have used single allele lines[12,17]. Therefore much of the earlier data, particularly from population cages maintained at different temperatures and from comparative studies of the heat-resistancet properties of *Adh* variants, must now be viewed with caution.

There may well be further cryptic variation at the *Adh* locus which new techniques will reveal, although methods of sequential gel electrophoresis have not detected 'hidden' variation at this locus[18]. Nevertheless, it is clear that interpretation of the maintenance of the *Adh* polymorphism must now be in terms of a tri-allelic system[19].

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Inter-locus allozyme mobility correlations and species divergence

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Summary. An analysis of allozyme data from numerous sets of related vertebrate and *Drosophila* species shows that species divergence does not generally seem to be accompanied by an overall increase or decrease in enzyme charge. The 2 significant results came from vertebrate inter-generic comparisons.

The cellular environment of certain highly specialized tissues appears in several cases to favor the functioning of isozymes whose electrical charge and hence electrophoretic mobility is recognizably and generally different from the corresponding isoenzymes in other tissues. It has, for example, been pointed out that the creatine kinase and lactate dehydrogenase isozymes restricted to the neural tissue of advanced teleosts characteristically possess a high net negative charge[1], and this is also true of the fructose 16 biphosphate aldolase isozyme found in the eye and brain of many vertebrates (including fishes)[2]. It has further been suggested that the low net charge of serum albumin of the marine iguana *Amblyrhynchus cristatus* when compared with that of 2 species of land iguana may have been critical for successful reptilian adaptation to an aquatic environment[3], and that generally charge (and concentration) of albumin are important factors in controlling water loss in reptiles[4]. Thus in at least some instances adaptation of proteins to particular cellular or ecological environments may be accompanied by a change in charge.

If adaptation of a population to a new environment does favor a change in protein charge, then it is conceivable that there could be selection for a general charge increase (or decrease) in physiologically important enzymes. Thus in comparing related species which have diverged from a common ancestor and subsequently adapted to new environments, the average anodal mobility of proteins may differ significantly between species. That is, there may be mobility correlations over protein loci between species. This is the hypothesis tested here. It should be noted that the theory of neutral variation[5–7] would not predict such correlations.

Many studies have been published describing patterns of allozymic variation at appreciable numbers of loci in related species, and the majority of these studies employ an internally consistent notation indicating the relative mobilities of the screened allozymes. We have tested for correlations in allozyme mobility between related vertebrate species and related *Drosophila* species, using studies scoring a minimum of 15 loci and 15 individuals per locus. Most of the sources of data are given in an earlier study[8]. The relative mobility of the product of the most frequent allele in each species was used in the comparisons.

14 of the vertebrate surveys and 1 *Drosophila* survey compared just 2 related species, and here the numbers of loci at which the most frequent alleles differed in mobility ranged from 1 to 12. Such data give an inadequate data

Interspecific comparisons of allozyme mobility involving surveys screening 3 or more species

Group	N	χ^2	d.f.	p
Vertebrates, overall	30 (141)	120.74	111	0.2483
Icteridae[11]	1 (7)	14.42	6	0.0253
Cichlidae[12]	1 (6)	18.29	5	0.0026
Drosophila, overall	5 (27)	27.74	22	0.1845

N is the number of surveys analyzed, the number in parenthesis being the total number of species screened.

base for testing the hypothesis meaningfully. In the single study[9] where 12 loci differed in the mobility of their common alleles, 9 were faster in the 1st species than the 2nd, and 3 were slower.

More interesting are those studies comparing 3 or more species. Each study was analyzed by Friedmann's test[10], using only those loci whose most frequent allele differed in mobility in at least 1 species. At each locus, species were ranked according to allozyme mobility, and the summed ranks of each species computed. Large differences in summed ranks between species are indicative of the mobility correlations that are being looked for. The test statistic for each data set is a χ^2 with degrees of freedom equal to one less than the number of species surveyed.

A total of 5 *Drosophila* surveys and 30 vertebrate surveys proved suitable for such analysis, and of these only 2 of the vertebrate surveys gave results significant at the 5% level, indicating the existence of mobility correlations. Neither the overall vertebrate nor overall *Drosophila* χ^2-values were significant (table). However, the 2 significant surveys (table) merit further discussion. Both are unusual in that they describe intergeneric rather than intra-generic comparisons, yet only 5 of the 30 vertebrate data sets were concerned with inter-generic comparisons. The study on Icteridae (Aves)[11] involved 7 species of 6 genera, that on Cichlidae (Pisces)[12] 6 species of 4 genera.

In the absence of an a priori hypothesis that these 2 studies would yield significant results, it could be argued that in 30 surveys it is not surprising that 2 are statistically significant. In fact, the probability of obtaining, by chance, in 30 studies, 1 study with $p \leqslant 0.0026$ and 1 with $p \leqslant 0.0253$ is equal to (30) (29) $(0.9747)^{28}$ (0.0253-0.0026) (0.0026)= 0.0251. To this should be added the probabilities of obtaining even more unlikely results, values which are extremely small. Therefore we can conclude a posteriori that the probability of obtaining by chance at least 2 such significant results is less than 0.05.

Thus we have little evidence favoring the hypothesis that speciation is generally accompanied by a net alteration in enzyme change. However, we do have evidence for correlated changes in allozyme mobility between species in 2 data sets, both describing inter-generic differentiation. It may be that niche separation of species in different genera is generally greater than that of species in the same genus, and therefore any selection for an overall alteration in enzymic charge is likely to be stronger and more readily apparent. It follows that if charge per se is subject to selection, then the results from these 2 studies may be viewed as good evidence for the selectionist hypothesis. However, the majority of surveys accord with expectations of the neutral model in not showing consistent differences in allozyme mobility between related species.

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Hydrophobic properties of model compounds with peptide-like chemical environment: N-acetyl-N′-methyl-amino acid amides

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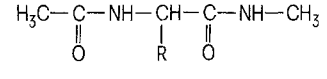
Summary. Capacity factors in reversed-phase HPLC and distribution constants in octan-1-ol/water of N-acetyl-N′-methyl-amino acid amides have been measured as a function of temperature. The HPLC capacity factors are proposed as estimates of the hydrophobicity of the amino acid side chains.

Hydrophobic effects are predominant in determining a number of different physicochemical and biological properties of peptides and proteins. The stabilization of the native conformation of proteins[2-4], the binding of biologically active peptides to enzymes and receptors[5] and the retention of small peptides in reversed-phase liquid chromatography[6] are among a few instances where the hydrophobicity of the amino acid residues is thought to play a substantial role.

Current hydrophobicity scales for the amino acid side chains are derived from various experimental data on the amino acids, e.g. solubility in alcohols[3,7], partition between immiscible solvents[8,9], chromatographic retention[10,11] and surface tension[12]. It is open to question, however, whether these scales measure the hydrophobicity of the side chain itself, since the zwitter-ionic nature of the amino acids is likely to affect hydrophobicity, for example by changing the type and extent of hydratation. Moreover, in no case was unambiguous evidence presented that a true, thermodynamically defined hydrophobic effect had been measured. Ideally, true hydrophobicity is characterized by a large positive excess of entropy change due to desolvation of apolar molecules and concomitant loss of water structure[2,13].

We have set out to measure the hydrophobic properties of N-acetyl-N′-methyl-amino acid amides with the general formula:



These compounds are un-ionized and may be considered as small peptide fragments, since the single amino acid side chain, (R), is contained between 2 amide bonds, much the same as it is in peptides.