Age and Sex Effects on Human Mutation Rates: An Old Problem with New Complexities

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Human mutation/Paternal age/Apert syndrome/Weinberg/Pre-meiotic selection/Mutation load.

Base substitution mutations are far more common in human males than in females, and the frequency increases with paternal age. Both can be accounted for by the greater number of pre-meiotic cell divisions in males, especially old ones. In contrast, small deletions do not show any important age effect and occur with approximately equal frequency in the two sexes. Mutations in most genes include both types, and the sex and paternal age effect depends on the proportion of the two types. A few traits, of which Apert Syndrome is best understood, are mutation hot spots with all the mutations occurring in one or two codons, usually at one nucleotide. They occur with very high frequency almost exclusively in males and the frequency increases rapidly with paternal age. It has been suggested that the mutant cells have a selective advantage in the male germ-line prior to meiosis. Evidence for this surprising, but important, hypothesis is discussed. A possible mechanism is the conversion of asymmetrical stem-cell divisions into symmetric ones. Some traits with complex etiology show a slight paternal age effect. There is also a short discussion of the high deleterious mutation rate and the role of sexual reproduction in reducing the consequent mutation load.

MUTATION FREQUENCIES THAT ARE GREATER IN MALES AND INCREASE WITH PATERNAL AGE

As long ago as 1912 Weinberg¹ noticed that children with achondroplasia (short limbed dwarfism) from normal parents tended to be late-born in the sibship. He astutely suggested that, if this is confirmed, it would argue for mutation as the causative factor. Coming only 12 years after the rediscovery of Mendelism and at a time when mutation was only vaguely understood, this is indeed a remarkable insight. Weinberg is best known for the mathematically trivial Hardy-Weinberg rule, but this is just the simplest of the many important findings of this gifted scientist. He was also a busy physician who practiced medicine for 42 years and delivered more than 3500 babies.²

Whether the increase in late-born children was the result of paternal age, maternal age, or birth order was not determined. The question remained unresolved for more than 40 years. Finally, in 1955 Penrose³ showed that the effect was mainly, if not entirely, the consequence of paternal age. The association with maternal age was no greater than would be expected from the age-correlation between mates. Penrose found no independent effect of birth order.

The large paternal age effect suggests that the mutation rates may differ in the two sexes. In the early days, it was not possible to distinguish paternal from maternal mutations at autosomal loci, but X-linked traits permitted this distinction. The first person to exploit that possibility was J. B. S. Haldane,⁴ who studied X-linked hemophilia. He noticed that the great majority of affected males come from heterozygous mothers, and from this estimated that the average male mutation rate is about ten times that of females. This is only a crude result, since at that time heterozygote detection was inexact and hemophilia was not always distinguished from other bleeding diseases that mimic it. Further studies provided additional evidence of a greater male rate, but did not permit an accurate estimate of the male:female ratio.⁵ A similar excess of carriers among the parents of affected males was found for Lesch-Nyhan syndrome.⁶

A more recent example is X-linked OTC (ornithine transcarbamylase) deficiency. Tuchman et al.⁷ reported that 2/28 of affected males had new mutations (the others had heterozygous mothers) as did 12/15 of heterozygous females. The high proportion of affected males from heterozygous mothers again argues for a higher male mutation rate. The low fitness of affected males and carrier females means that equilibrium is attained quickly, so equilibrium equations are appropriate. We can obtain a quantitative estimate as fol-

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¹Special lecture
lows:

Let \( q_m \) and \( q_f \) be the equilibrium allele frequencies in males and females, \( u_m \) and \( u_t \) the mutation rates in males and females, and \( h \) the selective disadvantage of heterozygous females (homozygous females and affected males have fitness 0). From equations 11 and 12 in Crow and Denniston\(^9\)

\[
q_m = (2u_t + u_m(1-h))/(1 + h)
\]
\[
q_f = (u_m + u_t)/(1 + h)
\]

Letting \( R = u_m/u_t \), the proportion of new mutants in males is

\[
u/m = (1+h)/(2 + R(1-h)) = 2/28.
\]

New mutants among heterozygous females is

\[
u = (u_m + u_t)/2q_f = (1+h)/2 = 12/15.
\]

Solving the two equations, \( h = 0.6 \) and \( R = 51 \), with a large standard error because of the small sample size.

Additional evidence comes from a study by Thomas,\(^9\) who noted that there are 13 X-linked diseases that are lethal or sterilizing in females. No males are found. The traditional explanation has been that the trait is a prenatal lethal in males, but the data are also consistent with a high male mutation rate. The reason is that affected females are new paternal mutations and, since they do not reproduce, there are no affected males. If this hypothesis is correct, it argues that the female rate is very low. Of course not all 13 diseases need have the same cause.

One of the 13 conditions has been analyzed, Rett Syndrome.\(^9\) Among 27 sporadic cases, 26 were paternal mutations. Since the female has two X chromosomes, either of which can mutate, the male/female mutation ratio, \( R \), is 2(26/1), or 52. Recent studies suggest that this interesting syndrome is a disorder caused by loss of imprinting.\(^11\)

With the coming of molecular methods it has become possible, using linked molecular markers, to determine the parent of origin for dominant autosomal mutations. A good illustrative example is retinoblastoma, since the disease requires two mutations, often one germinal and one somatic. This allows a comparison of the sex ratio for germinal and somatic mutations. Among 72 germinal mutations\(^13\) 85% were paternal, so \( R = 85/15 = 5.67 \). The somatic mutation rates, as expected, were about the same in the two sexes, showing that the excess male rate in germ cells is a germinal phenomenon and not an intrinsic property of the retinoblastoma gene.

Risch et al.\(^13\) have done an extensive survey of parental age for a variety of dominant traits. Usually there is a much larger paternal than maternal age effect, the latter mainly attributable to correlation between ages of mother and father. The paternal age effect is sometimes not significantly different from linear; in others the mutation rate accelerates with age. Whether the two groups differ significantly from each other is not clear.

How can we explain the sex difference and the paternal age effect? The most obvious explanation is the larger number of cell divisions in the germ cells prior to meiosis in the male than in the female; and the difference is greater in old males. According to Vogel and Motulsky\(^14\) (Figs. 9.13 and 9.14), there are 24 cell divisions from zygote to an ovum (23 chromosome replications, since there is only one in the two meiotic divisions). In the male, in contrast, there are 30 divisions prior to puberty (15 years), followed by stem cell divisions that occur 23 times per year, then 4 gonial divisions and two meiotic (one replication). Thus the number of chromosome replications from the zygote to a sperm produced by a man of age \( T \) is

\[
N_T = 30 + 23(T – 15) + 5
\]

At age 20, this is 150; at age 30, 380; at age 40, 610; and at age 50, 840. At age 30, the male/female ratio is 380/23 = 16.5.

In general the data – greater male than female mutation rate and increase with paternal age – agree with the cytological observations of pre-meiotic cell divisions in the two sexes. From the OTC and Rett data it appears that, although the number of cell divisions is linear throughout most of the pre-meiotic period, the increase in mutation rate is somewhat greater. Whether this is generally true is not yet clear.

EXCEPTIONS TO THE SEX AND PATERNAL AGE EFFECT

Duchenne muscular dystrophy is X-linked, but it shows a much smaller sex and grandpaternal age effect. This condition is caused by a very large gene with 55 exons. Among 198 mutations, 114 (57.6%) were deletions, 8 were duplications, and 76 were base substitutions.\(^15\) A similar study of Neurofibromatosis 1\(^17\) yielded 385/615 = 62.3% deletions. This is also caused by a very large gene and it too has a substantial fraction of deletions. In a survey of deletions of less than 20 bp in this and other large genes, Glaser and Jabs (personal communication) reported that among 847 cases of small deletions where the parental origin could be obtained, 392 or 46% were paternal. Rather than having a large male excess, the proportion in the two sexes is roughly the same. Furthermore, there is no significant age effect in either sex.

Both Duchenne muscular dystrophy and neurofibromatosis show a slight sex and paternal (or grandpaternal) age effect. This is caused by there being a mixture of base substitutions, which have a large effect, and deletions which have little or no such effect.

It is not known when the deletions occur. The similarity of the frequency in the two sexes and the absence of an age effect suggests that the event very likely happens only once in the life-cycle. Perhaps this is at meiosis; any of several mechanisms might be suggested. A study of X-linked loci might reveal, by a lesser frequency in males, that homologous exchange is involved. Some data suggest that this may be the case.\(^15\)

Mutations at most loci are a mixture of base substitutions.
and small indels, mostly deletions. From the data on the proportion of base substitutions, we can calculate the male/female ratio for base substitutions. Retinoblastoma is a large gene of 180 kb and would be expected to include deletions. Among 368 germinal mutations, 227 or 0.62 were base substitutions and 0.38 were indels.\textsuperscript{19} Let \( R_B \) = the male/female ratio for base substitutions. From the Glaser and Jabs review, the male/female ratio for deletions is 0.46/0.54 = 0.85. Note that these are small deletions (< 20 bp); curiously, large deletions occur preferentially in the male. The overall M/F mutation ratio is 5.67. So, using logs since we are dealing with ratios, we have the equation

\[
0.62 \log R_B + 0.38 \log 0.85 = \log 5.67
\]

which gives \( R_B = 18.1 \). The numerical value is uncertain because of limited numbers, but this illustrates the method.

If the average age of male reproduction is 30 years, the expected male/female ratio is 380/23 = 16.5. This suggests that the mutation rate is linear with the number of cell divisions. But, as mentioned earlier, the X-linked conditions, Rett and OTC, appear to show an accelerating age relationship. The data are too limited at this stage for any definitive conclusion as to the linearity or non-linearity of the paternal age effect; and is the relationship the same for all loci?

I would not find non-linearity surprising. I would expect that, with advancing age, various cellular mechanisms for mutation reduction – fidelity of transcription, proofreading, error correction – decay with age as does everything else (as I can say from personal experience).

In an analysis of the human database in 2000,\textsuperscript{20} consisting of 21,591 mutations in 1039 genes, base substitutions constitute 70\% and small indels 23\%, with gross indels making up most of the rest. Repeat expansions are less than 1\%, although this fraction might be larger if the study were done today. Altogether, we would expect a substantial sex and paternal age effect for the totality of dominant and X-linked mutations.

**HOT SPOTS**

Thus far, I have considered mutations that occur at multiple sites scattered along the chromosome; this is typical of most mutations. A few, such as those causing Achondroplasia, Apert syndrome, MEN 2A and 2B, and Crouzon and Pfeiffer syndromes, are hot-spot mutations; that is to say, the mutations occur at specific sites in one or two codons. All of these occur in three genes, FGFR3, FGFR2, and RET. The mutations occur almost exclusively in males and there is a striking increase with paternal age. The location of mutations at hot spots means that the mutation rate per nucleotide is extremely high. It is likely that, were it not for the extreme sex and paternal age effect, Weinberg might not have noticed the unusual distribution of birth orders; it is another example of a correct general conclusion reached from an extreme special case.

Achondroplasia is a mutation of the fibroblast growth factor receptor (FGFR3).\textsuperscript{21}\textsuperscript{22} The normal sequence in the region of interest is TAC GGG, coding for tyrosine and glycine. The most common mutation, a transition, converts GGG to AGG, leading to arginine. Considerably less common, as expected, is a transversion, converting GGG to CGG, also leading to arginine. Notice that these mutations occur at a CpG site, known to produce hotspots.

Apert syndrome causes a variety of symptoms, including misshapen head, fused fingers, and mental retardation.\textsuperscript{26} The mutations occur mainly at two adjacent codons in FGFR2, TCG coding for serine and CCT coding for proline. The transversions TCG \( \rightarrow \) TGG and CCT \( \rightarrow \) CGT both produce Apert syndrome.\textsuperscript{22} Mutations at another site in the FGFR3 gene produce Muenke-type craniosynostosis.\textsuperscript{23} This seems to be another hot spot. The mutations, C \( \rightarrow \) G transversions, are all at one nucleotide. Of 10 cases in which the parental origin could be determined, all were paternal and there was an elevated age of fathers.

**SPERM ANALYSIS**

I have long wished for confirmation of the clinical data by direct analysis of sperm. This is possible only by special techniques, such as using a restriction site at the appropriate position. Three laboratories have reported frequency data.\textsuperscript{22,24,25} The third reference gave results that provided no support for an increase with age. I suspect that the difference is technical, since the measurements are at the very edge of sensitivity of the technique employed. The first reference, from Wilkie’s laboratory, made use of a 4-cutter restriction enzyme that cuts between C and G at the serine codon. The sperm data agree well with the age distribution in the clinical observations (Fig. 1). The data of Tiemann-Boege et al.\textsuperscript{24} on achondroplasia can be interpreted as in rough agreement.\textsuperscript{26}

| Relative frequency | Age (years) |
|-------------------|-------------|
| 1.0               | 22          |
| 2.0               | 32          |
| 3.0               | 42          |
| 4.0               | 52          |

**Fig. 1.** Relative frequency (ordinate) of Apert Syndrome in children of fathers of different ages (abscissa). The smooth line is the best fitting exponential curve. From Risch et al.\textsuperscript{13}
Why is the mutation frequency so enormous? Wilkie and his colleagues suggest that, rather than a high mutation rate, there is pre-meiotic selection in the germ-line for the heterozygous mutation. A small selection differential is sufficient to generate a high apparent mutation rate and can mimic the paternal age data in Fig. 1. Premiotic selection in favor of a mutation that is highly deleterious in somatic tissue is very surprising, a priori; in fact, the conventional wisdom is that there is no selection in the germ line. Nevertheless, let us pursue the idea.

As a simple, but instructive, example, assume that the number of stem-cell lineages is such that one mutation occurs each year, not in the same lineage. In the stem-cell stages the divisions are asymmetrical. One daughter cell is discarded and the other continues the line. Thus, the mutations accumulate linearly, 1 at age 0, 11 at age 10, and 41 at age 40, all measured as years after puberty. Therefore by 40 years after puberty (55 years from birth), 41 mutations will have accumulated.

To consider selection, assume that the mutant cells have an intercellular survival advantage of $x$ per cell generation, or $23x = s$ per year. It is convenient to count back from time $T$. At age $T$ one mutation will occur; at age $T-1$ there will have been one mutation that now is increased to $(1 + s)$; at age $T-2$ there will be a mutation that now has a frequency $(1 + s)^2$, and so on (Fig. 2). The accumulated number of mutations $T$ generations after puberty will be

$$N_T = 1 + (1+s) + (1+s)^2 + \ldots + (1+s)^T = [(1+s)^{T+1} - 1]/s.$$

For $x = 0.002$ ($s = 0.046$), the number of mutants at successive 10-year periods will be 1, 14, 34, 66, 116, 217, 378, and 643, as seen in Fig. 3. This curve mimics the shape in Fig. 1. This of course is only a rough agreement; the data are not up to a more exact treatment. The point is that very weak intercellular selection, a selective advantage of 0.002, is sufficient to account for the clinical observations.

Recall that, by age 30, 90% or more of the cell divisions occur in stem-cell stages where the cell divisions are asymmetric. Within a stem-cell line, it makes no difference when the mutation occurred; there will be the same number at the end of the line (assuming, reasonably, that at most one mutation occurs in a given stem-cell lineage).

In contrast, with symmetric division leading to exponential growth, a mutation that occurs early will have many more descendants than a mutation that occurs later. This leads to a “jackpot” distribution, with an enormous difference in the number of descendants per mutant. It is similar to the Luria-Delbrück distribution of mutant cells in a growing bacterial culture.\(^\text{27}\)

With symmetric division mutant clones are expected to differ enormously in size. This will be reflected in a greatly increased variance in the number of mutant sperm per man. This was observed by the Wilkie group; the variance was enormously greater than the binomial or Poisson variance that would be expected with mutations in stem cells. (There is some exponential cell growth prior to puberty and again just before meiosis, but these constitute a small fraction of the cell divisions, not enough to account for the observed extremely high variance.)

Wilkie’s group did an additional test.\(^\text{22}\) Luckily, many of the men were heterozygous for an A/G SNP very close to the codon of interest. The mean ratio of A to G in the collection of sperm was 1:1, as expected. But the variance in the A:G ratio among men was enormous, adding to the evidence for selection.

There is additional evidence for selection.\(^\text{22,28}\) (a) The observed rate of $C \rightarrow G$ is higher than $C \rightarrow T$, despite the former being a transversion. This makes sense if we assume that the $C \rightarrow T$ mutation rate is higher, but the selective
advantage is less. (b) The variance is greater for the C → G than for the C → T change, as expected if selection is more intense for the former. (c) C → A produces a stop codon; it leads to no substantial increase in mutation numbers. (d) Other mutations in the two codons do not appear to produce a selective advantage; some are synonymous, others are not expected to enhance the FGF binding affinity. The average mutation number of these is about 20 fold less than those for which selection is postulated. (e) The two mutations with high frequency in sperm show no such increase in blood, where the postulated selective mechanism would not occur. (f) Several examples of sequential double mutations have been found, which would be extremely improbable if mutation were the only mechanism.

POSSIBLE SELECTIVE MECHANISMS

Figures 2 and 3 say nothing about the selective mechanism. Furthermore, they do not reflect the enormous increase in variance that was observed. Such an enormous variance in mutant frequency and SNP ratio among fathers implies symmetric cell division and exponential growth (the Luria-Delbrück effect). At present there is no suggestion as to how an asymmetric stem cell division pattern can be converted to a symmetric one. If a stem cell at the time of mutation were suddenly to start multiplying exponentially at the same cell division rate, the effect would be too large for the observed data. So we need to postulate some sort of mechanism that only partially converts the division pattern from asymmetric to symmetric. Perhaps symmetric divisions are interspersed among asymmetric ones. I note that selection favoring the mutant cells does not necessarily imply that the mutant cells divide faster; they might even divide more slowly. The mechanism of selection could be, rather than a different cell division rate, a conversion from asymmetric to symmetric division. But, as I said the conversion must somehow be only partial or occasional, else the mutant cells far outstrip the normal cells.

At present, there is no clue as to how such a process might work, although several kinetic models are possible. It is indeed a fascinating problem. It may presage an entirely new mechanism for pre-meiotic selection.

PRE-MEIOTIC SELECTION, MEIOTIC DRIVE, AND GAMETE COMPETITION

If there is indeed selection, why should a mutation that causes devastating disabilities in the individual confer an advantage in the germ line? Perhaps the fact that the mutation is a gain of function means that the new function can have a different result in germ and soma; what is good for the germ line may not be for somatic tissue. Cancer cells provide a precedent, perhaps relevant. Alternatively, the phenomenon might somehow be restricted to cells with stem-cell kinetics.

From the population standpoint, premeiotic selection, whatever the mechanism, is of great interest. The consequences are much the same as those of gametic selection or meiotic drive. Any of these is a mechanism by which somatically deleterious alleles are kept at a higher frequency in the population than can be expected from mutation-selection balance. Too many such alleles would be disastrous. As long ago as 1932, Haldane (p 123) wrote “Clearly a higher plant species is at the mercy of its gametes. A gene which greatly accelerates pollen tube growth will spread through a species even if it causes moderately disadvantageous changes in the adult plant.” He was discussing plants, but the statement applies equally to animals.

A recent paper argues that mild transmission distortion is common in human populations. This is not likely to explain the hot-spot problem, since it offers no explanation of the paternal age effect. Meiotic drive also seems unpromising, since Apert heterozygotes produce the two kinds of sperm in the canonical 1:1 ratio. However, this need not imply that the two kinds are equally functional.

Mendelian 1:1 segregation appears to be well protected by selective mechanisms. Independent modifiers of the segregation ratio will increase if they bring the ratio closer to 1:1. Large distortions of segregation ratios are rare; there are Drosophila and mouse examples but none in the human species. But very minor departures from a 1:1 ratio might be found at various loci without ordinarily being detected. This is indeed what some data seem to show.

Pre-meiotic selection seems the best bet. If the results of Goriely et al. are confirmed, they will have opened a new direction for research in human population genetics, with possible broad implications for other long-lived species.

CONCLUSION

The data collectively suggest that there are three main classes of Mendelian mutations. More than one can occur in the same gene.

1. Small deletions and duplications (< 20 bp)
   - No significant age effect
   - Roughly equal numbers of paternal and maternal origin

2. Base substitutions
   - Mainly, but not entirely paternal
   - Large paternal age effect, which may be nonlinear

3. Hot spots
   - Essentially all paternal
   - Very large, nonlinear paternal age effect
   - May be due to selection, rather than high mutation rate

Usually those base substitutions with a moderate sex and age effect occur at many sites. For example, in mild X-
Complex Conditions

A number of human conditions with complex inheritance show a paternal age effect. Usually the effect is much less than for single-gene dominant or X-linked mutations, but since many studies are based on very large populations, a small average paternal age increase – months rather than years – is statistically significant. An early example is Ols-han’s study of congenital heart defects. Relative to age 15–29, those over 50 have an increased risk of 2 or 3 fold.”

A striking and puzzling example is schizophrenia. Malaspina summarized data from a very large study done in Israel, involving 87,500 births. The relative risk according to paternal age is

| Paternal Age | Relative Risk |
|--------------|---------------|
| < 30         | 1.00          |
| 30–34        | 1.20          |
| 35–39        | 1.56          |
| 40–44        | 1.79          |
| 45–49        | 1.89          |
| ≥ 50         | 2.60          |

One might suspect that schizophrenic men, because of personality problems, might tend to marry and reproduce late. But the data do not support this as an explanation. The families were classified, admittedly crudely, into those with at least one affected in the extended family and those without. The paternal ages were, respectively, 29.2 and 33.9. Since sporadic cases are more likely than familial cases to be new mutations, this supports mutation as the cause.

Several other studies support the general conclusion qualitatively. Yet, the sheer magnitude of the effect invites skepticism. Taken at face value, the Malaspina data suggest that a large fraction of the incidence of schizophrenia is due to new dominant mutations. A priori, this seems very unlikely. We shall have to wait for further studies to unravel this tangle.

This does, however, suggest a strategy for finding mutations affecting complex diseases. One could select newborns or young children whose father was exceptionally old. This would constitute an enriched sample, with increased likelihood of having a new mutation. Then one could look for DNA differences between father and child. In this way one might hope to find component genes for traits with complex etiology.

Evidence from Another Source, Molecular evolution

Kimura has argued convincingly that a great deal of molecular evolution is selectively neutral and is driven by mutation. Noncoding regions, pseudogenes, and third codon positions are usually neutral, or nearly so. Their speed of evolution should be a direct reflection of the mutation rate. Miyata was the first to apply this idea. He compared the rate of evolution of arginosuccinate pseudogenes on the Y chromosome and chromosome 7 in human ancestry.

The theory is simple. Let M be the male mutation rate and F the female. Then the rates on different chromosomes are:

- Y chromosome: \( \frac{M}{3} \)
- Autosome (A): \( \frac{M + F}{2} \)
- X chromosome: \( \frac{2F + M}{3} \)

From this, letting \( R = \frac{M}{F} \), \( Y/X = 3R/(R + 2) \) and \( Y/A = 2R/(R + 1) \). The observed value of \( Y/A \) was 2.2, not significantly different from 2, implying that \( R \) is very large. From more extensive data, in primates \( R \) is in the range 4–6. Phylogenies that split farther back are more reliable, since they are less likely to be biased by the existence of polymorphisms before the split. A polymorphism means that the method measures the time to the origin of the polymorphism rather than the split in the lineage. This can bias the estimate of \( R \) for the following reason. The Y chromosome has 1/4 the effective population number of an autosomal (1/2 because of haploidy and 1/2 because of being in only one sex). Thus there is more random drift and less polymorphism for Y-linked loci. Hence, polymorphism causes an underestimate of \( R \) from \( Y/A \) comparisons.

Studies of the most distantly related primates give \( R \) values around 5–6. This value is considerably less than the ratio as estimated from human diseases. There are two reasons for this. One is that the evolutionary data include other primates with shorter reproductive life spans. A second reason is that these phylogenies go far back; the human reproductive age has increased considerably in the meantime. So, although there may be quantitative discrepancies, male-driven evolution agrees qualitatively with what would be expected.
Absolute Mutation Rate

The total mutation frequency, as inferred from evolutionary studies, is something of the order of 100 new mutations per generation. This seems utterly frightening. Surely, the overwhelming majority of these must be essentially neutral. More relevant is the frequency of new deleterious mutations, which is more than one per zygote. That is still high. Why aren’t we extinct?

I think the answer lies in the effectiveness of sexual reproduction in reducing the mutation load. With epistasis, recombination permits elimination of harmful mutations in groups. This is not possible with asexual reproduction. Truncation selection, in which all the individuals with more than a threshold number of mutations are eliminated mimics extreme epistasis and is known to be especially efficient. Strict truncation is not required; a very crude approximation is almost as effective. I suspect that something like this operated in our resource-limited, hunter-gatherer ancestors, permitting them to tolerate a mutation rate that in an asexual species would be fatal.

We are clearly out of equilibrium between mutation and selection – if our ancestors ever were at equilibrium. Selection is much relaxed in recent times, at least in affluent societies. Much of the selection that does occur, such as by differences in family sizes, is irrelevant to reducing the mutation load. Adjustment of mutation rates to conform to an increased age of reproduction in our recent evolutionary past is surely a very slow process. For all these reasons, mutations are almost certainly accumulating at a faster rate than in the past.

Is this a problem? Surely it will be eventually, but probably not immediately. Most mutations are mild and the time scale is several generations – hundreds or thousands of years. Furthermore, it is not obvious how to deal with the situation. The problem can be alleviated by continual environmental improvement, but we have little idea how much longer this can continue? And how likely is it that wars or environmental catastrophes will force us back toward an early, tougher environment in which accumulated mutations that are mild in the current environment become severe? Fortunately, we have time to learn more before the problem becomes pressing. Meanwhile, we can expect increasing control over single-gene mutations.

Sexual reproduction is one reason, perhaps the main one, why our ancestors were able to delay the reproductive age. There are clear advantages to a long childhood and adolescence. It permits greater time for growth and for learning and teaching. I think we may owe this luxury to our sexual mode of reproduction. In any case it would be very difficult to go back to asexual reproduction, even if we wanted to. One reason, but not the only one, is imprinting.

Now and in the near future we have other problems – global warming, water shortage, loss of habitat, famine, war, and population growth (last year the world population surpassed the number of nucleotides in a single cell). I hope that we can solve at least some of these problems. Then perhaps we can have the luxury of worrying about the mutation rate.

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REFERENCES

1. Weinberg, W. (1912) Zur Vererbung des Zwergwuchses. Arch. Rassen-u. Gesel. Biolog. 9: 710–718.
2. Crow, J. F. (1999) Hardy, Weinberg and language impediments. Genetics 152: 821–825.
3. Penrose, L. R. (1955) Parental age and mutation. Lancet 269: 312–313.
4. Haldane, J. B. S. (1947) The mutation rate of the gene for hemophilia and its segregation ratios in males and females. Ann. Eugen. 13: 262–271.
5. Oldenburg, J., Schwaab, R., Grimm, T., Zerres, K., Hakenberg, P., Brackmann, H. H., Olek, K. (1993) Direct and indirect estimation of the sex ratio of mutation frequencies in hemophilia A. Amer. J. Hum. Genet. 53: 1229–1238.
6. Francke, U., Felsenstein, J., Gartler, S. M., Migeon, B. R., Dancis, J., Seegmiller, J. E., Bakey, F., Nylan, W. L. (1976) The occurrence of new mutants in the X-linked recessive Lesch-Nyhan disease. Amer. J. Hum. Genet. 28: 123–137.
7. Tuchman, M., Plante, R. J., Garcia-Perez, M. A., Rubio, V. (1996) Relative frequency of mutations causing ornithine transcarbamylase deficiency in 78 families. Human Genet. 97: 274–276.
8. Crow, J. F., Denniston, C. (1985) Mutation in human populations. Adv. Human Genet. 14: 59–123.
9. Thomas, G. H. (1996) High male:female ratio of germ-line mutations: An alternative explanation for postulated gestational lethality in males in X-linked dominant disorders. Amer. J. Hum. Genet. 58: 1364–1368.
10. Trappe, R., Laccone, F., Cobilanschi, J., Meins, M., Hupke, P., Hanefeld, F., Engel, W. (2001) MECP2 mutations in sporadic cases of Rett Syndrome are almost exclusively of paternal origin. Amer. J. Hum. Genet. 68: 1093–1101.
11. Pescucci, C., Meloni, I., Renieri, A. (2005) Is Rett syndrome a loss-of-imprinting disorder? Nature Genet. 37: 10–11.
12. Dryja, T. P., Morrow, J. F., Rapaport, J. M. (1997) Quantification of the paternal allele bias for new germine mutations in the retinoblastoma gene. Human Genet. 100: 446–449.
13. Risch, N., Reigh, E. W., Wishnick, M. W., McCarthy, J. G. (1987) Spontaneous mutation and parental age in humans. Amer. J. Human Genet. 41: 218–248.
14. Vogel, F., Motulsky, A. (1997) Human Genetics; Problems and Approaches. Springer Verlag, Berlin.
15. Grimm, T. G., Meng., S., Liechti-Gallati, S. et al. (1994) On the origin of deletions and point mutations in Duchenne muscular dystrophy: most deletions arise in oogenesis and most
point mutations result from events in spermatogenesis. J. Med. Genet. 31: 183–186.
16. Zatz, M., S. Cumita, D., Campiottto, S., Canovas, M., Cerqueira, A., Vainzof, M., Passos-Bueno, M. R. (1998) Paternal inheritance of different mutations in maternally related patients occur in about 3% of Duchenne familial cases. Amer. J. Med. Genet. 78: 361–365.
17. Lazaro, C., Gaora, A., Ainsworth, P., Tenconi, R., Vidaud, D., Kruyer, H., Ars, E., Volpiri, V., Estivill, X. (1996) Sex differences in mutational rate and mutational mechanism in the NF1 gene in neurofibromatosis type 1 patients. Human Genet. 98: 696–699.
18. Dryja, T. P., Morrow, J. F., Rapaport, J. M. (1997) Quantification of the paternal allele bias for new germline mutations in the retinoblastoma gene. Human Genet. 100: 446–449.
19. Lohmann, D. R. (1999) RB1 gene mutations in retinoblastoma. Amer. J. Human Genet. 64: 283–288.
20. Antonarakis, S. E., Krawczak, M., Cooper, D. N. (2000) Disease-causing mutations in the human genome. Eur. J. Pediat. 159: S173–S178.
21. Bellus, G. A., et al. (1995) Achenondroplasia is defined by recurrent G380R mutations of FGFR3. Amer. J. Human Genet. 58: 657–670.
22. Goriely, A., McVean, G. A. T., Rojmyr, M., Ingemarsson, B., Wilkie, A. O. M. (2003) Evidence for selective advantage of pathogenic FGFR2 mutations in the male germ line. Science 301: 643–646.
23. Rannan-Eliya, S.V., Taylor, I. B., de Heer, I. M., van den Ouweland, A. M. W., Wall, S. A., Wilkie, A. O. M. (2004) Paternal origin of FGFR3 mutations in Muenke-type craniosynostosis. Hum. Genet. 115: 200–207.
24. Tiemann-Boege, L., Navidi, W., Grewal, R., Cohn, D., Eskenazi, B., Wyrobek, A. J., Arnheim, N. (2002) The observed human sperm mutation frequency cannot explain the achenondroplasia paternal age effect. Proc. Natl. Acad. Sci. USA 99: 14952–14957.
25. Glaser, R. L., Broman, K. W., Schulman, R. L., Eskenazi, B., Wyrobek, A. J., Jabs, E. W. (2003) The paternal-age effect in Apert syndrome is due, in part, to the increased frequency of mutations in sperm. Amer. J. Human Genet. 73: 939–947.
26. Wilkie, A. O. M. (2005) Bad bones, absent smell, selfish tests: the pleiotropic consequences of human FGFR receptor mutations. Cytokine Growth Factor Rev. 16: 187–203.
27. Luria, S. E., Delbrück, M. (1943) Mutations of bacteria from virus sensitivity to virus resistance. Genetics 28: 491–511.
28. Goriely, A., McVean, G. A. T., van Pelt, A. M. M., O’Rourke, A. W. O., Wall, S. A., de Rooij, D. G., Wilkie, A. O. M. (2005) Gain-of-function amino acid substitutions drive positive selection of FGFR2 mutations in human spermatogonia. Proc. Natl. Acad. Sci. USA 102: 6051–6056.
29. Haldane, J. B. S. (1932) The Causes of Evolution. Longmans, Green and Company, London.
30. Zollner, S., Wen, X., Hanchard, A., Herbert, M. A., Ober, C., Pritchard, J. K. (2004) Evidence for extensive transmission distortion in the human genome. Amer. J. Human Genet. 74: 62–72.
31. Moloney, D. (2001) What can we learn about mechanisms of mutation from a study of craniosynostosis? Ann. Rev. Coll. Surg. Engl. 83: 1–9.
32. Crow, J. F. (1991) Why is Mendelian segregation so exact? Bioessays 13: 305–312.
33. Leith, E. G. (1987) Ronald Fisher and the development of evolutionary theory. II. Influences of new variation on evolutionary process. Oxford Series in Evol. Biol. 4: 212–263.
34. Eshel, I. (1985) Evolutionary genetic stability of Mendelian segregation and the role of free recombination in the chromosomal system. Amer. Natur. 125: 412–420.
35. Becker, J., Schwaab, R., Möller-Taube, A., Schwaab, U., Schmid, W., Brackmann, H. H., Grimm, T., Olek, K., Oldenburg, J. (1996) Characterization of the factor VIII defect in 147 patients with sporadic hemophilia A. Family studies indicate a mutation type-dependent sex ratio of mutation frequencies. Amer. J. Human Genet. 58: 657–670.
36. Tartaglia, M., Corbeddu, V., Chang, H., Shaw, A., Kalidas, K., Crosby, A., Patton, M. A., Sorcini, M., van der Brigt, I., Jeffery, S., Gelb, B. D. (2004) Paternal germline origin and sex ratio distortion in transmission of PTPN11 mutations in Noonan syndrome. Amer. J. Human Genet. 75: 492–39.
37. Hassold, T., Abrauzzo, M., Adkins, K., et al. (1996) Human aneuploidy: Incidence, origin, and etiology. Environ. Mol. Mutag. 28: 167–175.
38. Antonarakis, J. P. et al. (1995) Factor VIII gene inversions in severe hemophilia: Results of an international consortium study. Blood 86: 2206–2212.
39. Olshan, A. F., et al. (1994) Paternal age and the risk of congenital heart defects. Teratology 50: 80–84.
40. Malasaina, D., Harlap, S., Heiman, D., Nahon, D., Feldman, D., Susser, E. S. (2001) Advancing paternal age and the risk of schizophrenia. Arch. Gen. Psychiat. 58: 361–367.
41. Kimura, M. (1983) The Neutral Theory of Molecular Evolution. Cambridge Univ. Press, Cambridge.
42. Miyata, T., Hayashida, H., Kuma, K., Mitsuysasu, K., Yasunaga, T. (1987) Male-driven molecular evolution demonstrated by different rates of silent substitutions between autosomal and sex chromosome-linked genes. Proc. Japan. Acad. B 63: 327–331.
43. Makova, K. D., Li, W. H. (2002) Strong male-driven evolution of DNA sequences in humans and apes. Nature 416: 624–626.
44. Nachman, M. W., Crowell, S. L. (2000) Estimate of the mutation rate per nucleotide in humans. Genetics 156: 297–304.
45. Eyre-Walker, A., Keightley, P. D. (1999) High genomic deleterious mutation rates in hominids. Nature 397: 344–347.
46. Crow, J. F. (2000) The origins, patterns and implications of human spontaneous mutation. Nature Rev. Genet. 1: 40–47.
47. Crow, J. F. and Kimura, M. (1979) Efficiency of truncation selection. Proc. Natl. Acad. Sci. USA 76: 396–399.