



Isoantigens of Gamma Globulin in Pigs

B. A. Rasmusen

Science, New Series, Volume 148, Issue 3678 (Jun. 25, 1965), 1742-1743.

Your use of the JSTOR database indicates your acceptance of JSTOR's Terms and Conditions of Use. A copy of JSTOR's Terms and Conditions of Use is available at <http://www.jstor.org/about/terms.html>, by contacting JSTOR at jstor-info@umich.edu, or by calling JSTOR at (888)388-3574, (734)998-9101 or (FAX) (734)998-9113. No part of a JSTOR transmission may be copied, downloaded, stored, further transmitted, transferred, distributed, altered, or otherwise used, in any form or by any means, except: (1) one stored electronic and one paper copy of any article solely for your personal, non-commercial use, or (2) with prior written permission of JSTOR and the publisher of the article or other text.

Each copy of any part of a JSTOR transmission must contain the same copyright notice that appears on the screen or printed page of such transmission.

Science is published by The American Association for the Advancement of Science. Please contact the publisher for further permissions regarding the use of this work. Publisher contact information may be obtained at <http://www.jstor.org/journals/aaas.html>.

Science

©1965 The American Association for the Advancement of Science

JSTOR and the JSTOR logo are trademarks of JSTOR, and are Registered in the U.S. Patent and Trademark Office. For more information on JSTOR contact jstor-info@umich.edu.

©2000 JSTOR

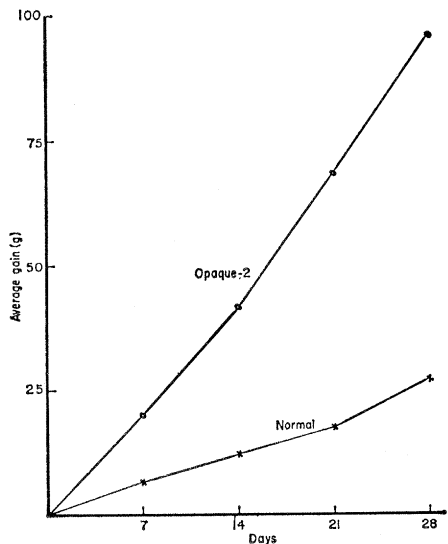


Fig. 1. Curves showing average weekly gains of rats fed on *opaque-2* maize and Indiana hybrid 453.

Fortification Mixture; diet D, the same as diet C except containing 20 percent soybean meal (8), no casein, and 65 percent corn starch.

The results of feeding the rats on

Table 2. Gains in weight, protein consumption, and protein efficiency ratios (28 days).

Animal No.	Gain (g)	Protein eaten (g)	PER*
<i>Diet containing casein</i>			
C-1	70	20.3	3.4
C-2	54	17.2	3.1
C-3	72	31.6†	2.3†
C-4	73	21.0	3.5
C-5	43	17.4‡	2.5‡
C-6	85	26.7	3.2
Average	66		3.1
<i>Diet containing opaque-2 maize</i>			
A-7	86	30.0	2.9
A-8	102	38.0†	2.7†
A-9	134	41.6	3.2
A-10	81	30.7	2.6
A-11	96	37.0	2.6
A-12	85	38.2†	2.2†
Average	97		2.8
<i>Diet containing Indiana hybrid 453</i>			
B-13	32	38.6†	0.8†
B-14	33	19.8	1.7
B-15	23	21.5†	1.1†
B-16	23	14.3	1.6
B-17	23	19.7†	1.2†
B-18	30	19.5	1.5
Average	27		1.6
<i>Diet containing soybean meal</i>			
D-19	67	35.6†	1.9†
D-20	95	34.0	2.8
D-21	95	36.0	2.6
D-22	101	37.2‡	2.7‡
D-23	78	28.7	2.7
D-24	105	37.3	2.8
Average	90		2.7

* Protein efficiency ratio (grams gained divided by grams protein eaten). The percentage of protein ($N \times 6.25$) in the diets was as follows: diet C, 8.1; diets A and B, 9.5; and diet D, 10.0. † Value not included in average because of food scattering. ‡ Rat scattered the food slightly, but value was included in average.

these diets are summarized in Table 2. In spite of special feed cups, scattering was a problem in all groups, with greatest incidence in the group fed on hybrid maize (diet B). Protein efficiency ratios (PER) were calculated for all animals, but the average PER values include only animals which did not scatter food, or scattered only slightly. Howe *et al.* (9) surveyed the PER values of nine unsupplemented, ground, whole cereal grains fed to rats on diets containing 7.8 to 10 percent protein, and found oats to have the highest value (1.8) when compared with casein (2.5). Barley, rice, maize, bulgar, rye, wheat, sorghum, and millet were below oats in PER. Our data indicate that in young rats *opaque-2* maize proteins have a food value equal to that of heat-treated soybean meal, and superior to any cultivated cereal grain.

Diets A and B were labeled with 1 percent metallic oxide, and the digestibility of the proteins was measured as described by Mertz *et al.* (10). White rats weighing approximately 100 g were used, four animals being fed on each diet. Protein digestibility values of 80, 83, 86, and 87 percent, respectively, were obtained with the rats on diet A (*opaque-2*) and values of 84, 84, 87, and 87 percent, respectively, were obtained with the rats on diet B (Indiana hybrid 453). Better digestibility of proteins therefore does not appear to be a factor contributing to the growth-promoting properties of *opaque-2* maize.

In a previous feeding test with the same number of rats (3), the average gain in weight for 28 days was 86 g for rats fed a different strain of *opaque-2* maize and 23 g for rats fed Indiana hybrid 257, a 3.7-fold difference. In the present test, a 3.6-fold difference was observed. Figure 1 shows average weekly gains of the animals on diets A and B. Similar curves were obtained in the previous feeding test.

The greater efficiency of *opaque-2* proteins in rats provides a basis for assuming that *opaque-2* proteins would also be superior to ordinary maize proteins in the diet of man and domestic animals. Loci homologous to *opaque-2* probably exist in other cereals, and methods for detection and use of mutants should be developed.

Note added in proof: Opaque kernels in high protein background gave endosperms containing 19.2 percent protein, of which 2.7 percent was lysine. Non-opaque kernels on the same ear yielded

endosperms with 19.6 percent protein, of which 1.3 percent was lysine. Thus, the opaque gene exerts its effect in a high protein background, and selection is feasible for lines that are high in both lysine and protein. Such lines would provide more lysine per gram of corn than diet A.

EDWIN T. MERTZ

OLIVIA A. VERON

LYNN S. BATES

Department of Biochemistry

OLIVER E. NELSON

Department of Botany and

Plant Pathology,

Purdue University, Lafayette, Indiana

References and Notes

1. E. T. Mertz, L. S. Bates, O. E. Nelson, *Science* **145**, 279 (1964).
2. E. T. Mertz, O. A. Veron, L. S. Bates, O. E. Nelson, unpublished data.
3. E. T. Mertz, O. A. Veron, O. E. Nelson, *Fed. Proc.* **24**, 629 (1965).
4. E. T. Mertz and R. Bressani, *Cereal Chem.* **34**, 63 (1957).
5. Nutritional Biochemicals Corp., Cleveland, Ohio.
6. General Biochemicals Corp., Chagrin Falls, Ohio.
7. National Casein Sales, Chicago, Ill. The casein contained 13 percent nitrogen.
8. Animal Nutrition Research Council representative meal supplied by P. H. Derse, Wisconsin Alumni Research Foundation, Madison. The meal contained 8 percent nitrogen.
9. E. E. Howe, G. R. Jansen, E. W. Gilfillan, *Am. J. Clin. Nutrition* **16**, 315 (1965).
10. E. T. Mertz, S. S. Rennert, E. W. Cole, *J. Nutrition* **56**, 437 (1955).
11. This is journal paper No. 2501, Purdue Agricultural Experiment Station, Lafayette, Indiana. Supported in part by a grant from the Corn Industries Research Foundation.

15 February 1965

Isoantigens of Gamma Globulin in Pigs

Abstract. Two gamma globulin isoantigens in pigs have been identified by hemagglutination-inhibition tests. Two codominant autosomal alleles, $G1^a$ and $G1^b$, determine three phenotypes, $G1(a+b-)$, $G1(a+b+)$, and $G1(a-b+)$.

Genetic differences in types of γ -globulins have been described for a number of mammalian species (1-3). The hemagglutination-inhibition system used to identify isoantigens of γ -globulins in pigs is modeled after that used for typing Gm and Inv, the hereditary antigenic determinants of γ -globulin in man (2).

Antibodies against the serum factor designated $G1_n$ were first produced by isoimmunization of pigs according to a procedure similar to that described for rabbits (3), but repeated immunizations were required to produce antibodies, and inhibition of agglutination was not easily interpreted. Wilson and Steinberg

(4) found a high frequency of antibodies to G_m and Inv in children aged 6 months to 5 years, and serums from young pigs may similarly have a high frequency of isoantibodies to γ -globulins without deliberate immunization. There were antibodies to G_{1_a} in 2 out of 20 serum samples from pigs which were approximately 4 months old, and in another there were antibodies to a complementary serum factor, designated G_{1_b} (5). One of the serums with antibodies to G_{1_a} and the single serum with antibodies to G_{1_b} were used as agglutinators in all of the tests reported here.

For the test for G_{1_a}, one volume of a 2 percent suspension of red cells (washed seven times) from a K_b-positive (6) pig of group O (7) was mixed with two volumes of an antiserum (diluted 1:16) which contained incomplete antibodies to K_b. Cells from a group-O pig were used since naturally occurring antibodies to pig O are rare (7) and would not be likely to interfere with the tests for G₁ factors. The mixture was incubated for 2½ hours at room temperature (24°C) to coat the red cells with antibody. The coated cells were then washed four times, and saline (0.91 percent NaCl) was added to make a 1-percent suspension. In each of two tubes one drop (1/30 ml) of the suspended red cells was added to a mixture of equal parts of antiserum to G_{1_a} (diluted 1:16) obtained from a pig, aged 4 months, and the diluted serum to be tested (1:4 and 1:8, respectively). The agglutinator and test serum were previously incubated at room temperature for 2 hours. The mixture of test serum, agglutinator, and coated cells was incubated at room temperature for 5 minutes, centrifuged gently (270g) for 1 minute, and examined macroscopically for agglutination. After an additional 2 hours incubation the tubes were again examined for agglutination, before and after centrifuging. The G₁(a+) test serums strongly inhibited agglutination of the coated cells by antibody to G_{1_a}; G₁(a-) serums inhibited weakly or not at all. For the test for G_{1_b}, the serum (diluted 1:8) obtained from a pig aged 4½ months was used as the agglutinator, and incomplete antibodies to L_c were used for coating the L_c-positive (6) red cells of the same group-O pig used as a source of cells in the test for G_{1_a}. Controls consisted of: known G₁(a+b-) and G₁(a-b+) serums tested with each agglutinator and appropriate coated cells; diluted test serum, saline, and coated cells; saline, agglutinator,

Table 1. Inheritance of G₁ types in pigs.

Mating types	Matings (No.)	Offspring		
		a+b-	a+b+	a-b+
a+b- × a+b+	6	20	22	0
a+b- × a-b+	2	0	14	0
a+b+ × a+b+	5	7	26	8
a+b+ × a-b+	4	0	14	20
a-b+ × a-b+	4	0	0	30

and coated cells; and saline plus coated cells. The red cells coated with incomplete antibodies to K_b or L_c can be agglutinated by the addition of rabbit antiserum to pig globulin as well as by the appropriate isoantibodies.

Porcine γ -globulins (Cohn Fraction II) and albumin (Cohn Fraction V) (8) were checked for freedom from other serum proteins by immunoelectrophoresis, and saline solutions of each fraction (20 mg/ml) were used in inhibition tests. There was no inhibition of agglutination by albumin, whereas the γ -globulin (diluted to 20 mg/ml) inhibited antibody to G_{1_a} up to a further dilution of 1:8, and inhibited antibody to G_{1_b} up to 1:4096. These data suggest that most of the pigs from which the pool for preparation of the γ -globulin was obtained were G₁(a-b+).

The distribution of G_{1_a} and G_{1_b} in serums from adult males and females (chosen to avoid including samples from pairs of full sibs) was: Duroc pigs, 2 G₁(a+b-), 10 G₁(a+b+), 2 G₁(a-b+); Yorkshire pigs, 1 G₁(a+b-), 3 G₁(a+b+), 12 G₁(a-b+).

No serums have been found to be G₁(a-b-), and it appears likely that allelic genes are responsible for factors G_{1_a} and G_{1_b}. The globulin types of 161 offspring, tested when 3 months old or older, from 21 matings are given in Table 1. These data are in accord with the hypothesis that G^{1_a} and G^{1_b} are codominant alleles, so that genotypes G^{1_a} G^{1_a}, G^{1_a} G^{1_b}, and G^{1_b} G^{1_b} determine phenotypes G₁(a+b-), G₁(a+b+), and G₁(a-b+). The existence of additional alleles, including G¹⁻ and G^{1^{ab}}, is not excluded by these limited data from two breeds of pigs.

Specific G₁ types are not associated with specific haptoglobin, transferrin, prealbumin or amylase (9) types, nor are they associated with specific red-cell antigens in the A-O, B, C, E, F, G, H, I, J, K, or L (6, 7, 10) systems. Results of segregation in offspring from matings of animals of known G₁ and

red-cell types have excluded sex linkage and close linkage between the G₁ locus and the A, B, C, E, F, G, H, J, K, and L loci controlling red-cell antigens.

B. A. RASMUSEN

Department of Animal Science,
University of Illinois, Urbana

References and Notes

1. R. Grubb and A.-B. Laurell, *Acta Pathol. Microbiol. Scand.* **39**, 390 (1956); A. Kelus and J. K. Moor-Jankowski, *Nature* **191**, 1405 (1961); S. Dray, *Proc. Intern. Congr. Genet. 11th Congress* **2**, 165 (1963).
2. A. G. Steinberg, in *Progress in Medical Genetics*, A. G. Steinberg and A. G. Bearn, Eds. (Grune and Stratton, New York, 1962), vol. 2, pp. 1-33.
3. S. Dubiski, Z. Dudziak, D. Skalba, *Immunology* **2**, 84 (1959).
4. J. A. Wilson and A. G. Steinberg, *Transfusion*, in press.
5. I have chosen terminology and notation which conforms to that proposed by Andresen (6) for describing blood groups in pigs. The γ -globulin system is symbolized by "G₁." The factors in this system are symbolized by "G¹" followed by a lower-case letter as a subscript: G_{1_a} and G_{1_b}. The antigens (G_{1_a} and G_{1_b}) are symbolized in the same way as the factors since each of these antigens has only one known factor; if new antigens or factors are discovered, the designations for the antigens can be expanded accordingly (for example, antigen G_{1_{ab}} would have both factors G_{1_a} and G_{1_b}). Symbols for alleles are italicized and superscripts are used instead of subscripts: G^{1_a} and G^{1_b}. Phenotypes are designated to reveal the results of serum typing; for example, a serum that is positive for G_{1_a} and negative for G_{1_b} has the phenotype G₁(a+b-).
6. E. Andresen, *Ann. N.Y. Acad. Sci.* **97**, 205 (1962); ———, *A Study of Blood Groups of the Pig* (Munksgaard, Copenhagen, 1963). K_b and L_c are antigenic factors of red cells of pigs.
7. B. A. Rasmusen, *Genetics* **50**, 191 (1964).
8. Porcine blood fractions obtained from Nutritional Biochemicals Corporation, prepared using the procedure of E. J. Cohn, L. E. Strong, W. L. Hughes, Jr., D. J. Mulford, J. N. Ashworth, M. Melin, H. L. Taylor, *J. Amer. Chem. Soc.* **68**, 459 (1946).
9. G. C. Ashton, *Nature* **186**, 991 (1960); F. K. Kristjansson, *Genetics* **48**, 1059 (1963).
10. E. Andresen and L. N. Baker, *ibid.* **49**, 371 (1964).
11. Supported in part by PHS grant GM 08752. Cecelia Szurszewski provided technical assistance.

13 May 1965

Degeneration of the Eyes of

Tyrosine-Deficient Chick Embryos

Abstract. Subjecting 4-day-old chick embryos to a yolk-sac perfusion medium lacking tyrosine resulted in arrest of retinal pigmentation and in degeneration of the neural retina. Phenylalanine was ineffective in replacing tyrosine. Possibly retinal tyrosinase played a part in initiating the degenerative changes.

The first successful study of amino acid deficiencies in chick embryos was reported by Klein, *et al.* (1), who applied the explantation techniques of Spratt (2) and of Hayashi and Herrmann (3) in studies with defined