diseases, large familial correlations) are the same cases in which LD and HWD tests are likely to be useful (see Zöllner and von Haeseler 2000; Terwilliger, in press). In a study of more-common phenotypes and larger, more diverse populations, it is highly unlikely that marginal effects of single-risk alleles of a given locus are going to be etiologically important—in which case, LD and HWD tests will have little or no power (see Terwilliger and Weiss 1998; Terwilliger and Göring 2000; Weiss and Terwilliger, in press). And small populations with unusual histories are also more likely to have some population-level deviation from HWE in general, and, if one does not ascertain population controls, then there is no way to validate this critical assumption of the model. Although the paranoia about population stratification that leads people to mistrust case-control samples may be exaggerated, the absence of a sample of controls poses even greater danger.

Acknowledgments

Support from a Hitchings-Elion Fellowship from the Burroughs-Wellcome Fund is greatly appreciated. Discussions with Drs. Iiris Hovatta, Harald H. H. Göring, John Blangero, Patrik Magnusson, and Kenneth M. Weiss are gratefully acknowledged.

JOSEPH D. TERWILLIGER
Department of Psychiatry, Columbia University, Columbia Genome Center, and Division of Neuroscience, New York State Psychiatric Institute, New York

References

Chase GA (1977) Genetic linkage, gene-locus assignment, and the association of alleles with diseases. Transplant Proc 9: 167–171

Deng HW, Chen WM, Recker RR (2000) QTL fine mapping by measuring and testing for Hardy-Weinberg and linkage disequilibrium at a series of linked marker loci in extreme samples of populations. Am J Hum Genet 66:1027–1045

Göring HHH, Terwilliger JD (2000) Linkage analysis in the presence of errors. IV. Joint pseudomarker analysis of linkage and/or linkage disequilibrium on a mixture of pedigrees and singletons when the mode of inheritance cannot be accurately specified. Am J Hum Genet 66:1310–1327

Hovatta I, Varilo T, Suvisaari J, Terwilliger JD, Ollikainen V, Arajärvi R, Juvonen H, et al (1999) A genomewide screen for schizophrenia genes in an isolated Finnish subpopulation, suggesting multiple susceptibility loci. Am J Hum Genet 65:1114–1124

Nielsen DM, Ehm MG, Weir BS (1999) Detecting marker-disease association by testing for Hardy-Weinberg disequilibrium at a marker locus. Am J Hum Genet 63:1531–1540

Risch N (1990) Linkage strategies for genetically complex traits. I. Multilocus models. Am J Hum Genet 46:222–228

Terwilliger JD. On the resolution and feasibility of genome scanning approaches to unraveling the genetic components of multifactorial phenotypes. In: Rao DC, Province MA (eds) Genetic dissection of complex phenotypes: challenges for the new millennium. Academic Press, New York (in press)

Terwilliger JD, Göring HHH (2000) Gene mapping in the 20th and 21st centuries: statistical methods, data analysis, and experimental design. Hum Biol 72:63–132

Terwilliger JD, Shannon WD, Lathrop GM, Nolan JP, Goldin LR, Chase GA, Weeks DE (1997) True and false positive peaks in genomewide scans: applications of length-biased sampling to linkage mapping. Am J Hum Genet 61:430–438

Terwilliger JD, Weiss KM (1998) Linkage disequilibrium mapping of complex disease: fantasy or reality? Curr Opin Biotechnol 9:578–594

Weiss KM, Chakraborty R, Majumder PP, Smouse PE (1982) Problems in the assessment of relative risk of chronic disease among biological relatives of affected individuals. J Chronic Dis 35:539–551

Weiss KM, Terwilliger JD. How many diseases do you have to study to map one gene with SNPs? Nat Genet (in press)

Zöllner S, von Haeseler A (2000) A coalescent approach to study linkage disequilibrium between single-nucleotide polymorphisms. Am J Hum Genet 66:615–628

Address for correspondence and reprints: Dr. Joseph D. Terwilliger, Columbia University, Unit 109, 1150 St. Nicholas Avenue, Room 520C, New York, NY 10032. E-mail: jdt3@columbia.edu
© 2000 by The American Society of Human Genetics. All rights reserved.
0002-9297/2000/6701-0034$02.00

Am. J. Hum. Genet. 67:259–261, 2000

QTL Fine Mapping, in Extreme Samples of Finite Populations, for Complex Traits with Familial Correlation Due to Polygenes

To the Editor:

Recently, Deng et al. (2000) developed a QTL fine-mapping approach on the basis of Hardy-Weinberg (HW) or linkage-disequilibrium (LD) patterns in extreme samples of large and random-mating populations in which HW equilibrium holds. This approach is based on robust linkage results that have already localized a quantitative-trait locus (QTL) to a large genomic region (e.g., ~30 cM). The purpose is to fine map the QTL to a small region of ~1 cM, through examination of the patterns of deviation from HW and linkage equilibrium at a series of closely linked marker loci in extreme samples of populations. The deviation can be measured by a number of indices (including some test statistics—e.g., the test statistics for HW equilibrium) (Deng et al. 2000). Our approach is an extension of those of Feder et al. (1996) and Nielsen et al. (1998) for fine mapping of disease-

susceptibility loci. Feder et al. have successfully used this approach by examining HW disequilibrium patterns in affected cases, at a series of closely linked marker loci, to fine map a susceptibility locus for hereditary hemochromatosis, to a region of ~600 kb.

As pointed out by Terwilliger (2000 [in this issue]), in the development of our fine-mapping approach for genes underlying common complex traits, familial correlations of complex traits are ignored, and large randomly mating populations are assumed. In response to the concerns of Terwilliger, we performed simulations to investigate the performance of our approach in finite populations, in the presence of familial correlations for the trait under study.

The simulation procedures are roughly the same as those described by Deng et al. (2000), except that families with familial correlations for the trait are simulated for finite populations. In brief, an evolving population of size N is simulated for 50 generations. In each generation, random pairs of individuals are mated to generate the next generation. The number of children per family is generated from a Poisson distribution with a mean of 2. To maintain a constant population size of N from generation to generation, if the number of children generated is $>N$, random children are discarded, so that the total number of children is N for the next generation. If the number of children simulated is $<N$, random parental pairs are included, to generate more children (according to the Poisson distribution for each pair), until there are N children generated for the next generation. Without loss of generality, the family correlation is simulated via 10 unlinked biallelic background polygenic QTLs. The effect of the background polygenic QTLs is indexed by the heritability (h^2_{pg}) attributable to them. Each polygenic QTL has the same recessive effect, so that its heritability is $h^2_{pg}/10$; the frequency of the allele causing lower trait values is .2. The correlations among family members can be easily computed from h^2_{pg} and from the heritability (h^2) of the QTL being tested. In simulations, the frequency of the allele causing lower trait values at the QTL is $p = .1$, the marker-allele frequency $p_M = .2$, and $h^2 = .20$. If the marker is linked to the QTL, $D^0_{A_1M}$ (the amount of LD simulated at the 0 generation) = .08. The initial LD may be caused by various evolutionary scenarios, such as admixture of populations differentiated at the QTL and marker frequencies. At the 50th simulated generation, extreme samples are taken from the simulated populations. For a series of closely linked marker loci that are simulated around the QTL being tested, we performed QTL fine mapping by the five-point moving-average technique (Deng et al. 2000). The first stage of fine mapping (Deng et al. 2000) is more robust than the second stage and is little affected by finite population sizes and familial correlations. Therefore, we present, in figure 1, only the results for

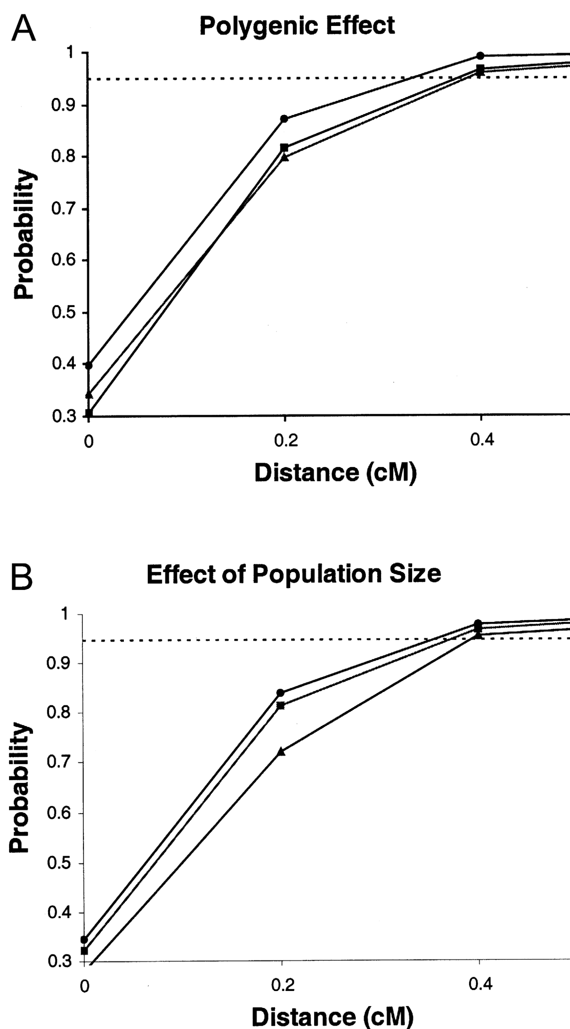

Figure 1 Effects of polygenic QTLs and population sizes on the QTL fine-mapping approach of Deng et al. (2000). For plot *A*, the population site is 5,000; circles, triangles, and squares represent the data for situations in which h^2_{pg} = .0, .3, and .6, respectively. For plot *B*, h^2_{pg} = .5; circles, squares, and triangles represent the data for situations in which the population sizes are 15,000, 5,000, and 2,000, respectively.

the second stage of fine mapping, when highly dense markers (~0.2 cM apart from one another) are typed around the QTL position. In figure 1, the Y-axis is the probability that the true QTL position is within a certain distance (X-axis) from the peak of the LD measure q_{excess} (Deng et al. 2000) at a series of closely linked markers. Measurement of disequilibrium by q_{excess} uses individuals from the bottom and the top 10 percentiles (100 each) in study populations. The results for other LD measures (including those for HW disequilibrium—the χ^2_2 and χ^2_4 [Deng et al. 2000]) are essentially the same.

Even with small population sizes (as small as 2,000)

and large familial correlations (as reflected by h^2_{pg}, which is as large as .6), our QTL fine-mapping approach not only remains powerful but also is valid and robust (fig. 1). Under the parameters simulated, the correlation between full sibs is .36, and that between a parent and a child is .27, when $h^2_{pg} = .5$ (plot *B* in fig. 1). Finite population sizes and familial correlations may lower the power of our QTL fine-mapping approach, especially when the marker is extremely close (<0.2 cM) to the true QTL position. However, the effect is very small. In particular, when the distance of the peak from the true QTL position is >0.5 cM, our power of QTL fine mapping is little affected. Recall that the purpose of our QTL fine-mapping approach is to narrow a large genomic region found in regular linkage analyses to a small region of ~1 cM, for further physical mapping to clone the QTL. With finite population sizes and familial correlations, our approach can have >95% probability to correctly position the QTL to a region <0.8 cM (fig. 1).

Acknowledgments

This study is partially supported by grants from the National Institutes of Health, Health Future Foundation, and Hunan Normal University and by a student tuition waiver to W.-M. Chen from Creighton University.

HONG-WEN DENG[1,2] AND WEI-MIN CHEN[1]
[1]*Osteoporosis Research Center and Department of Biomedical Sciences, Creighton University, Omaha; and* [2]*Laboratory of Molecular and Statistical Genetics, College of Life Sciences, Hunan Normal University, ChangSha, Hunan, China*

References

Deng HW, Chen WM, Recker RR (2000) QTL fine mapping by measuring and testing for Hardy-Weinberg and linkage disequilibrium at a series of linked marker loci in extreme samples of populations. Am J Hum Genet 66:1027–1045
Feder JN, Gnirke A, Thomas W, Tsuchihasi Z (1996) A novel MHC class I-like gene is mutated in patients with hereditary haemochromatosis. Nat Genet 13:399–408
Nielsen DM, Ehm MG, Weir BS (1998) Detecting marker-disease association by testing for Hardy-Weinberg disequilibrium at a marker locus. Am J Hum Genet 63:1531–1540
Terwilliger JD (2000) Inflated false-positive rates in Hardy-Weinberg and linkage-equilibrium tests are due to sampling on the basis of rare familial phenotypes in finite populations. Am J Hum Genet 67:258–259 (in this issue)

Address for correspondence and reprints: Dr. Hong-Wen Deng, Osteoporosis Research Center, Creighton University, 601 North 30th Street, Suite 6787, Omaha, NE 68131. E-mail: deng@creighton.edu
© 2000 by The American Society of Human Genetics. All rights reserved.
0002-9297/2000/6701-0035$02.00

Am. J. Hum. Genet. 67:261–262, 2000

Reply to Deng and Chen

To the Editor:
The simulations done by Deng and Chen (2000), in response to my letter (Terwilliger 2000), are completely consistent with one of the points that I was trying to make. The pointwise mean and variance of the distribution of the test statistics are slightly inflated in extreme samples from small populations, so that, when such analyses are performed over much larger genomic regions, as in a genome scan, these seemingly minor pointwise effects can be dramatic. This is the real danger in such studies, which could lead to a potential sea of false positives in the literature, swamping the likely dearth of true-positive findings (see Weiss and Terwilliger, in press). The effects of “extreme sampling” are going to be much greater when the frequency of the phenotype is <10% (which is very common for a disease phenotype) and/or the effective population size is smaller (e.g., because of rapid population expansion and/or more-extreme isolation), as seen in the schizophrenia study by Hovatta et al. (1999). But, even under this “best-case scenario,” Deng and Chen showed that there is an inflation of mean and variance of their statistics under H_0, even for $P \leqslant .05$, and, when one gets closer to the critical values needed in a genomewide sense (which must be more, not less, strict than those used in linkage analysis—$P < .0001$), the inflation must be larger still (also see Terwilliger and Göring 2000 and Terwilliger, in press). Furthermore, under the model that I described, the familial correlations in phenotype could have absolutely nothing to do with genetic factors at all (like “ability to speak Finnish” in a sample of Americans); yet the same problems would result, because familial phenotypes correlate with familial substrata of the population, leading to potentially increased rates of false evidence of both Hardy-Weinberg and linkage disequilibrium, compared with what is seen in random samples from the whole population.

JOSEPH D. TERWILLIGER
Department of Psychiatry, Columbia University, Columbia Genome Center, and Division of Neuroscience, New York State Psychiatric Institute, New York

References

Deng HW, Chen WM (2000) QTL fine mapping in extreme samples of finite populations for complex traits with familial correlation due to polygenes. Am J Hum Genet 67:259–261 (in this issue)
Hovatta I, Varilo T, Suvisaari J, Terwilliger JD, Ollikainen V,