

February 26, 1982

Dr. Howard B. Hamilton  
Chief, Clinical Laboratories  
Radiation Effects Research Foundation  
5-2 Hijiyama Park, Minami Ward  
Hiroshima City 730  
JAPAN

Dear Howard:

Many thanks for your letter of February 8. I have been thinking on and off about your problem and have discussed it from time to time with several people who might prove to be helpful. It seems to me that the most conservative plan that could be carried out immediately is to try to collect 50 mls. of blood from the more heavily irradiated subjects and their progeny and to store frozen cell pellets for later extraction of DNA. Alternatively and preferably, the DNA could be made directly from fresh cells and saved until personnel and facilities were available for making phage libraries. The overall objective would be to compare DNA from  $F_1$ s with the DNA of the two parents. One could anticipate detecting at least two kinds of lesions. First, base changes which are perceived as restriction site polymorphisms; these might be best detected by intensive restriction mapping of domains of the genome which have already been extensively studied. As an example of such a region, you might look at the view by Maniatis in last year's Annual Review of Genetics. Each restriction site is encoded by four, five or six bases in general, so you can see that by surveying fifty or a hundred restriction sites (a quite simple task once the library is made) that one gets a very rapid sense of how frequent point mutations occur or have occurred in the germ line at random without respect to whether the base change produces a detectable change in a gene product. The second kind of test that might prove more profitable in view of the presumed nature of radiation induced lesions would be to screen several genetic domains for deletions or rearrangements. This could be done either using well characterized genetic domains such as the one for globin or by selecting isolated clones of human DNA at random and comparing restriction of the nucleus maps of the DNA from the two parents and the  $F_1$ . In this case unusual restriction fragments will be observed using several restriction enzymes reflecting the basic alteration and the structure of that region of the genome rather than a simple base change that affects a single restriction site.

There is notdoubt that this approach presents an appreciable amount of work. However, there are at least three reasons that I can see for making some first steps toward taking it. First, the procedures for making DNA libraries have gotten a good deal simpler in the last year or two and can be expected to become very simple within the next few years. Secondly, it seems inherently preferable to me to do extensive explorations of radiation effects in a small number of subjects who have been exposed to extremely high doses of radiation ~~and their progeny~~. Thirdly, this population which you are dealing with is a vanishing resource and the storage of cells or DNA would provide some insurance against the loss of this experimental resource even if the cloning and subsequent analysis could not be carried out until some later date.

One item of interest which I didn't explore with you while I was there is whether there appears to be any increased incidence in cancer among the F<sub>1</sub>s of highly exposed victims. Any evidence for creation of or induction of an oncogene would obviously spark heightened interest in this project among several people here. Next week I shall be seeing two of the people who are prominent in efforts to map the human genome, Ray White and David Botstein, and I plan to discuss the situation with them and seek their suggestions. I will let you know if I have any further ideas.

With best regards,

Harold E. Varmus, M.D.  
Professor

HEV/jm