	0.263	0.718	0.357
T <sub>3</sub> Pearson's correlation		1	0.091	0.350	-0.435	-0.363	-0.311	0.358	-0.315	0.059
Sig.(two-tailed)			0.712	0.141	0.063	0.127	0.195	0.132	0.190	0.810
fT <sub>4</sub> Pearson's correlation			1	0.525*	-0.250	-0.276	0.003	0.100	-0.606**	-0.363
Sig.(two-tailed)				0.021	0.303	0.253	0.991	0.683	0.006	0.127
fT <sub>3</sub> Pearson's correlation				1	-0.793**	-0.201	-0.875**	0.908**	0.070	0.250
Sig.(two-tailed)					0.000	0.409	0.000	0.000	0.777	0.302
T <sub>4</sub> :T <sub>3</sub> Pearson's correlation					1	0.170	-0.421	0.389	0.384	0.180
Sig.(two-tailed)						0.486	0.072	0.100	0.105	0.461
fT <sub>4</sub> :fT <sub>3</sub> Pearson's correlation						1	-0.467*	0.446	-0.016	0.595**
Sig.(two-tailed)							0.044	0.055	0.949	0.007
%T <sub>4</sub> Pearson's correlation							1	-0.985**	-0.379	-0.494*
Sig.(two-tailed)								0.000	0.109	0.032
%T <sub>3</sub> Pearson's correlation								1	0.240	0.379
Sig.(two-tailed)									0.322	0.110
%fT <sub>4</sub> Pearson's correlation									1	0.514*
Sig.(two-tailed)										0.024
%fT <sub>3</sub> Pearson's correlation										1
Sig.(two-tailed)										

\* SE: standard error

Table 5: The percentages of plasma thyroid hormones of Iranian Sarabi calves according to gender

Age	N	Gender		%T <sub>4</sub>	%T <sub>3</sub>	%fT <sub>4</sub>	%fT <sub>3</sub>
1-14 days	6	Male	Mean	97.510	1.760	0.030	0.02
			SE	0.220	0.190	0.004	0.002
	8	Female	Mean	97.410	1.730	0.030	0.01
			SE	0.240	0.170	0.005	0.003
1-2 months	7	Male	Mean	98.330	1.060	0.030	0.01
			SE	0.618	0.390	0.002	0.002
	7	Female	Mean	96.780	2.080	0.030	0.02
			SE	0.620	0.300	0.004	0.002
3-6 months	7	Male	Mean	97.900	1.380	0.030	0.01
			SE	0.550	0.384	0.002	0.0005
	7	Female	Mean	98.180	1.260	0.030	0.01
			SE	0.220	0.177	0.002	0.0007
Total	20	Male	Mean	98.080	1.240	0.030	0.01
			SE	0.380	0.260	0.001	0.0008
	22	Female	Mean	97.400	1.720	0.030	0.01
			SE	0.310	0.177	0.002	0.001

\* Correlation is significant at the 0.05 level (2-tailed)

\*\* Correlation is significant at the 0.01 level (2-tailed)

## DISCUSSION

In this study, total means of  $T_4$  concentrations were lower than the values reported in buffalo calves [12], calves [13] and in dromedary camels [14] and higher than the values reported in sheep [4] and lambs [15]. However, these results are consistent with the values reported in cow [16]. The total means of  $T_3$  has a little difference with the values reported in sheep [4] and higher than the values reported in calves [13] and goats [17], but the levels of this hormone were lower than the other values reported in buffalo calves [12], goats [8] and in cow [16]. Neonatal lambs and kids had higher levels of  $T_4$  and  $T_3$  as compared with growing lambs and adults [18, 19]. Also, the lowest values of these hormones were reported in elderly animals [7]. Additionally, the levels of  $fT_4$  were higher and of  $fT_3$  were lower in the Iranian Sarabi calves than the other report in sheep [4], respectively. In this study,  $T_4$ :  $T_3$  and  $fT_4$ :  $fT_3$  ratio did not vary significantly among various age groups. This result was consistent with the other report in buffalo calves [1]. In addition, the percentages of thyroid hormones in Iranian Sarabi calves have a little difference with the values reported in sheep [20].

There are no gender-dependent significant differences in plasma thyroid hormones of Sarabi calves; this result was consistent with those reported in goats [3] and sheep [4]. Age had significant effect only on plasma  $T_4$  and  $fT_4$  concentrations in the Sarabi calves, with values being lower in 1-2 months-old calves. According to these data from the Iranian Sarabi calves, there was no significant correlation between the age groups concerning the studied parameters. The lowest value of  $T_4$ :  $T_3$  ratio was seen during the first two weeks after birth followed by a gradual increase until 3-6 months of age. As, with an advance of age, the levels of  $T_3$  being lower and of  $T_4$  being higher, so this changes for  $T_4$ :  $T_3$  ratio seems normal. Unfortunately, there is little data about the changes of these ratios in farm animals. In most species, the levels of plasma  $T_4$  are higher than the other thyroid hormones, especially during the first weeks of life [4, 8, 12, 19]. Similar to other researches, the highest concentrations of thyroid hormones and their percentages were observed during the early two weeks of life in the Iranian Sarabi calves. The most important factors which they effects on the plasma concentrations of thyroid hormones are age, breed, nutrition, gender, season, climate and geographical traits of the region, hair fiber growth, the rearing technology, some physiological conditions, method of measurement and

disease [3-5, 21-24]. These factors maybe affects the levels of plasma thyroid hormones synchronously at any time, so in this study the exact explanation of these differences with other results is not possible.

In conclusion, it seems that age (significantly) and other factors particularly season and gender and their related physiological and endocrinal changes have the highest effect on the plasma thyroid hormones concentrations in Sarabi calves.

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