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The Protein and Protein-Bound Carbohydrates in Four Species of Nonhuman Primates' Sera (32858)

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Nonhuman primates constitute the group of animals phylogenetically, anatomically, physiologically, biochemically, and immunologically closest to man. Although it is widely believed and predicated that biochemical experimentation on nonhuman primates will throw light on human metabolism, our knowledge concerning the chemical composition of their various tissues and fluids is still very primitive.

Serum is the most important body fluid which is readily accessible. Among the major constituents of serum are the proteins of which the glycoproteins are an important subgroup. This paper reports comparative studies of total serum protein and glycoprotein levels of *Macaca mulatta* (rhesus), *Theropithecus gelada* (baboon), *Hylobates lar* (gibbon), and *Pan troglodytes* (chimpanzee).

Materials and Methods. Serum samples were obtained from various nonhuman primate colonies available at the Delta Regional Primate Research Center of Tulane University. Some serum samples of *Macaca mulatta* were kindly made available by Dr. D. R. Anderson of the USAF School of Aerospace Medicine, Brooks Air Force Base, Texas. All of the serum samples were kept in a deep freezer (—25°C) until analyzed.

Total serum protein was determined by the biuret method according to Weichselbaum (1) using Lab-trol (Dade Reagents, Inc., Miami, Florida) as a standard. Hexose associated with protein was determined by the tryptophane–H_2SO_4 method described by Shetlar *et al.* (2). The Winzler (3) modifications of the cystein–sulfuric acid method and of the Elson-Morgan reaction were used for the determination of protein-bound fucose and hexosamine, respectively. Sialic acid associated with protein was determined by the thiobarbituric acid method of Warren (4) after hydrolysis of the protein sample with 0.1 *N* H_2SO_4 at 80°C for 1 hour. Detailed procedures for the hydrolysis of serum samples for sialic acid determination were recently described by Shetlar (5). Microelectrophoresis of the serum protein on a cellulose acetate membrane was performed according to the procedures described by Grunbaum *et al.* (6,7).

Results. The analytical data on serum protein and protein-bound carbohydrates of the four species of nonhuman primates are summarized in Table I. For comparison, the reported values for humans are also included.

The total serum protein concentrations of *Pan troglodytes, Theropithecus gelada,* and *Macaca mulatta* are very close to human total serum protein levels, while that of *Hylobates lar* is significantly lower. Protein-bound hexose concentration was found to be higher than

TABLE I. Protein and Protein-Bound Carbohydrate Levels in 4-Species of Adult Nonhuman Primate Sera.

Species	No. of animals	Body wt. (kg)	Sex[a]		Protein (gm/100 ml)	Protein-bound sugars (mg/100 ml)			
			F	M		Hexose	Hexosamine	Sialic acid	Fucose
Pan troglodytes (chimpanzee)	26	30–60	16	10	7.6 ± 0.34[b] (6.9–8.3)	129.6 ± 15.0 (107.2–158.4)	106.3 ± 7.9 (91.6–120.8)	61.0 ± 7.5 (50.9–72.6)	11.9 ± 1.6 (7.1–14.7)
Hylobates lar (gibbon)	19	5–8	10	9	6.4 ± 0.44 (5.8–7.2)	112.7 ± 10.1 (99.1–128.4)	81.8 ± 9.8 (81.8–96.3)	58.8 ± 9.8 (46.4–76.6)	8.7 ± 0.7 (7.1–10.4)
Theropithecus gelada (baboon)	16	7–12	6	10	6.9 ± 0.35 (6.3–7.4)	139.8 ± 11.9 (121.1–157.8)	93.1 ± 7.8 (79.0–106.0)	57.3 ± 5.8 (49.1–67.7)	9.1 ± 0.8 (8.2–10.5)
Macaca mulatta (rhesus monkey)	62	6–10	32	30	7.3 ± 0.44 (6.5–8.2)	121.8 ± 12.4 (99.2–146.2)	110.1 ± 9.3 (96.0–129.7)	62.7 ± 5.0 (52.4–75.1)	9.2 ± 1.2 (7.2–11.8)
Human					7.25 ± 0.75[c] (5.75–8.75)[c]	121.0 ± 2.1[d]	83.0 ± 4.1[d]	60.0 ± 7[d]	8.9 ± 0.6[d]

[a] F, female; M, male.

[b] Values are the mean ± SD, standard deviation; the range is given in parentheses.

[c] Grunbaum *et al.* (6), by micro-Kjeldahl analysis of 221 Caucasian males, 15–35 years of age.

[d] Winzler (3), analysis of 20 normal adults; expressed as mean ± SE.

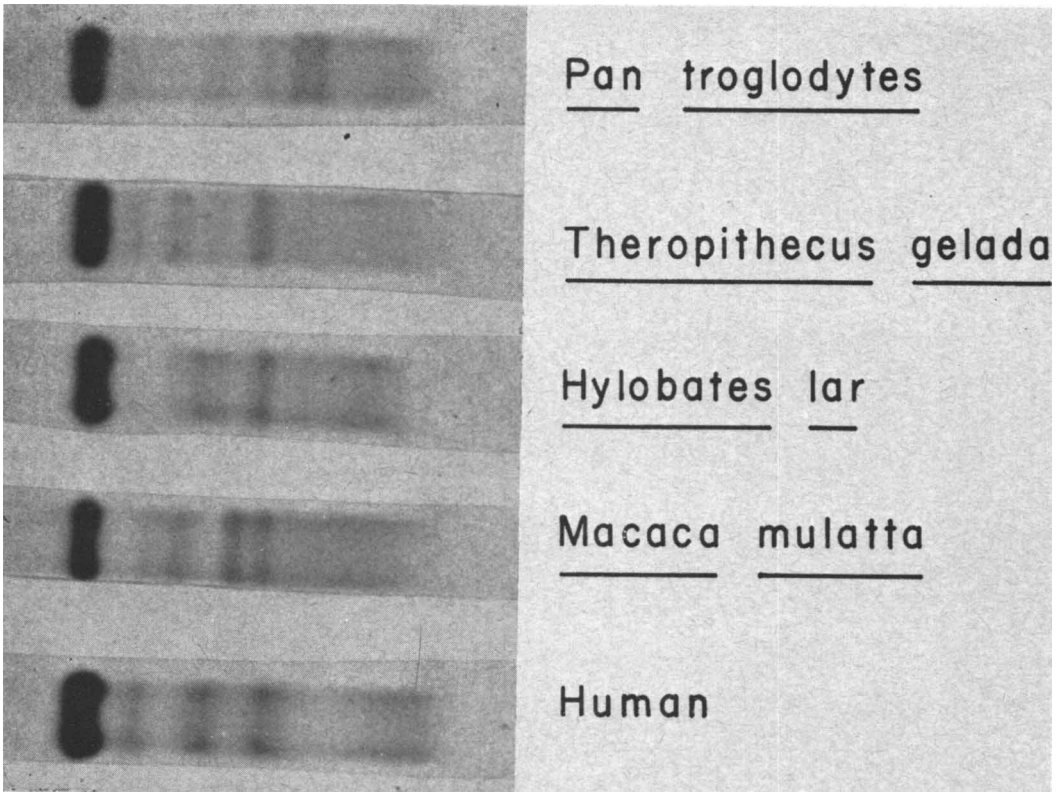

FIG. 1. Electrophoretic patterns of serum proteins in various nonhuman primates. Eletrophoresis was performed at room temperature on a cellulose acetate membrane for 20 min at a constant voltage of 250 with veronal buffer, pH 8.6, ionic strength 0.075 (6, 7).

human levels only in *Theropithecus gelada;* the values of the other three species are fairly close to that of humans. In all species studied except *Hylobates lar,* protein-bound hexosamine concentrations are considerably higher than those of humans. Generally, the levels of sialic acid and fucose associated with serum protein in the four species examined are comparable to human levels.

Discussion. Although Asatiani (8) reported the levels of protein-bound carbohydrates in rhesus monkeys, no information was given concerning the size and the number of animals used in his report. It is of interest to note that levels of hexosamine associated with serum protein in *Theropithecus gelada, Pan troglodytes,* and *Macaca mulatta* are significantly higher than in humans. Since serum protein-bound sugars are greatly elevated in various disease states (3,5), the sera used in this study were obtained only from healthy animals. It is, however, impossible to exclude those animals which may have had subclinical disease.

Attempts were also made to compare the relative distribution of protein and protein-bound carbohydrates in the electrophoretic fractions by microelectrophoresis of the serum proteins on a cellulose acetate membrane. As shown in Fig. 1, the serum electrophoretic patterns among various nonhuman primates studied are so different that it is impossible to draw any conclusions from these studies.

Summary. Blood sera from four species of nonhuman primates were analyzed for their protein and protein-bound sugar content. Generally, the levels of total protein, protein-bound hexose, sialic acid, and fucose of *Pan troglodytes* (chimpanzee), *Theropithecus gelada* (baboon), and *Macaca mulatta* (rhesus monkey) are comparable to those of humans. In all species examined except *Hylobates lar* (gibbon), the protein-bound hexosamine levels

are considerably higher than those of humans. In *Hylobates lar,* the total serum protein concentration is lower than in humans, while the protein-bound hexosamine level is very close to human levels. The electrophoretic patterns of the four species of nonhuman primates examined are very different from that of humans and from each other.

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Inhibition of Water Diuresis in *Mustelus canis* by Extracts of Homologous Brain and Spinal Cord* (32859)

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Marine elasmobranchs maintain their body fluids hyperosmotic to sea water, continually taking up water through their gills and integument, while maintaining water balance through the renal excretion of a urine which is hypo-osmotic to both sea water and plasma. An increased uptake of water occurs when these fish migrate to fresh water or are placed in dilute sea water. In these circumstances plasma osmolality becomes reduced, urine flow rate and glomerular filtration rate increase, and the ratio of urine osmolality to plasma osmolality decreases (1). A similar response to dilution is observed in higher vertebrates (2–4) and it has been repeatedly suggested that endocrine control of water excretion exists in the elasmobranchs as it does in higher forms. In teleosts the caudal neurosecretory system of the spinal cord has been implicated in the control of water excretion, and the neurosecretory cells in the spinal cords of elasmobranchs have been considered as having analogous functions (5,6). However, despite many conjectures no experimental evidence has supported the presence of a humoral factor capable of influencing water excretion in elasmobranchs, nor is there evidence directly implicating the caudal neurosecretory cells or any other elasmobranch tissue as the source of these postulated factors. In a search for such factors we have examined the effect of injections of extracts of various tissues taken from the smooth dogfish, *Mustelus canis,* on the renal function of other *Mustelus* undergoing water diuresis while in dilute sea water.

Methods. Donor fish, (male and female, weighing between 1 and 4 kg) were placed in fresh running dilute sea water (70–80%) for 2–3 days, a period sufficient to promote development of the maximum drop in plasma osmolality and the maximum water diuresis obtainable in this salinity. Donor fish were then anesthetized with intravenous sodium pentobarbital. Tissues were removed and frozen. Four to 5 gm of tissue were homogenized in the cold, mixed with an equal volume of dogfish Ringer solution (7), and this mixture was centrifuged at 500 or 12,000*g*. The resulting supernatant fluid was frozen until injected into the caudal vessel of a recipient fish.

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