CONSERVED LINKAGE WITHIN A 4-cM REGION OF MOUSE CHROMOSOME *9* AND HUMAN CHROMOSOME *11*

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ABSTRACT

A six-point cross was carried out to determine the gene order and distances among loci on mouse chromosome *9*. Our results are consistent with the following arrangement: centromere – *Lap-1* – (1.2 ± 0.8) – *Es-17* – (3.0 ± 1.0) – *Ups* – (1.3 ± 0.7) – *Alp-1* – (23.1 ± 3.4) – *Mod-1* – (10.9 ± 2.6) – *Acy-1*. This study provides the first estimate of the distances between *Es-17*, *Ups* and *Alp-1*. Exceptions to the preferred association of alleles of *Es-17* and *Ups* have been found in three feral populations and one inbred strain. Evidence is presented for the homology of this chromosome region with the *ESA4* – *UPS* – *APO-AI* region on the long arm of human chromosome *11*.

ELECTROPHORETIC variants of two mouse enzymes, uroporphyrinogen I synthase (UPS) (EC 4.3.1.8)[1] and esterase 17 (ES-17) (EC 3.1.1.6), were recently mapped to the proximal region of mouse chromosome *9* (ANTONUCCI, CHAPMAN and MEISLER 1982; OTTO and VON DEIMLING 1983). Eight inbred strains were found to carry the common allele at both loci, whereas a single strain, LP/J, has a basic variant of both enzymes, suggesting a degree of linkage disequilibrium among inbred strains. The isoelectric points of the major isomers of UPS and ES-17 were nearly identical (MEISLER and CARTER 1980; OTTO and VON DEIMLING 1983).

The reported similarities in isoelectric points, chromosomal linkage and strain distribution were consistent with the formal possibility that the two enzyme activities might be encoded by a single gene. The present study was undertaken to test that possibility. In addition to direct measurement of the distance between *Ups* and *Es-17*, we have determined the gene order of six linked markers on chromosome *9*. The analysis was facilitated by the genotype of strain LP/J, which in addition to the rare variants of *Es-17* and *Ups* also carries variant alleles for four other loci previously mapped to this chromosome. Evidence that this linkage group is conserved in the human genome is also presented.

[1] In the past this enzyme has also been referred to as porphobilinogen deaminase (PBGD). It has recently been renamed hydroxymethylbilane synthase by the Nomenclature Committee of the International Union of Biochemistry (Eur. J. Biochem. **125:** 1–13, 1982).

MATERIALS AND METHODS

Animals: Mice of strains LP/J, C57BL/6J, DBA/2J, and 129/SvJ were purchased from The Jackson Laboratory, Bar Harbor, Maine. YBR/Ki mice were purchased from the Kirschbaum Memorial Mouse Colony in Rootstown, Ohio, and are maintained at the University of Michigan. Samples from the following strains were provided by J. HILGERS, Netherlands Cancer Institute, Amsterdam: A2G/A, ACR/A, BIMA/A, BIR/A, B6/ByA, B10/ScSnA, C57BL/LiA, DD/HeAf, FVB/NA, GRS-Mtv-2$^-$/A, GRS/A, LTS/A, LIS/A, MAS/A, MOL3/JA, NFS/NA, O1R/A, SL/NiA, STS/A, TSI/A, WLL/BrAf, 129/MA and 129/SVSLA. In addition, individuals from laboratory stocks of *M. m. musculus* (Belgrade), *M. m. musculus* (Brno), *M. m. musculus* (Skive), *M. m. castaneous*, *M. m. molossinus*, PAC/Cv and MOR/Cv were obtained from VERNE CHAPMAN, Roswell Park Memorial Institute.

Male LP/J mice were mated with female C57BL/6J mice to produce an F_1 generation. The F_1 animals of both sexes were backcrossed to LP/J and to C57BL/6J in reciprocal matings. One hundred and twenty-seven progeny from the backcross to C57BL/6J and 170 progeny from the backcross to LP/J were examined.

Isoelectric focusing of UPS: Whole blood was collected by cardiac puncture, and erythrocyte lysates were prepared as described (MEISLER and CARTER 1980). Lysates were treated with hydroxylamine to dissociate enzyme-substrate intermediates and thereby simplify the isozyme patterns (Meisler *et al.* 1981). Immediately prior to isoelectric focusing, 90 μl of each hemolysate were mixed with 10 μl of 2 M NH_2OH, pH 8.0, and incubated for 30 min at 37°. Samples were then applied directly to PAG plates (pH 4.0–6.5) (LKB); after focusing, UPS activity was visualized by a fluorescent staining method. Focusing and staining protocols have been previously described (MEISLER and CARTER 1980).

Electrophoretic analysis of esterase-17 (ES-17): Kidneys were homogenized in 3 volumes of distilled H_2O. Homogenates were centrifuged at 12,000 × *g* for 10 min immediately before electrophoresis. Isoelectric focusing and staining were performed by the method of OTTO and VON DEIMLING (1983), using PAG plates (pH 4.0–6.5).

Electrophoretic analysis of leucine arylaminopeptidase-1 (LAP-1): Intestines were scraped clean and homogenized in 2 volumes of distilled H_2O. Homogenates were centrifuged at 12,000 × *g* for 10 min on the day of electrophoretic analysis. LAP-1 phenotypes were identified by loading 5 μl of intestine homogenate onto 7% acrylamide, 0.025% methylene-bisacrylamide gels (9 × 9 cm). Electrophoresis was carried out at 200 V for 40 min or until the major bromphenol blue dye front was close to the bottom of the gel. The running buffer and gel buffer were 0.025 M Tris, 0.19 M glycine, pH 8.5. LAP-1 activity was detected using the substrate L-leucyl-β-naphthylamide (WOMACK, LYNES and TAYLOR 1975). The LAP-1 phenotypes detected by this method are shown in Figure 1. The heterozygous AB phenotype can be reliably distinguished from LP/J (BB) but not from C57BL/6J (AA). For this reason, only the backcross to LP/J was typed for LAP-1.

Apolipoprotein A-1 (ALP-1) analysis by two-dimensional electrophoresis: ALP-1 phenotypes were determined by two-dimensional gel electrophoresis of the supernatant fraction of liver or kidney homogenates prepared as described for malate oxidoreductase (MOD-1) and ES-17, respectively. Aliquots of the tissue extracts were solubilized in 3 volumes of a solution containing 9 M urea, 2% NP-40, 2% β-mercaptoethanol and 2% ampholines, pH 3.5–10 (LKB). The first dimension gel was prepared, and electrophoresis was carried out as described by NEEL *et al.* 1984. The second dimension was performed on gradient gels in the presence of sodium dodecyl sulfate as previously described (ROSENBLUM *et al.* 1982). The pattern of soluble proteins visible in mouse kidney by staining with Coomassie brilliant blue or with silver (MERRIL *et al.* 1981) is presented in Figure 2. The predominant albumin and hemoglobin components are probably the result of contamination with blood. ALP-1 is more easily analyzed in kidney after Coomassie staining, since silver staining visualizes several minor proteins with similar mobility (Figures 2 and 3). This is not a problem in liver samples, which can be easily scored by staining with silver (Figure 3c) or Coomassie blue. ALP-1 is a major protein in serum and is readily detected on gels stained with Coomassie blue (Figure 3d) or with silver.

Electrophoretic analysis of MOD-1 and amino acylase (ACY-1): Livers were homogenized in an equal volume of distilled H_2O and centrifuged at 12,000 × *g* for 10 min. Supernatants were removed and stored at −80° until electrophoretic analysis.

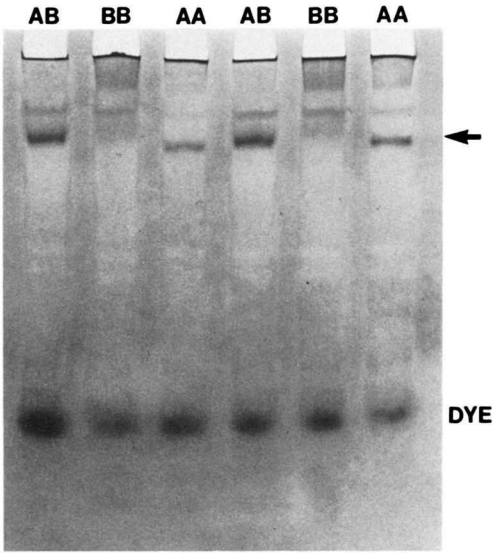

FIGURE 1.—Electrophoretic phenotypes for LAP-1 in parental strains and heterozygotes. Soluble extracts from intestine were analyzed by electrophoresis at pH 8.5. In the backcross to LP/J, offspring with the band indicated by the arrow were designated as AB; those without a discrete band at this position were designated BB. AA, C57BL/6J; BB, LP/J; AB, (C57BL/6J × LP/J)F_1.

MOD-1 isozymes were separated by isoelectric focusing on PAG plates, pH 3–10 (LKB), at 8°. Gels were prefocused for 15 min at 28 mA. Samples were applied to 5- × 10-mm filter paper applicators (18 μl/applicator) which were placed on the gel surface about 3 cm from the anodal wick. Focusing was at 28 mA until a potential of 2000 V was attained. The gels were then washed in 200 ml of 0.1 M Tris-HCl (pH 7.6) for 15 min and stained for MOD-1 enzyme activity in 100 ml of the staining solution of SHOWS, CHAPMAN and RUDDLE (1970).

ACY-1 phenotypes were determined by cellulose acetate electrophoresis of liver supernatants. Approximately 2 μl of each sample were applied to Titan III cellulose acetate plates (Helena Lab) which had been wetted in 50 mM Tris glycine buffer (pH 8.0) and blotted. Electrophoresis was at 300 V (1.5 mA/plate) in 50 mM Tris-glycine buffer for 30 min. ACY-1 activity was visualized in an agar overlay made by mixing an equal volume of 2% melted agarose solution (60%) with a staining solution (QAVI and KIT 1980) containing 2 mg/ml of *N*-acetyl methionine, 0.6 mg/ml of L-amino acid oxidase, 1 mg/ml of horseradish peroxidase and 1 mg/ml of *o*-dianisidine in 50 mM Tris-glycine buffer. Plates were incubated at 37° for 30 min or until enzyme activity appeared.

RESULTS

Comparison of isoelectric points of UPS and ES-17: The isoelectric points of the major isomers of UPS and ES-17 were compared directly by focusing samples of kidney homogenates and erythrocytes on the same gel. After focusing the gel was divided in two; the kidney samples were stained for ES-17 and the erythrocyte samples for UPS (Figure 4). The result confirms the previous reports of the similarity of isoelectric points of the major isozymes of the two enzymes. UPS B and ES-17B^2 focus at nearly identical positions on the PAG plate. The major isozymes of UPS A and ES-17A^2 are separated by 1 cm on these gels, with UPS A more acidic than ES-17A^2. Since the tissue isomers of UPS which predominate in kidney are more acidic than the eryth-

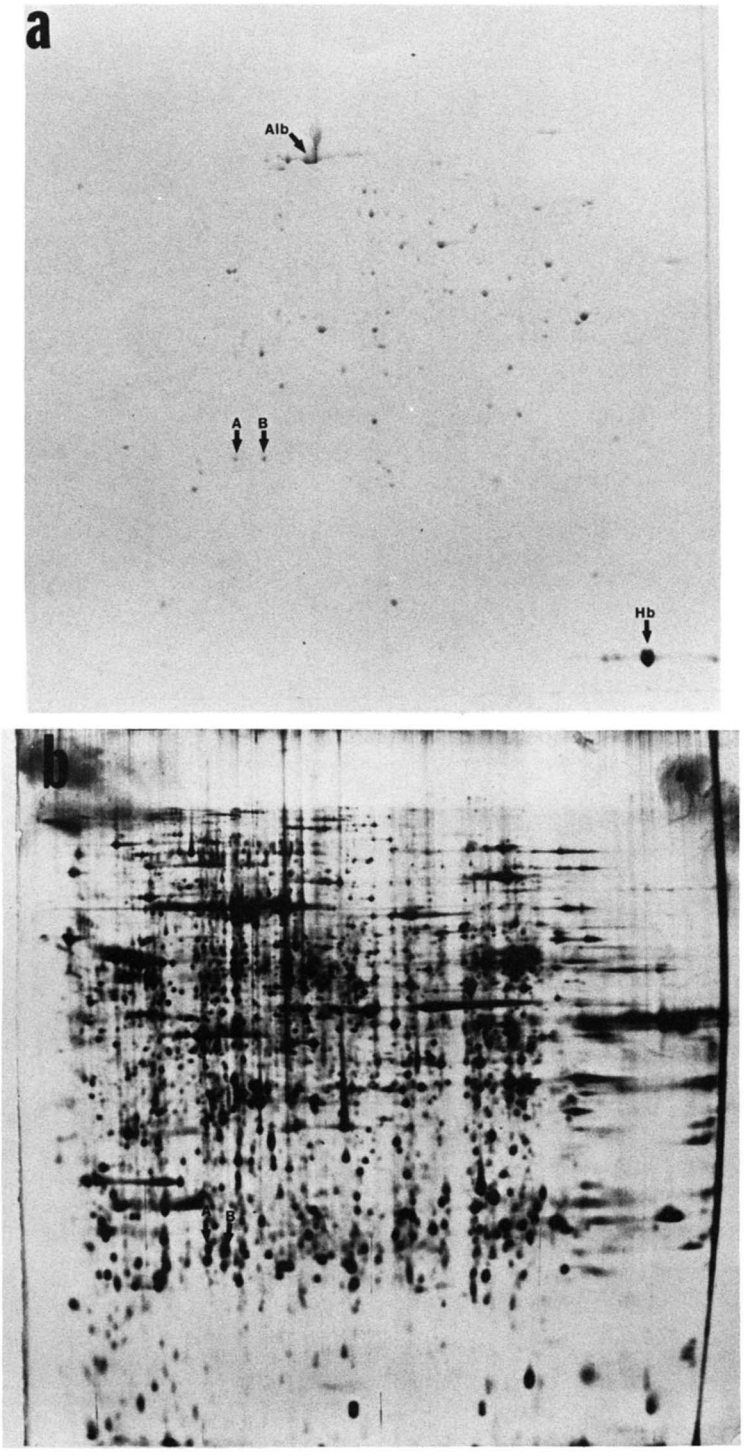

FIGURE 2.—ALP-1 phenotype of a heterozygous (C57BL/6J × LP/J)F_1 individual. Two-dimensional gel electrophoresis of soluble proteins from kidney was carried out as described in MATERIALS AND METHODS. The gel was first stained with Coomassie brilliant blue (A) followed by silver staining (B). The positions of albumin (Alb) and hemoglobin (Hb) polypeptides are indicated.

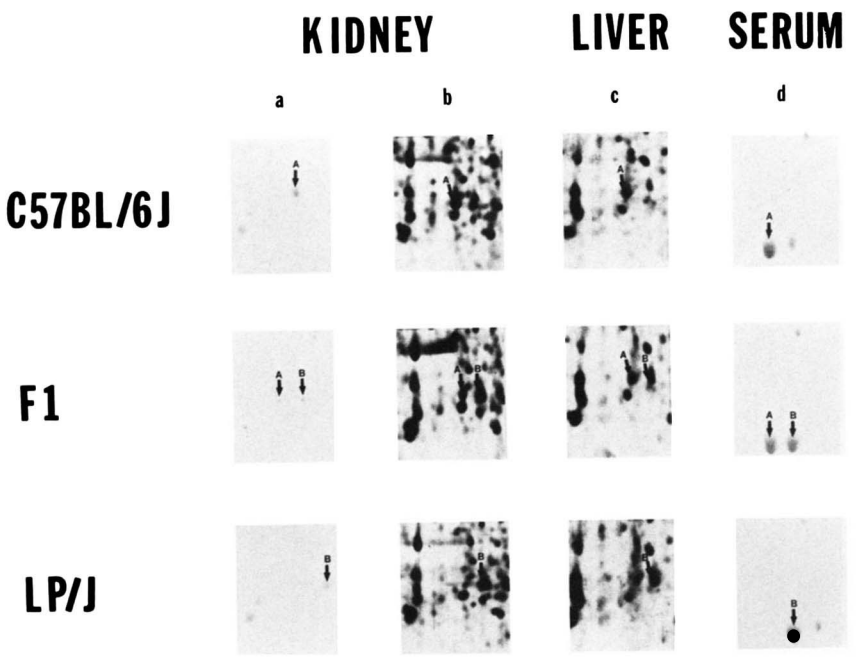

FIGURE 3.—ALP-1 phenotypes can be determined by two-dimensional electrophoresis of liver, kidney or serum. Samples were analyzed as described in MATERIALS AND METHODS. Gels were stained with Coomassie brilliant blue (a and d) or with silver (b and c). a and b, kidney; c, liver; d, serum.

rocyte isozymes shown here (MEISLER and CARTER 1980), it is clear that UPS and ES-17 can be physically separated by isoelectric focusing.

Genetic recombination between Ups and Es-17: UPS and ES-17 phenotypes of offspring from the backcross of (C57BL/6J × LP/J)F_1 animals to C57BL/6J and to LP/J were determined. Among 297 individuals tested, nine were recombinant for *Ups* and *Es-17* (Tables 1 and 2). The combined estimate of the distance between the two loci is 3.0 ± 1.0 cM. There was no significant difference in recombination frequency for male and female heterozygotes within this region.

Gene order of linked loci: Individuals in both backcross generations were typed for the linked marker *Alp-1* (Tables 1 and 2). In addition, individuals from the backcross to LP/J were typed for *Lap-1* (Table 2). Two recombinants between *Es-17* and *Lap-1* were found among 170 animals tested; both were also recombinant between *Ups* and *Lap-1*. Four recombinants between *Alp-1* and *Ups* were observed among 297 animals tested; these four individuals were not recombinant between *UPS* and the other two markers. The resultant gene order for the four loci is: *Lap-1* – *Es-17* – *Ups* – *Alp-1*. As predicted by the closeness of the linkages, no double recombinants were observed among these four loci.

To orient this linkage group with respect to the centromere, the 15 recombinant animals from the backcrosses were typed for *Mod-1*, an outside marker

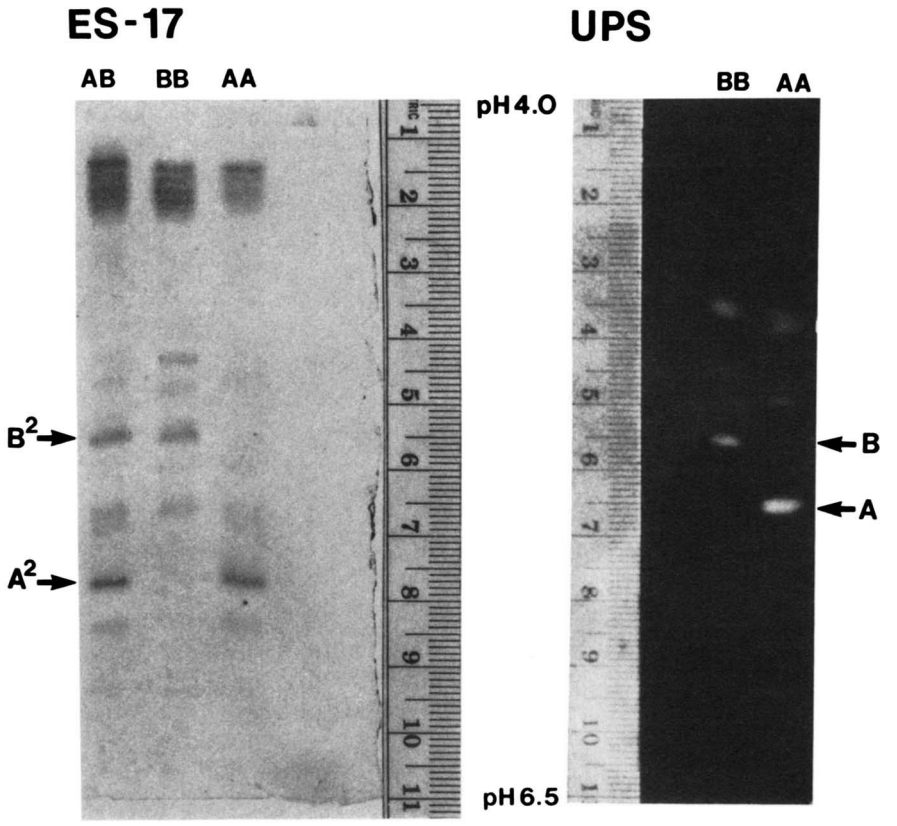

FIGURE 4.—Isoelectric points of UPS and ES-17 isomers. Samples of erythrocyte lysates and of kidney homogenates were applied to a single PAG plate, pH 4.0–6.5. After focusing, the gel was divided in two; the erythrocyte samples were stained for UPS and the kidney samples were stained for ES-17. Comparison with standards indicates that the isoelectric points of the major isozymes marked by arrows are within the range pH 5.3–5.65.

located approximately 30 map units distal to *Lap-1* (RODERICK and DAVISSON 1981). Of ten recombinant animals in the backcross to LP/J, nine were concordant for *Alp-1* and *Mod-1*, whereas one was a double recombinant (Table 3). Thus, *Alp-1* is closer to *Mod-1* than is *Lap-1*, and the gene order is: centromere – *Lap-1* – *Es-17* – *Ups* – *Alp-1* – *Mod-1*. The five recombinants from the backcross to C57BL/6J were consistent with this gene order (Table 3).

Recombination with distal markers: To better define the region distal to *Alp-1*, animals from both backcrosses were also typed for *Mod-1* and *Acy-1*. Recombination frequencies for the interval between *Alp-1* and *Acy-1* are presented in Table 4. The observed frequencies of recombination confirm the gene order *Alp-1* – *Mod-1* – *Acy-1* and are generally consistent with previous estimates of these distances (WOMACK, LYNES and TAYLOR 1975; WOMACK, TAYLOR and BARTON 1978; DOUGLAS and DAWSON 1979; LANE and WOMACK 1979; EICHER *et al.* 1980; NAYLOR *et al.* 1982).

TABLE 1

Segregation of alleles of Ups, Es-17 *and* Alp-1 *in the backcross to C57BL/6J*

Class	Genotype	No.	Total	% recombinants
Parental	$\frac{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}$	57	122	
	$\frac{Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}$	65		
Single crossover *Es-17 – Ups*	$\frac{Es\text{-}17^b\ Ups^a\ Alp\text{-}1^a}{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}$	2	3	2.4
	$\frac{Es\text{-}17^a\ Ups^b\ Alp\text{-}1^b}{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}$	1		
Single crossover *Ups – Alp-1*	$\frac{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^b}{Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}$	2	2	1.6

Frequency of recombination ± SE: *Es-17 – Ups*, 3/127 = 0.024 ± 0.014; *Ups – Alp-1*, 2/127 = 0.016 ± 0.011; *Es-17 – Alp-1*, 5/127 = 0.039 ± 0.017.

TABLE 2

Segregation of alleles of Ups, Es-17, Lap-1 *and* Alp-1 *in the backcross to LP/J*

Class	Genotype	No.	Total	% recombinants
Parental	$\frac{Lap\text{-}1^a\ Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	84	160	
	$\frac{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	76		
Single crossover *Lap-1 – Es-17*	$\frac{Lap\text{-}1^a\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	2	2	1.2
	$\frac{Lap\text{-}1^b\ Es\text{-}17^a\ Ups^a\ Alp\text{-}1^a}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	0		
Single crossover *Es-17 – Ups*	$\frac{Lap\text{-}1^a\ Es\text{-}17^a\ Ups^b\ Alp\text{-}1^b}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	3	6	3.5
	$\frac{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^a\ Alp\text{-}1^a}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	3		
Single crossover *Ups – Alp-1*	$\frac{Lap\text{-}1^a\ Es\text{-}17^a\ Ups^a\ Alp\text{-}1^b}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	1	2	1.2
	$\frac{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^a}{Lap\text{-}1^b\ Es\text{-}17^b\ Ups^b\ Alp\text{-}1^b}$	1		

Frequency of recombination ± SE: *Lap-1 – Es-17*, 2/170 = 0.012 ± 0.008; *Es-17 – Ups*, 6/170 = 0.035 ± 0.014; *Ups – Alp-1*, 2/170 = 0.012 ± 0.008; *Lap-1 – Ups*, 8/170 = 0.047 ± 0.016; *Es-17 – Alp-1*, 8/170 = 0.047 ± 0.016; *Lap-1 – Alp-1*, 10/170 = 0.059 ± 0.017.

TABLE 3

Genotypes of recombinant animals from the two backcrosses at five loci

	Lap-1		*Es-17*		*Ups*		*Alp-1*		*Mod-1*	No. of individuals
C57BL/6J	a		a		a		a		b	
LP/J	b		b		b		b		a	
Backcross to LP/J	a	*	b		b		b		a	2
	b		b	*	a		a		b	3
	a		a	*	b		b		a	3
	b		b		b	*	a		b	1
	a		a		a	*	b	*	b	1
Backcross to C57BL/6J			b	*	a		a		b	2
			a	*	b		b		a	1
			a		a	*	b		a	2

The recombinant individuals from the two backcrosses analyzed in Tables 1 and 2 were tested for the outside marker *Mod-1*. The most likely gene order is indicated from left to right; the positions of recombinations are marked with asterisks.

TABLE 4

Recombination frequencies between Alp-1 *and distal loci* Mod-1 *and* Acy-1

	Sex of F_1 parent		
	Female	Male	Total
Backcross offspring	178	95	273
Parental genotypes	114	66	180
Recombinants			
Alp-1 – Mod-1	41	22	63
Mod-1 – Acy-1	23	7	30
Double recombinants	3	1	4

Data from the backcrosses to LP/J and C57BL/6J are pooled.
Frequency of recombination ± SE: *Alp-1 – Mod-1*, 63/273 = 0.231 ± 0.034; *Mod-1 – Acy-1*, 30/273 = 0.109 ± 0.026; *Alp-1 – Acy-1*, 93/273 = 0.340 ± 0.028.

Strain distribution of alleles of Ups *and* Es-17: The genotypes of the following additional inbred strains have been determined. *Ups*^{a}: BIMA/A, BIO/ScSnA, B6/ByA, BIR/A, C57BL/LiA, DD/HeAf, GRS/A, GRS-Mtv-2$^-$/A, LTS/A, OIR/A and STS/A. *Ups*^{b}: 129/MA, 129/SvJ[2] and 129/SVSL. *Es-17*^{a}: B6/ByA, BIR/A, C57BL/LiA, GRS-Mtr-2/A and LTS. Both the *Ups* and *Es-17* alleles carried by more than 30 inbred strains have now been determined, with only one example of discordance from the a-a or b-b combination (Table 5). In contrast, two of the five feral populations are discordant. The a-a combination

[2] The allele carried by strain 129/SvJ was incorrectly reported to be *Ups*^{a} in an earlier publication (ANTONUCCI, CHAPMAN and MEISLER 1982).

TABLE 5

Distribution of Ups *and* Es-17 *alleles in inbred strains and feral mice*[a]

Es-17	*Ups*	Feral mice	Inbred strains
a	a	None	A2G/GA, ACR/A, BIMA/A, BIR/A, BIO/ScSnA, B6/ByA, DBA/2J, FVB/NA, LIS/A, MAS/A, MOR/Cv, NFS/A, OIR/A, PAC/Cv, SL/NiA, STS/A, TSI/A, WLL/BrAf, YBR/Ki, plus several substrains of A, AKR, BALB, C57BL, C3H, CBA, GRS and RIII
b	b	*M. m. musculus* (Belgrade) *M. m. musculus* (Brno) *M. m. musculus* (Skive)[b]	LP/J, MOL3/JA
a	b	None	129[c]
b	a	*M. m. castaneous* *M. m. molossinus*	None

[a] References: ANTONUCCI, CHAPMAN and MEISLER (1982); OTTO and VON DEIMLING (1983); MEISLER and CARTER (1980); and this publication.

[b] The *Ups*[a] allele is also present in this population with a gene frequency of 0.07 in 15 individuals.

[c] Sublines 129/SvJ, 129/MA and 129/SVSL.

which predominates in inbred strains was not observed in any of the feral mice tested.

DISCUSSION

It is evident from the analysis of the two backcrosses constructed for this study that *Ups* and *Es-17* are distinct and closely linked loci. Our results provide a detailed map of this chromosome region: centromere – *Lap-1* – (1.2 ± 0.8) – *Es-17* – (3.0 ± 1.0) – *Ups* – (1.3 ± 0.7) – *Alp-1* – (23.1 ± 3.4) – *Mod-1* – (10.9 ± 2.6) – *Acy-1*. This gene order is consistent with the previous ordering of *Lap-1*, *Alp-1* and *Mod-1* by LANE and WOMACK (1979); the distances between loci that we observe differ in some instances from previous estimates (for review see RODERICK and DAVISSON 1981).

Genetic variation at the *Alp-1* locus was first recognized by EICHER *et al.* (1980). Using the technique of one-dimensional electrophoresis of mouse serum on cellulose acetate, EICHER *et al.* mapped an electrophoretic variant to chromosome *9* and the locus was designated *Sep-1*. ELLIOTT (1979) independently identified variants of a liver protein by two-dimensional electrophoresis and mapped it to the same region of chromosome *9*, using the locus designation *Ltw-1*. Subsequent work by ELLIOTT and coworkers demonstrated that liver protein *LTW-1* is identical with the apolipoprotein associated with high density lipoproteins in serum (MILLER *et al.* 1983). The locus designation *Alp-1* was suggested by LUSIS and coworkers (1983) in a recent study of genetic variation at this locus using one-dimensional electrophoresis of proteins isolated from serum high density lipoproteins. We have utilized *Alp-1* because it is the

more descriptive designation. It would be desirable to reconcile the nomenclature of the mouse locus with that of the homologous human locus, which is currently designated *APO-AI*.

Among the 30 inbred strains that we have typed for *Ups* and *Es-17*, we have found only one strain in which the preferred association of alleles is not conserved. The nonrandom association of alleles of *Ups* and *Es-17* is consistent with other evidence that the standard inbred strains are derived from common progenitors. In contrast to the observations in inbred strains, no preferred association of alleles is evident in five populations of feral mice. The a-a allele combination characteristic of inbred strains is not represented among the feral mice tested.

In recent years there have been several reports of linkage homologies between the human and mouse genomes (LALLEY, MINNA and FRANKE 1978; PEARSON *et al.* 1979; LUNDIN 1979; DALTON *et al.* 1981). Conservation of 15 two- or three-locus regions are cited in recent reviews (WOMACK 1983; NADEAU and TAYLOR 1983). Human UPS has recently been mapped to chromosome region 11q23 → qter (MEISLER *et al.* 1981; WANG *et al.* 1981). It was, therefore, of interest to us that a locus encoding a human esterase, *ESA4*, has also been mapped to this region of human chromosome *11* (WANG *et al.* 1981) and might represent a homolog of mouse *ES-17*.

Esterase A4 was first described in 1962 (ECOBICHON and KALOW 1962). It is present in many human tissues and was first classified with the acrylesterases (EC 3.1.1.2) (SHOWS 1972) but is now considered to be an acetylesterase (EC 3.1.1.6). Evaluation of homology between nonspecific esterases of different species is complicated by the fact that the biological function of these enzymes is unknown and they can be identified only by their activity against nonphysiological substrates. The large number of distinct mammalian esterases is an additional complication (PETERS 1982; WOMACK 1983). Thus, in order to establish homology of esterases, as many criteria as possible should be taken into consideration. The known characteristics of human esterase A4 and mouse esterase 17 are compared in Table 6. Common properties between the two esterases include their broad tissue distribution, catalytic activity against acetates but not butyrates and resistance to inhibition by organophosphorous and mercuric compounds. These similarities strongly suggest that these two loci are indeed homologous. The close linkage of both acetylesterases to the gene encoding UPS indicates that this chromosome region has been conserved during evolution.

This conclusion is strongly supported by the recent assignment of the locus for human apolipoprotein A-I to chromosome *11* (CHEUNG *et al.* 1984; LAW *et al.* 1984). Mouse *Alp-1* and human *APO-AI* are clearly homologous, since the products of both loci are similar in physical characteristics and are major protein components of serum high density lipoprotein particles. Thus, a minimum of 4.3 map units between mouse *Es-17* and *Alp-1* appears to be conserved in the human genome. It seems likely that the gene order is also conserved. The possible location of the human equivalents of the mouse chromosome *9* loci *Lap-1* and *Thy-1* on chromosome *11* is also very likely; for

TABLE 6

Characteristics of human esterase A4 and mouse esterase 17 indicating homology

	Human ESA4[a]	Mouse ES-17[b]
Molecular wt	49,000	<50,000[c]
Substrate specificity		
α-Naphthyl acetate	+	+
Indoxyl acetate	+	+
5-Bromo indoxyl acetate	+	+
N-Methyl indoxyl acetate	+	+
α-Naphthyl butyrate	−	−
Indoxyl butyrate	?	−
Inhibitor sensitivity		
10^{-4} M $HgCl_2$	Resistant	Resistant
2×10^{-5} M E600[a]	Resistant	Resistant
2×10^{-2} M NaF	Slight	?
10^{-3} M eserine	Resistant	Resistant
Tissue distribution		
Moderate to high activity	Kidney, GI tract, liver, spleen, ovary, testis	Same
Low to moderate activity	Lung, adipose, brain, heart, skeletal muscle	Same
No activity	Erythrocytes	Same

[a] Data from COATES, MESTRINER and HOPKINSON (1975).
[b] Data from OTTO and VON DEIMLING (1983).
[c] VON DEIMLING (unpublished observation).

example, the probability that the human homolog of *Lap-1* is linked to *ESA4* can be estimated to be 0.91 using the method developed by NADEAU and TAYLOR (1983).

Mouse chromosome *9* carries four distinct gene regions that are found on four different human chromosomes (WOMACK 1983): *Mpi-1* on human chromosome *15*, the amino acylase – β-galactosidase region on human chromosome *3*, the *Mod-1* – *Pgm-3* region on human chromosome *6q* and the *Es-17* – *Ups* – *Alp-1* region on human chromosome *11q*. Such conservation of small chromosome regions appears to be characteristic of the genomes of mammalian species (NADEAU and TAYLOR 1984).

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