

## LETTERS TO THE EDITOR

### The Gene Bomb

I would like to comment on Tom Wilkie's comments on *The Gene Bomb*<sup>1</sup> (*J Med Genet* 1997;34:438-9), since I feel he missed the whole point of the book and presented to the readers of this journal a distorted view of the issues I attempted to raise. They have implications for all of us.

First, the subtitle, which somewhat more accurately portrays the issues, *Does Higher Education and Advanced Technology Accelerate the Selection of Genes for Learning Disorders, ADHD, Addictive and Disruptive Behaviors?* was left out.

Second, no mention was made of part I of the book which reviews data showing the following.

(1) Epidemiological studies using structured diagnostic instruments have shown a highly significant increase in frequency, and decrease in the age of onset, in younger cohorts for all the major psychiatric disorders including depression, manic depressive disorder, schizoaffective disorder, suicide, obsessive-compulsive disorders, panic disorder, phobias, alcoholism, and drug abuse.

(2) Using the Achenbach Child Behavior Checklist, Achenbach performed epidemiological studies in 1976 and 1989. Of 112 behavioural scores, 42 were worse in 1989 while none was worse in 1976. In addition, behaviours relating to specific diagnoses, such as ADHD and conduct disorder, increased from 10% in 1976 to 18.2% only 13 years later. In 1976, 8.2% of children had to repeat a grade while in 1989 17.8% had to repeat a grade.

(3) Studies of the same localised region in Sweden by Gillberg have shown an increase in the frequency of autism from 4/10 000 in 1980, to 7.5/10 000 in 1984, to 11.5/10 000 in 1988.

(4) US Department of Education studies have shown a progressive increase in the frequency of learning disabilities from 890 000 cases in 1976 to 2 714 000 cases in 1993. At the same time the population has only increased from 210 000 000 to 240 000 000.

To date there have been no adequate explanations for these trends and observations, very few professionals, including geneticists, have either been aware of or concerned about these trends, and no one has offered any satisfactory explanation for them. I work with children with hereditary behavioural disorders (ADHD, Tourette syndrome, learning disabilities, autism, Asperger syndrome, conduct disorder, and others) on a daily basis and I and their parents are concerned. I am particularly concerned about the trend in learning disabilities. These have increased from 2 000 000 in 1988 to 2 700 000 in 1993. At this rate the frequency of learning disorders is doubling every 15 years.

In attempting to understand these trends, I noticed the following. First, all of these disorders have a strong genetic component. Second, there is a strong inverse correlation between the frequency of these disorders,

especially learning disorders, and years of education. Not surprisingly, those with learning disorders tend to drop out of school, while those without learning disorders are more likely to go to college and possibly graduate school. Third, there is an extraordinarily high inverse correlation between the number of years of education and the age of initiation of child birth. At the beginning of this century only 2% of the population attended college. The rate dramatically increased after the second world war and now 38% attend college. All of these trends in behaviour disorders have occurred in the latter half of this century. At the present time the spread in years of the age of onset of childbearing, for those who drop out of school before completing a high school education versus those who go to college, is 8.9 years. Fourth, like compound interest, anything that turns over more rapidly will more rapidly increase in frequency. This can provide a very strong selection for the genes associated with learning disability, totally independent of the number of children people have. Comparing 1920 to 1990 shows that the magnitude of the selective force (spread in years by percent attending college) has increased 40 fold.

How does one respond to these observations or concerns? There are several possibilities. One is to shoot the messenger, as Wilkie has chosen to do. A second is to provide alternative explanations for these trends. I would prefer this so I can then stop worrying about the subject. The third is to accept it as a legitimate concern and either disprove the hypothesis or continue research into the genes involved in these disorders so that their frequencies in the population by age can be monitored.

In the book I suggested two approaches. The first were "motherhood and apple pie" approaches that no one should object to, such as special programmes to encourage children with ADHD and learning and related disorders to remain in school, and programmes for day care for children of college students so that if they wished, they could start a family before completing their education. The second approach is to continue (or accelerate) the research towards providing a better understanding of the genes involved. Among other things this would allow couples to participate in totally voluntary programmes by which they could make informed choices about reproductive decisions, as we now encourage people with a family history of cystic fibrosis or Huntington's disease to do.

Finally, I would like to point out that when the various behavioural disorders, most of which appear to be polygenic, are combined they occur in over 35% of the general population. This is 35 times the frequency of all the single gene disorders combined. I would suggest that those who propose that geneticists should not be involved in studies of the genetics of human behaviour, and prefer that we do not engage in responsible talk about some of the social implications of these studies, because it may give geneticists a bad name, are guilty of sticking their head in the sand. If geneticists don't become involved and play some role in attempting to understand the cause of these trends, I suspect no one will. In his book, *And the Band Played On*, Randy Shilts severely chastised the professional community for failing to mount an early response to the AIDS epidemic, because they preferred not to become involved or felt uncomfortable with the subject. Are we going

to be similarly chastised in the next century because we also failed to care, or felt uncomfortable with the subject, and made no attempt to understand these trends?

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1 Comings DE. *The gene bomb. Does higher education and advanced technology accelerate the selection of genes for learning disorders, ADHD, addictive, and disruptive behaviors?* Duarte, CA: Hope Press, 1996.

### Gaucher disease plus

In a recent letter, Uyama *et al*<sup>1</sup> discussed a Japanese sibship of three subjects with glucocerebrosidase deficiency. These patients had oculomotor abnormalities characteristic of neuronopathic Gaucher disease along with several more unusual clinical manifestations described in other populations, including valvular calcifications, communicating hydrocephalus, and corneal opacities.<sup>2-5</sup> Yet although the patients are reported to have glucocerebrosidase deficiency and point mutations on both glucocerebrosidase alleles, the authors suggest that the patients have a "Gaucher-like" disorder.<sup>1</sup>

Gaucher disease is characterised by a vast phenotypic heterogeneity that is continually expanding.<sup>6</sup> Recently, there has been a greater appreciation that diverse symptoms including congenital ichthyosis,<sup>7</sup> hydrops fetalis,<sup>7</sup> primary pulmonary hypertension,<sup>8</sup> valvular calcifications,<sup>3-5</sup> and even Parkinsonian symptoms<sup>9</sup> can be seen in patients with Gaucher disease. Thus, we would suggest that the symptoms observed in the patients described by Uyama *et al*<sup>1</sup> are just part of the broader Gaucher phenotype.

Although the authors mention other genes near to glucocerebrosidase, they discount the likelihood of a contiguous gene effect.<sup>1</sup> Since the discovery of human metaxin,<sup>10</sup> a gene located immediately downstream and transcribed convergently to the glucocerebrosidase pseudogene, we have characterised the locus more completely and have found that the region encompassing glucocerebrosidase is particularly gene rich.<sup>11,12</sup> The physical relationship of metaxin and glucocerebrosidase on human chromosome 1q21 described by Uyama *et al*<sup>1</sup> is incorrect, for actually the pseudogene for human metaxin and not the gene is immediately downstream to the glucocerebrosidase gene.<sup>10</sup> We suggest that patients with these more unusual Gaucher phenotypes may indeed have "Gaucher disease plus", an interruption in a second contiguous gene, rather than a "Gaucher-like" illness. In Hunter syndrome, it has recently been shown that some patients with atypical phenotypes have deletions including segments of nearby genes.<sup>13</sup> Further characterisations of all the genes around the glucocerebrosidase locus may provide a more complete explanation for the phenotypic variation observed in Gaucher disease.

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## Molecular characterisation of cystic fibrosis patients in the state of São Paulo (Brazil)

To characterise the CF patient population in the state of São Paulo (Brazil), we estimated the frequency of mutations  $\Delta F508$ , G542X, N1303K, G551D, and R553X, which are some of the most frequent mutations worldwide. In a sample of 120 CF chromosomes, the frequencies we obtained were as follows:  $\Delta F508=31.7\%$ , G542X=8.3%, N1303K=2.5%. The G551D and R553X mutations were not detected in this sample. The mutation detection rate was 42.5% based on the analysis of these five mutations. We conclude that further screening for more mutations will be necessary for molecular diagnosis and carrier detection because of the ethnic heterogeneity of this population. The present study represents a step towards the molecular characterisation of CF patients in the state of São Paulo.

Cystic fibrosis is the most common autosomal recessive disease, with an incidence of 1 in 2000-3000 births in various groups.<sup>1</sup> The CF gene was cloned in 1989.<sup>2,4</sup> The major mutation causing CF is a three base pair deletion which results in the loss of phenylalanine 508 ( $\Delta F508$ ).<sup>4</sup> This mutation is found on about 70% of CF chromosomes worldwide and varies considerably between populations. The CF Genetic Analysis Consortium has identified over 620 other sequence alterations in the CF gene. Most of these mutations are rare. Only a few mutant alleles have a worldwide frequency of 1% or greater, although they may occur at higher frequencies in selected populations.<sup>5</sup>

Knowledge of the spectrum of CF gene mutations in different populations allows for the possibility of genetic testing. In Brazil, Raskin *et al*<sup>6</sup> and Martins *et al*<sup>7</sup> evaluated the frequency of  $\Delta F508$  in the state of São Paulo. Their results showed frequencies of 52% (60/116) and 31% (15/48), respectively. In view of these differing results and in order to contribute to the molecular characterisation of CF patients in the state of São Paulo, we have analysed the frequencies of CF mutations  $\Delta F508$ , G542X,<sup>8</sup> N1303K,<sup>9</sup> G551D,<sup>10</sup> and R553X.<sup>10</sup>

Sixty unrelated CF patients regularly followed at the University (UNICAMP) Hospital in Campinas were studied. Blood samples were obtained from these patients, who had been diagnosed with CF on the basis of typical clinical manifestations or a positive sweat test (>60 mEq/l) or both. Genomic DNA was extracted from peripheral blood leucocytes according to standard protocols.

DNA amplification was performed according to a general procedure.<sup>11</sup> The primers and annealing temperatures used for each amplified region have been reported before.<sup>4,12</sup>

Amplified samples from each subject were screened for the five previously mentioned mutations. The mutation  $\Delta F508$  was detected by heteroduplex DNA formation.<sup>13</sup> The mutations G542X and N1303K were detected by digestion with *Bst*NI<sup>12</sup> and mutations G551D and R553X were detected by digestion with *Hinc*II followed by ASO hybridisation. This combination of methods was used because digestion with *Hinc*II by itself does not distinguish mutations G551D and R553X.<sup>12</sup>

The frequencies of mutations  $\Delta F508$ , G542X, and N1303K in the sample studied were 31.7%, 8.3%, and 2.5%, respectively. Mutations G551D and R553X were not detected.

The main objective of this study was to contribute to the molecular characterisation of CF patients in the state of São Paulo. The observed frequency of the  $\Delta F508$  mutation (31.7%) is consistent with the ethnic composition of the white population in this state, which consists predominantly of immigrants from Italy, Portugal, and Spain where the frequency of this mutation is lower than in the rest of the world (70%).<sup>14,16</sup> Our result for  $\Delta F508$  differed significantly from that reported by Raskin *et al*<sup>6</sup> ( $\chi^2_{(1)}=9.77$ ,  $p<0.01$ ) but is in accord with that reported by Martins *et al*<sup>7</sup> ( $\chi^2_{(1)}=0.003$ ,  $p>0.95$ ). The sample studied by Raskin *et al*<sup>6</sup> might be composed essentially of patients with more severe phenotypes, which are correlated with the  $\Delta F508$  mutation.

The frequencies of the next two most common mutations in the sample studied are also consistent with the above ethnic composition. The G542X mutation is fairly common throughout Europe, particularly southern Europe,<sup>17</sup> and the N1303K mutation is common throughout Europe. Mutations G551D and R553X have higher frequencies in northern European countries, and for this reason they may be rare in the population of the state of São Paulo.

The most immediate application which results from determination of the spectrum of CF gene mutations in different populations is genetic testing. In the sample of CF chromosomes that we studied, 42.5% of them could be identified by the analysis of five mutations. Reaching a 90% detection rate in this population may require testing many additional low frequency mutations since São

Paulo, like the whole of Brazil, is genetically very heterogeneous.

In conclusion, population molecular characterisation is an important aspect in the study of CF but, as expected for any non-isolated population, it depends on the analysis of a great number of mutations. Our data should contribute further to the characterisation of CF in the state of São Paulo.

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