Antigenic Similarity of Human α_1-Antitrypsin to a Corresponding Protein in the Serum of Non-Human Primates

Serum of many species, including man, contains glycoproteins capable of inhibiting the proteolytic activity of trypsin and other proteinases. In man the protein which is responsible for more than 90% of the trypsin-inhibiting capacity is known as α_1-antitrypsin, a glycoprotein that has been well characterized[1]. Serum of non-human primates also has the capacity to inhibit the activity of trypsin. I have tested several species of primates and all have antitryptic activity similar to humans. The antitryptic activity of serum from chimpanzees was found to be 1.68 $\pm$ 0.53 mg of trypsin inhibited by 1 ml of serum (based on 19 individual serum samples) and that of Rhesus monkeys was slightly higher namely 1.8 $\pm$ 0.35 mg (based on 32 individual serum samples). Other macaques that were tested have serum antitrypsin levels within the same range. The mean serum antitryptic activity for humans was found earlier to be 1.49 $\pm$ 0.38 mg of trypsin inhibited by 1 ml of serum[2]. As in human serum, most of the trypsin-inhibiting activity of serum from these primates is found in the electrophoretic α_1-region. Therefore, I tested serum of non-human primates for a protein similar to the human α_1-antitrypsin.

The work of GOODMAN[3], HAFLEIGH and WILLIAMS[4], and SARICH and WILSON[5] shows that immunologic technics can successfully be used to measure the similarity of serum proteins of related species. A useful method for this approach is precipitation inhibition employing a radioactively labeled antigen as described by HERZENBERG et al.[6]. This method requires only small amounts of antigen and specific antiserum. The degree of cross-reactivity of a given serum is expressed in % inhibition of the precipitation reaction. This is determined by the amount of radioactively labeled antigen remaining in the supernate after the antigen-antibody precipitate is centrifuged to the bottom of the test-tube. The autologous antigen, in this case human α_1-antitrypsin, inhibits the precipitation reaction completely (=100% crossreactivity). The mean standard deviation for all primate serums combined was 5%. The procedure for the isolation of human α_1-antitrypsin from pooled plasma and the production of antiserum in rabbits have been described[7]. The method of GREENWOOD et al.[8] was used for labeling α_1-antitrypsin with ^{125}iodine (New England Nuclear Corp.). The antiserum was used in a concentration of $^1/_{100}$ and the radioactively labeled α_1-antitrypsin in a concentration of 0.10 mg/ml.

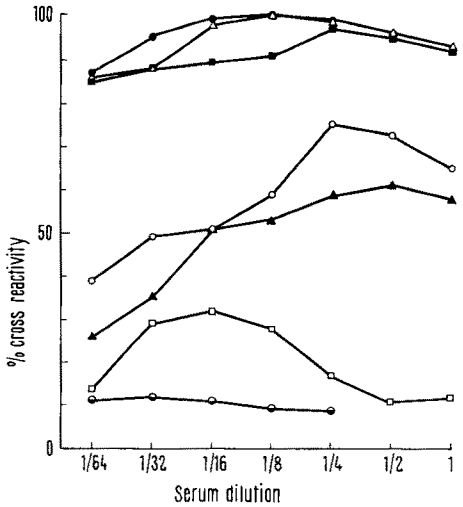
Several human serums were tested in this system and all were found to completely inhibit the precipitation at dilutions from $^1/_8$–$^1/_{200}$. All serums from primates were tested in dilutions from 1–$^1/_{64}$. The values given in the Table are the largest obtainable within this range of dilutions.

In preliminary experiments a few serums from primates were also tested in higher dilutions, but they never yielded a greater degree of crossreactivity than in the lower range of dilutions. The Figure shows the degrees of inhibition of some of the serums at various concentrations. Between 2 and 5 serum samples from different animals of each species were tested except in *Galago crassicaudatus*, *Lagothrix lagotricha* and *Cebus capucinus* from which only 1 serum sample was available. The results are in agreement with the immunologic studies of GOODMAN[3] and WANG et al.[9] for several serum proteins and those of SARICH and WILSON for albumin[5]. Like serum albumin, α_1-antitrypsin of man is most closely related to that of the Pongidae. Intermediate in closeness of relationship is that of the Cercopithecoidea, then that of Ceboidea, and most dis-

% crossreactivity of several primate serums with a rabbit antiserum to human α_1-antitrypsin

Hominoidea	*Gorilla gorilla*	100
	Pan troglodytes	100
	Pongo pygmaeus	100
	Hylobates lar	97
Cercopithecoidea	*Macaca speciosa*	76
	Macaca cynomolgus	68
	Macaca nemestrina	63
	Macaca mulatta	61
	Cercopithecus aethiops	74.5
	Cercocebus fulginosus	76
	Papio papio	75
Ceboidea	*Cebus capucinus*	32
	Lagothrix lagotricha	21
Prosimii	*Galago crassicaudatus*	12

Mean standard deviation: 5%.



% of crossreactivity at different dilutions of serum. ● Chimpanzee; △ Orangutan; ■ Gibbon; ○ Baboon; ▲ *Macaca mulatta;* □ *Cebus capucinus;* ◒ Galago.

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similar is that of Galago, the only representative of the Prosimii that was tested. The numerical values of crossreactivity of the Pongidae and Cercopithecoidea α_1-antitrypsin are close to those for transferrin[9] and albumin[5]. In Ceboidea and Prosimii the values of crossreactivity for albumin are consistently greater than those for α_1-antitrypsin indicating that albumin has behaved more conservatively during evolution than other serum proteins[3,10].

Zusammenfassung. Mit der Methode der Präzipitationshemmung nach HERZENBERG et al.[6] wurde die Kreuzreaktion von Seren subhumaner Primaten mit einem Antiserum gegen menschliches α_1-Antitrypsin gemessen. Die Ergebnisse stimmen mit der geltenden Taxonomie gut überein. α_1-Antitrypsin hat sich während der Evolution offenbar weniger konservativ verhalten als Serumalbumin.

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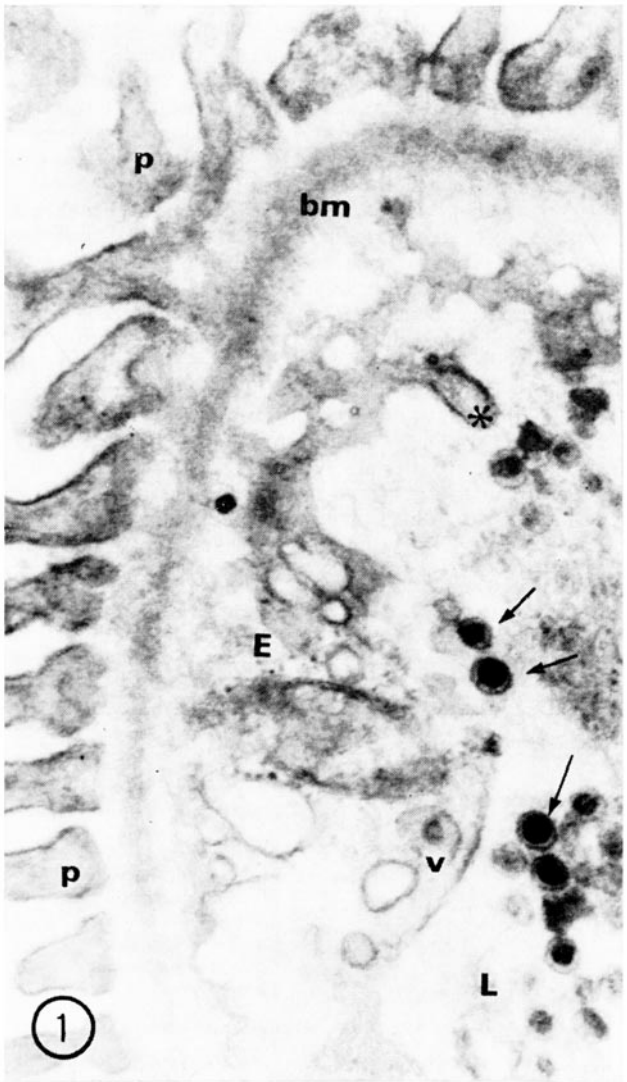
Changes in the Glomerular Endothelia Connected with Gross Virus Infection of Mice

Particles of GROSS virus are usually found both intracellularly and extracellularly in ultrathin sections of leukemic organs such as lymph nodes, spleen and thymus[1]. They have also been found in kidneys, but only in leukemic cells infiltrating this organ[2]. The aim of this paper is to present evidence that GROSS virus particles can also be found in connection with changes in the fine structure of mouse kidney endothelia without apparent presence of infiltrating leukemic cells.

In the present study, 3 mice of Charles River Swiss strain, infected with a filtrate of GROSS virus and with fully developed leukemia, were used. Pieces of kidney of experimental and control animals were immersed immediately after decapitation in cold 1.3% aqueous s-collidine buffered osmium tetroxide solution. The tissue was embedded in Epon 812. Ultrathin sections were stained with different lead salts and/or uranyl acetate.

Epithelial cells of renal tubules of all infected animals showed marked changes, consisting of a rearrangement of ultrastructural elements of mitochondria, a dilatation of endoplasmic reticulum and Golgi complex, and cytoplasmic vesiculation. Endothelial cells of the kidney glomeruli of infected animals were more irregular in form than control cells. In the cytoplasm of the endothelial cells many vesicles developed, the majority located at the luminal surface of the cytoplasm; some of them were open to the lumen of the glomerulus capillary, and inside some of the vesicles dense spherical particles were present. Particles of the same aspect but devoid of the limiting membrane were also found in the cytoplasm of the endo-



In the lumen of the capillary (L), many spherical particles apparently composed of a dense core with a single membrane are indicated by arrows. Magnification about × 60,000.

[1] L. DMOCHOWSKI, L. GROSS and F. PADGETT, Proc. Soc. exp. Biol. Med. *110*, 504 (1962).

[2] L. GROSS, in *Perspectives in Virology* (Ed. M. POLLARD; Hoeber Medic. Div., New York 1965), vol. 4, p. 226.

The endothelial cell (E) of the glomerulus of the infected mouse is separated by the basement membrane (bm) from the epithelial foot processes (p). Vacuoles containing dense particles (v) may be seen in the cytoplasm of the endothelial cell. Other dense particles, larger than ribosomes and devoid of any visible membrane, are also present in the same cell. A slender projection of the endothelial cell protruding into the lumen of the capillary is marked by an asterisk.