Mutation as the source of the abnormal gene for plasma cholinesterase

Sir,

Recently, you published an article by Evans and Magill (1974) suggesting that a mutation hypothesis (one of the alleles for the E_1 locus of cholinesterase had mutated) was the most likely hypothesis for explaining the genotype of two deceased parents among whose children there were three phenotypes; usual, silent, and intermediate in the family which they reported. The enzyme from two of the usual phenotypes had activity expected for the $E_1^sE_1^s$ genotype but from one had activity which was twice as high as that from the other two usual phenotypes in the sibship. They discussed three other hypotheses which were considered correctly by the authors to be most unlikely: (1) a child in the sibship did not come from the same two parents, (2) the atypical and silent genes were non-allelic, or (3) there was a suppressor gene.

There is one explanation for their family data which was not suggested and which in my opinion is far the most likely one. That is that the parents had the genotypes $E_1^sE_1^s$ and $E_1^sE_1^s$ and that the offspring, whose enzyme activity was high and the phenotype was usual, had a genotype $E_1^sE_1^s$ and that there was as much enzyme activity determined by his one E_1^s allele as is produced on the average in homozygotes for the E_1^s allele, or he had also the allele at the E_2 locus ($C5+$) which produces higher activity of either the usual or atypical forms of the enzyme (Harris *et al.*, 1962).

We have measured the levels of cholinesterase activity in 26 obligate heterozygotes ($E_1^sE_1^s$). These individuals had the usual phenotype and were parents or children of eight persons who were homozygous ($E_1^sE_1^s$) for the 'silent' allele and parents or children of 10 persons who were the $E_1^sE_1^s$ genotype. Five of the latter cases were described in Simpson and Kalow (1964). Although the mean activity for the 26 heterozygotes was 137 units (units were in μmol of acetylcholine hydrolysed by 1 ml of serum at 37° C during 1 h) and the mean for a population of 268 individuals who were probably homozygous for the usual gene and $C5-$ was 208 units (Simpson, 1971), the variances were large (the standard deviation for the heterozygotes was ± 38 and for homozygotes was ± 66 units) and consequently there were a few $E_1^sE_1^s$ heterozygotes whose activity was as high or higher than the mean for homozygotes. It is also known that the activity of the enzyme is influenced by age with a sharp decrease up to about the age of 20 years and then a gradual decrease (Scott *et al.*, 1970). In our series of 26 $E_1^sE_1^s$ heterozygotes, the highest activity was 233 units in a woman who was 26 years of age and $C5-$; an activity considerably above the mean for the $E_1^sE_1^sC5-$ genotype.

The above data show that there is considerable overlap in the distributions between activity for cholinesterase in sera from the $E_1^sE_1^s$ and $E_1^sE_1^s$ genotypes and that $C5$ and age also need to be considered. Scott *et al.* (1970) have discussed the problems of discriminating between these two genotypes in detail. The age of the offspring with the usual phenotype and high enzyme activity in Evans' and Magill's pedigree was 51 years and if his genotype is $E_1^sE_1^s$, as I am suggesting, his higher than expected activity might be due to the $C5+$ allele or simply to the fact that his value is in the extreme right tail of a normal distribution for activity from $E_1^sE_1^s$ individuals.

Yours, etc,

Nancy E. Simpson

REFERENCES

- Evans, R. T. and Magill, P. J. (1974). Evidence for mutation being the source of the abnormal gene for plasma cholinesterase. *Journal of Medical Genetics*, **11**, 117-120.
- Harris, H., Hopkinson, D. A., and Robson, E. B. (1962). Two-dimensional electrophoresis of pseudocholinesterase components in normal human serum. *Nature*, **196**, 1296-1298.
- Scott, E. M., Weaver, D. D., and Wright, R. C. (1970). Discrimination of phenotypes in human serum cholinesterase deficiency. *American Journal of Human Genetics*, **22**, 363-369.
- Simpson, N. E. (1971). A geneticist's look at pharmacogenetics. *International Journal of Clinical Pharmacology, Therapy and Toxicology*, **5**, 44-50.
- Simpson, N. E. and Kalow, W. (1964). The 'silent' gene for serum cholinesterase. *American Journal of Human Genetics*, **16**, 180-188.

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Sir,

It was with great interest that we read the comments of Dr Simpson on our recent article and her alternative to our mutation hypothesis, which she based upon the supposition that a patient, labelled by us as normal, was in fact a heterozygote for the normal and silent genes.

We believe her suggestion to be unlikely for the following reasons.

1. Measurement of the serum cholinesterase activity of 10 patients, shown in this laboratory to be of the $EuEs$ genotype, has not confirmed her observation of significant overlap between their enzyme levels and those of normals. The mean value on these subjects, obtained by the method of Johnson and Whitehead (1965) using acetyl choline bromide as substrate, was 151 IU/dl ($SD = 36$), the highest value being 214 IU/dl. These figures compare with one of 432 IU/dl in the patient in question, eight standard deviations above the mean for $EuEs$ heterozygotes, and with a mean of 385 IU/dl ($SD = 68$) in normals (Fishtal *et al.*, 1972).

2. No evidence for the presence of a silent gene has been found in any of the patient's three children.

3. If the total enzyme activity of 432 IU/dl were due to the presence of one normal gene only, it would be very difficult to explain the much lower levels seen in the three siblings in whom this gene is also found and who demonstrate serum enzyme activities less than half that of their brother.

4. The only possible support for Dr Simpson's thesis would derive from the presence of an allele at the second locus for serum cholinesterase (C5+).

Our gratitude is due to Dr Mary Whittaker of the University of Exeter who has very kindly repeated and confirmed our published findings and has also carried out the electrophoretic investigations necessary for the study of the second locus. No evidence for enzyme from this source has been found by Dr Whittaker in any members of this family.

The significance of Dr Simpson's reference to the age

of our patient is doubtful, since correction of his serum enzyme activity to that expected at 28 years, the age of his youngest brother, would result in an increase of some 15% and would render her theory even less likely.

We must therefore conclude that, despite her detailed argument to the contrary, evidence at present available is in support of our original hypothesis.

Yours, etc,

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REFERENCES

- Fishtal, A., Evans, R. T., and Chapman, C. N. (1972). An original approach to the diagnosis of scoline induced apnoea. *Journal of Clinical Pathology*, **25**, 422-426.
- Johnson, J. K. and Whitehead, T. P. (1965). Rapid estimation of serum cholinesterase activity using the Astrup micro equipment. *Journal of Clinical Pathology*, **18**, 435-440.