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Short Communication

COMPARISON OF ANTIGENICITY OF SERUM IMMUNOGLOBULIN G AMONG HUMAN, CYNOMOLGUS MONKEY, AFRICAN GREEN MONKEY AND SQUIRREL MONKEY

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SUMMARY: Antigenicity of IgG was compared among human, the cynomolgus monkey, the African green monkey and the squirrel monkey by the quantitative precipitation test using purified IgG of and rabbit anti-IgG serum to each species. Clear cross-antigenicity was observed between the cynomolgus monkey and the African green monkey and less clear cross-antigenicity between human and the cynomolgus monkey or the African green monkey. The cross-antigenicity observed between the squirrel monkey and the other three species examined was evidently weak.

Various species of nonhuman primates have widely been used as experimental animals for immunological studies. Because of the lack of specific antisera to nonhuman primates' immunoglobulins (Igs), however, most previous studies were carried out with antisera to human Igs. The cross-antigenicity between human and nonhuman primates' Igs should be examined in advance when we utilize antisera to human Igs for immunological studies using nonhuman primates.

In this short paper, we deal with the cross-antigenicity of IgG among human, the cynomolgus monkey (*Macaca fascicularis*), the African green monkey (*Cercopithecus aethiops*) and the squirrel monkey (*Saimiri sciureus*).

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Procedures for IgG purification and production of rabbit antisera to IgG were described elsewhere (2,3). Purity of IgG and monospecificity of the antisera were confirmed by immunoelectrophoresis. The quantitative precipitation test was carried out in small test tubes. One-tenth milliliter of antiserum was added to each of test tubes containing 15, 30, 60, 90, 120, 150 and 180 μ g of purified IgG. The final volume of the content in a test tube was adjusted to 1.0 ml with saline. The tubes were incubated for 1 hr at 37 C and then for 24 hr at 4 C . Precipitates formed were washed twice with cold saline, dissolved in 1.0 ml of 0.1 N NaOH, and determined for the protein content by the Folin-Ciocalteau's method (5). Supernatants were assayed for excess antigens or antibodies by the gel double diffusion test. The antibody content in 0.1 ml of antiserum was calculated by the following formula: (Weight of precipitate in μ g - Weight of added IgG in μ g), on the basis of the equivalence point where neither antigen nor antibody was detected in the supernatant. The degree of cross-antigenicity was expressed as the percentage assuming the amount of antibody precipitated in the homologous system to be 100%.

The quantitative precipitation reactions are illustrated in Fig. 1, involving different combinations of IgG and specific antisera as well as different volumes of IgG added. A maximum amount of antibody was precipitated in the combination of homologous antiserum and IgG. Average degree of cross-antigenicity was calculated from the results of five repeated tests. As shown in Table I, cross-antigenicity observed between IgGs of the cynomolgus monkey and the African green monkey averaged 82 - 90%. Those between human and the cynomolgus monkey or the African green monkey were 47 - 62%. Squirrel monkey IgG had cross-antigenicity of less than 31% with IgGs of human, the cynomolgus monkey and the African green monkey.

In most previous studies on the cross-antigenicity of a protein among human and nonhuman primates, investigators utilized specific antisera to the human proteins (4,6-8). Their experimental systems were able to recognize merely the antigenic sites of a nonhuman primate protein, which has the same antigenicity as that of human protein. In the present study, we prepared monospecific antisera not only to human IgG but also to nonhuman primate IgG so that we could determine the reciprocal antigenic relationship of IgG among human and three nonhuman primate species.

The present results have revealed that the cross-antigenicities of IgG detected by the use of antisera to human are nearly on the same level as that detected by the use of antisera to nonhuman primates. From these results, we

Fig. 1. Examples of the quantitative precipitation reaction: anti-IgG sera to human (a), cynomolgus monkey (b), African green monkey (c) and squirrel monkey (d) with purified IgG of human (Δ), cynomolgus monkey ($\bullet$), African green monkey ($\circ$) and squirrel monkey ($\blacktriangle$). Arrows indicate the equivalence point where neither antigen nor antibody was detected in supernatant.

Table I. Cross-antigenicity of IgG among human, cynomolgus monkey, African green monkey and squirrel monkey

Source of IgG	Cross reactivity to anti-IgG of			
	Human	Cynomolgus	Green	Squirrel
Human	100	47 (45-48)	47 (46-50)	28 (26-29)
Cynomolgus	62 (58-69)	100	82 (75-87)	31 (28-33)
Green	58 (53-67)	90 (88-91)	100	22 (20-26)
Squirrel	31 (27-32)	14 (13-16)	14 (14-15)	100 (%)

Average percentage of cross-antigenicity was calculated at equivalence point by (precipitated antibody with heterogeneous IgG/precipitated antibody with homologous IgG) $\times 100$.

considered that the total amount of antigenic sites existing on IgG of the three non-human primate species may be almost equal to that of human. In addition, the present result suggests also that the antigenicity of nonhuman primate IgG is partially identical with that of human IgG.

In short, the present study demonstrated that [1] clear cross-antigenicity of IgG was observed between the cynomolgus monkey and the African green monkey, both of which belong to the Old World monkeys, [2] weaker cross-antigenicity of IgG was observed between human and the cynomolgus monkey or the African green monkey, and [3] the squirrel monkey belonging to the New World monkeys showed a very low level of cross-antigenicity of IgG with human, the cynomolgus monkey and the African green monkey. These results on the cross-antigenicity of IgG among human and nonhuman primate species were in good agreement with their mutual taxonomic relationship that has generally been accepted (1).

REFERENCES

1. Dene, H., Goodman, M., Prychodko, W. and Moor, G. W. (1976): *Folia Primatol.*, 25, 35-61.
2. Fujimoto, K., Terao, K., Cho, F. and Honjo, S. (1983): *Japan. J. Med. Sci. Biol.*, 36, 171-176.
3. Fujimoto, K., Terao, K., Cho, F. and Honjo, S. (1986): *J. Med. Primatol.*, 15, 319-328.
4. Kagen, L. J. and Gurevich, R. (1967): *Immunology*, 13, 201-208.
5. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J. (1951): *J. Biol. Chem.*, 193, 265-275.
6. Monte-Wicher, V., Wicher, K. and Arbesman, C. E. (1970): *Immunochemistry*, 7, 839-849.
7. Safleigh, A. S. and Williams, C. A. (1966): *Science*, 151, 1530-1535.
8. Wang, A. C., Shuster, J., Epstein, A. and Fundenberg, H. H. (1968): *Biochem. Genet.*, 1, 347-358.