Genet. Res., Camb. (1985). **47**, *pp.* 53–57 *With 6 text-figures Printed in Great Britain*

Screening of guinea pig strains for electrophoretic isoenzyme polymorphisms

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(*Received 26 February 1985 and in revised form 13 August 1985*)

Summary

Seven strains of guinea pig (*Cavia porcellus*) with different degrees of inbreeding were investigated for genetic variability by starch gel electrophoresis. Variants for six out of 31 loci (*Gpd-1*, *Aat-1*, *Ak-2*, *Pgm-1*, *Ada* and *Gpi*) are reported. The data suggest that the strains all come from a small founding population and therefore are genetically more homogenous than the visible characteristics imply. Differences between isoenzyme data and other genetic characters are described and possible underlying genetic mechanisms are discussed.

1. Introduction

Guinea pigs (*Cavia porcellus*) have been the subject for biological research for more than 80 years. Until 1960 this species was of main interest for geneticists because of its large variability in coat colour and texture. Outstanding among these studies are those of Sewell Wright and co-workers (see Searle, 1968; Wright, 1963; for reviews) contributing fundamentally to genetic theory, genic interaction and to the theory of inbreeding.

No systematic study of biochemical genetic variation has been made, in contrast with the amount of data on the formal genetics of more than 100 biochemical loci in various laboratory strains of *Mus musculus* (Staats, 1980). Isoenzyme studies in defined guinea pig strains have been restricted to a limited number of loci such as carbonic anhydrase, phosphoglucose isomerase, phosphogluconate dehydrogenase, glucose-6-phosphate dehydrogenase and phosphoglucomutase (Carter, 1972; Carter & Festing, 1972; Carter, Hill & Weir, 1972). Other studies have been carried out with animals of unknown or undescribed origin (e.g. peptidases, Frick 1983). Some well-documented biochemical peculiarities of the guinea pig have been reviewed recently (Wriston, 1981, 1984). Although excellent overall reviews are available (Wagner & Manning, 1976; Altman & Katz, 1979) data on biochemical genetic variation of this species are lacking. Therefore a number of gene products was studied by electrophoretic techniques in several strains of *Cavia porcellus*. In this way data will become available for characterization of the most common laboratory strains.

2. Material and Methods

The inbred strains 2 and 13 are bred in our animal breeding station. Specimens of the strain 'C4-deficient' were a gift of Dr Tabeiner, I. Department of Dermatology, Vienna. 'Dunkin–Hartley' animals were obtained from Dr Adamiker, Institute of Animal Breeding, Himberg. The strains VI-G, VI-B and VI-W are maintained by Dr Majdic, Institute of Immunology, Vienna. They descended from a single pair of random bred guinea pigs, and were inbred by consecutive brother–sister mating for eight generations, selected only for coat colour. Since the fifth generation, the offspring have bred true for this character. While VI-G retains the gray colour of the parents, VI-B is totally black both on back, belly and eyes. VI-W shows white colour with black nose, black feet and ear-tips. From each strain three to eight animals were used. Preparations of samples, starch gel electrophoresis and enzyme staining were performed according to standard methods (Csaikl, 1984; Csaikl, Engel & Schmidtke, 1980).

Enzyme names and abbreviations are according to Harris & Hopkinson (1976) or according to the original description; locus and allele designations follow those for mice (Lyon, 1977 and Staats, 1980).

3. Results

Gene products of 19 enzymes and one unspecific protein, the albumin, have been investigated in seven guinea pig breeding groups with different degrees of inbreeding and from different sources. The different enzymes, their E.C.-numbers and the various loci

Table 1. *List of enzymes, E.C. numbers, loci and alleles in the various guinea pig strains studied.*

EC-number	Enzymes	Loci	Alleles found in strain						
			2	13	C4	DH	VI-W	VI-G	VI-B
1.1.1.1	ADH	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.8	GPD	*1*	*a*	*a*	*a*	*a*	*a, b*	*a, b*	*a, b*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.14	SDH	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.27	LDH	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.37	NAD-MDH	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.40	NADP-MDH	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.42	IDH	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.44	6-GPD	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.1.1.49	G-6-PD	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
1.15.1.1	SOD	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
2.4.2.1	NP	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
2.6.1.1	AAT	*1*	*a*	*a*	*b*	*a, b*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
2.7.3.2	CPK	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
2.7.4.3	AK	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a, b*	*a, b*	*a, b*	*a, b*	*a, b*
2.7.5.1	PGM	*1*	*a*	*a*	*a*	*a, b*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		3	*a*	*a*	*a*	*a*	*a*	*a*	*a*
3.5.1.14	ACY-1	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
3.5.4.4	ADA	*1*	*a*	*a*	*a, b*	*a, b*	*a*	*a*	*a*
4.1.2.13	ALD	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*
		2	*a*	*a*	*a*	*a*	*a*	*a*	*a*
5.3.1.9	GPI	*1*	*a*	*a*	*a, b*	*b*	*a*	*a*	*a*
					Unspecific proteins				
—	Alb	*1*	*a*	*a*	*a*	*a*	*a*	*a*	*a*

screened are listed in Table 1. For the albumin (Alb.) and the following 13 enzymes no polymorphism was detected in and between the strains under study: alcohol dehydrogenase (ADH), sorbitol dehydrogenase (SDH), lactate dehydrogenase (LDH, loci *1* and *2*), malate dehydrogenase (NAD–MDH, loci *1* and *2*), malic enzyme (ME or NADP-MDH, loci *1* and *2*), isocitrate dehydrogenase (IDH, loci *1* and *2*), 6-Phosphogluconate dehydrogenase (IDH, loci *1* and *2*), 6-Phosphogluconate dehydrogenase (6-GPD), glucose-6-phosphate dehydrogenase (G-6-PD), superoxide dismutase (SOD, loci *1* and *2*), purine nucleoside phosphorylase (PNP), creatine phosphokinase (CPK), aminoacylase-1 (ACY-1) and aldolase (ALD).

Genetic variability was observed for the following polymorphic systems. In the case of the glycerol-3-phosphate dehydrogenase (GPD, Fig. 1) also known as α-glycerophosphate dehydrogenase, two different loci designated Gpd-1 and Gpd-2 were detected. The anodic form was found to be heterozygous in the strains VI-G, VI-B and VI-W. The other strains were homozygous for the slow moving gene product. The second gene product was moving cathodally under the conditions used (Harris & Hopkinson 1976). This confirms in guinea pigs the results of Kömpf, Ritter & Schmitt, (1971, 1972) and Hopkinson, Peters & Harris (1974), who described two different genes in man and primate species. For this gene product all strains were homozygous. A third intermediate band found by these authors and interpreted as a molecular product of both gene products was not detected even if other buffer systems were used. This finding is in contradiction with results on laboratory mice where only one locus was described (Kozak, 1972, 1975). On the other hand a quite complicated banding pattern found in various fish species was interpreted as the product of three different independently segregating genes by Engel, Schmidtke & Wolf (1971). In the aspartate aminotransferase (AAT, Fig. 2), locus *1* was heterozygous in the 'Dunkin–Hartley'-strain. The 'C 4-deficient'-strain was homozygous for the slow moving gene product, while in the other strains only allele *Aat-1*^{a} coding for the fast moving gene product was present. Locus *2* was homozygous in all specimens, with variable activity. For the adenylate kinase (AK, Fig. 3) only the two strictly inbred strains 2 and 13 were homozygous for the two loci *Ak-1* and *Ak-2*. The other strains were heterozygous for the cathodally moving *Ak-2*. In the

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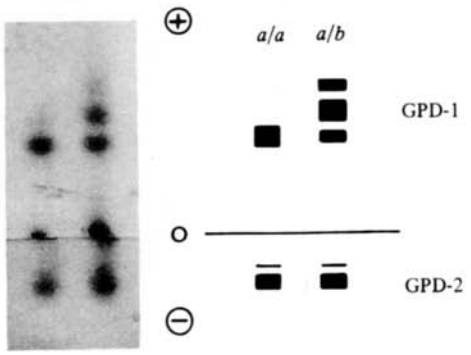

Fig. 1: GPD

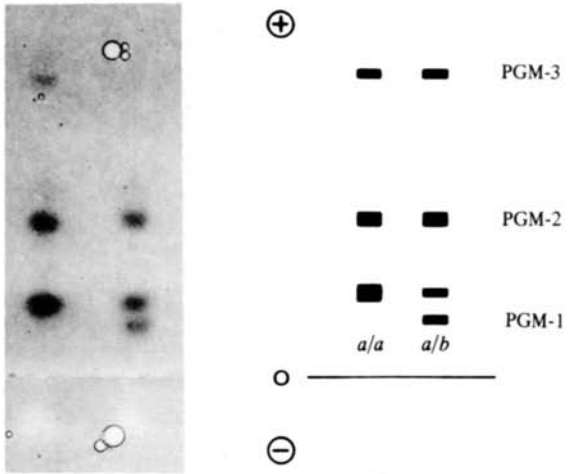

Fig. 4: PGM

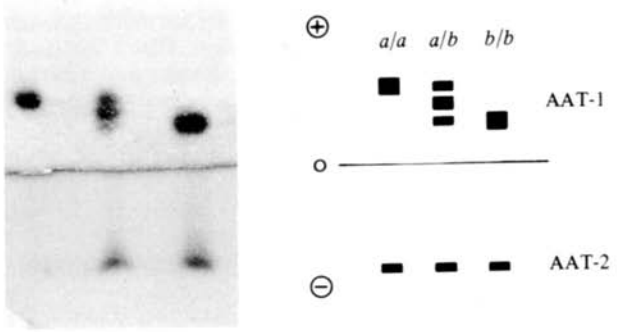

Fig. 2: AAT

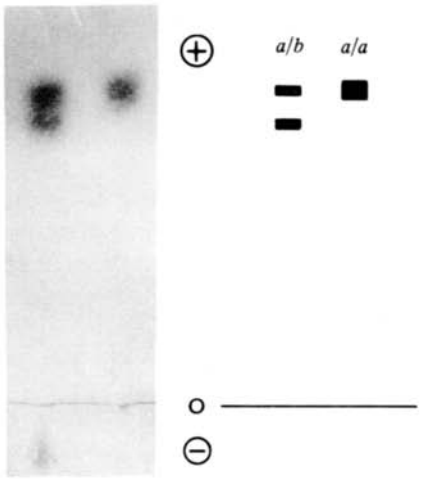

Fig. 5: ADA

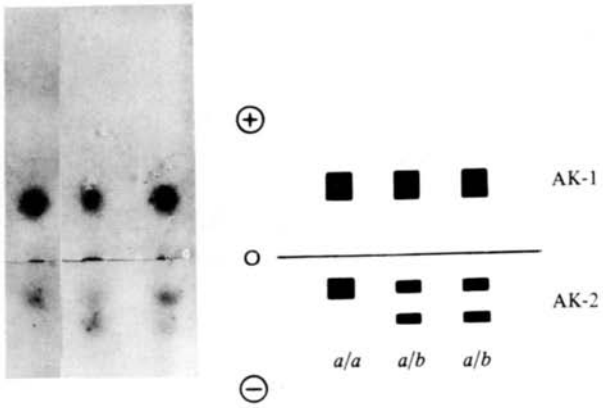

Fig. 3: AK

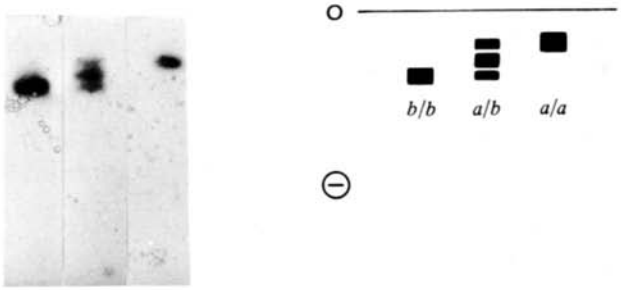

Fig. 6: GPI

Fig. 1–6. Electrophoretic and diagrammatic representation of glycerol-3-phosphate dehydrogenase (GPD), aspartate aminotransferase (AAT), adenylate kinase (AK), phosphoglucomutase (PGM), adenosine deaminase (ADA), and glucose phosphate isomerase (GPI).

case of phosphoglucomutase (PGM, Fig. 4) three loci could be detected, designated *1*, *2* and *3* according to the mobility of their gene products. *Pgm-1* segregated two different alleles, *a* and *b*, in the 'Dunkin–Hartley'-strain.

In the other strains this locus was homozygous. The other two loci were homozygous in all strains. This enzyme was also found to be polymorphic at this locus by Carter & Festing (1972). The partially inbred strains OM3 and R9 were homozygous for alternative alleles, while strain B was segregating for both alleles. For adenine deaminase (ADA, Fig. 5) a polymorphism was found both in the 'C 4-deficient' and the 'Dunkin–Hartley' strains. In the other strains only allele *Ada-1*^{b} segregating for the faster moving band was detected. In contrast to Carter & Festing (1972) two alleles could be detected in the guinea pig for the enzyme glucose phosphate isomerase (GPI, Fig. 6). Although this enzyme was named phosphoglucose-isomerase (PGI) by the former authors, we suggest

https://doi.org/10.1017/S0016672300024514 Published online by Cambridge University Press

the term GPI according to the nomenclature of mouse strains (Staats 1980). In strain 'C 4-deficient' the alleles *Gpi-1*^{a} and *Gpi-1*^{b} were present. The strain 'Dunkin–Hartley' was homozygous for allele *Gpi-1*^{b}, coding the faster moving band, while the other strains were homozygous for allele *Gpi-1*^{a}.

In four of the six polymorphic loci only one homozygous and the heterozygous phenotype were detected. Whether this is due to genetic mechanisms or is the result of sample error remains obscure. In the case of AAT and GPI both homozygous and the heterozygous phenotype were present in our material.

4. Discussion

In natural populations variation at the gene level results from spontaneous mutations followed by periodic bottlenecking, founder effects and intermittent random drift. Fixation of alleles can be caused additionally by selection. The more or less artificial strains of laboratory animals are established and maintained with the help of the same processes. Human error during the breeding process may serve as an additional factor. For studying such features starch gel electrophoresis of enzymes has proved to be of unique value (Ayala, 1976; Harris, 1980; Nei, 1975).

Studies on the biochemical–genetic variation in the guinea pig have been restricted to very few loci. This lack is in contrast to the sea of data about other laboratory animals such as mouse or rat where many genetic loci have been studied. Complementary data from wild populations of these two species have increased our knowledge of their genetic composition. In the house mouse many different alleles are fixed in various strains (see Staats, 1980, for review). The same situation is described in rat laboratory strains, although this species seems to be much more polymorphic than the mouse (Bender & Günther, 1977).

The results of the present study are in contrast with the situation found in these two species. The large number of alleles shared by the two well known and well defined guinea pig strains 2 and 13 is a quite astonishing result. These strains reveal strong differences in a number of genetic properties. For instance many antigens determined by genes linked to the complex of guinea pig leukocyte antigens are dissimiliar in the two strains; and they also differ in susceptibility to various immunological diseases (see Bacon *et al.* 1979, for review). Even the chromosomes show differences in the heterochromatic regions between strain 2 and the other strains studied (Whang-Peng, Lee & Hubbell, 1979). Both strains differ completely from the 'C 4-deficient' strain in only one enzyme, the AAT-1, and from the 'Dunkin–Hartley' strain in another enzyme, the GPI. Other pairwise comparisons of the strains show differences in one allele with the enzymes GPD-1, AAT-1, AK-2, PGM-1, ADA and GPI, while all other enzymes are homozygous for the same allele. In spite of different origin and different breeding history all strains under study seem to belong to one homogeneous gene pool. This homogeneity at the isoenzyme level contrasts with the situation for other characters in the guinea pig (Wagner & Manning, 1976; Altman & Katz, 1979, for review), and needs some further discussion. In our view the principles of genetic differentiation as cited above are not sufficient for an explanation.

I am indebted to Mr G. Reimer for his excellent technical assistance and to Mr P. Breit for his dynamic help with photographical work. The editorial help of Miss M. Sachs and the useful and stimulating discussion of my wife are acknowledged.

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