

The Colonel Harlan D. Sanders Award Address for 1998

JC Virus and Its Possible Role in Oncogenesis

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INTRODUCTION

First, let me express the honor I feel at being chosen this year's recipient of the Colonel Harlan D. Sanders Award of the March of Dimes Birth Defects Foundation. Looking at the list of past recipients, I would have to say that it has quickly become one of the most distinguished awards in the field of medical genetics, and I am delighted to be this year's awardee. The notification of the award carried with it the statement that it was customary in the acceptance talk to make some brief reference to the accomplishments for which the recipient was being recognized. But knowing that I was to be introduced by John Opitz, I found myself in something of a quandary. John is so thorough and so encyclopedic when the history of human genetics and its players are the subject, that I feared he would not leave me a great deal of running room for my own little talk. I might say, John has just lived up to my every expectation. But you know what, I think I have outfoxed John. I've chosen to talk briefly about some very recent developments, some unpublished, which illustrate not only how serendipity enters into our research lives but as well the unexpected twists and turns in the pursuit of a scientific problem.

Actually, although most of the developments I will discuss are quite recent, the initial step in these developments was taken in 1969. At that time, I was in my Amerindian phase, concerned with a largish multidisciplinary study on the American Indian. This involved numerous trips into the tropical rain forests of Central and northern South America, to work among some of the least acculturated of the surviving Amerindian tribes. Each of our expeditions had a slightly different agenda. For the expedition of 1969, Arthur Bloom, then the cytogeneticist of our department, agreed to do cytogenetic studies if we would collect the proper blood samples. As you all know, roughly one percent of the cultured lymphocytes of members of industrialized

populations exhibit gross chromosomal damage. It was our expectation that these clean-living Amerindians, far from the pollutants that characterize our society, would be found to exhibit a much lower frequency of chromosomal damage.

For the subjects of our cytogenetic study, we chose the Yanomama, then one of the more isolated and least acculturated of all the tribes of South America. But because there were already some contacts with this tribe, especially about its periphery, we chose to do our sampling in their heartland, the Parima Mountain Range, on the boundary between Venezuela and Brazil. This was possible because in one part of the range the tropical rain forest gives way to an undulating piece of savannah on which a small plane, careful to avoid the termite mounds, can land. The commitment of the very skilled pilots of the Mission Aviation Fellowship included servicing a very small Unevangelized Tribes Mission in that area, and Paul Johnson, the pilot for the Upper Orinoco region, agreed to try to put us down near a cluster of Yanomama villages. Since I'm talking to you today, he was obviously successful, and the team spent a busy 10 days on location collecting, among other activities, the blood samples for cytogenetic studies.

THE DISCOVERY OF "ROGUE CELLS"

To our surprise, back in the laboratory, 23 among a total of 4,969 cells scored showed a picture of extreme cytogenetic damage [Bloom et al., 1970]. We later termed these abnormal cells "rogue cells," now arbitrarily defined as cells containing five or more exchange-type aberrations for which precise karyotypic identification of the origin of the aberrant chromosomes was usually impossible [Awa and Neel, 1986]. No such cells were observed among 2,575 cultured lymphocytes from a second tribe studied at the same time, the Piaroa [Bloom et al., 1970]. In 1970, we attempted to repeat this observation in two additional Yanomama villages, with limited success (two rogue cells among 5,654 cells scored) [Bloom et al., 1973]. Accordingly, in 1971, we returned to one of the two Yanomama villages studied earlier. This time, only one among 4,917 cells scored was a rogue cell [Bloom et al., 1973]. I can assure you that at this point, there was considerable skepticism among our colleagues concerning the valid-

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ity of our observation. However, within the next decade similar cells were reported in cytogenetic studies of selected populations in England, Japan, and the former Soviet Union [Awa and Neel, 1986; Bochkov and Katosova, 1994; Fox et al., 1984; Lazutka, 1996; Neel et al., 1992; Salomaa et al., 1997; Scheid et al., 1993; Sevan'kaev et al., 1993; Tawn et al., 1985; Verschaeye et al., 1993] albeit, with one exception [Neel et al., 1992], never with a frequency approaching the original observation.

LINKING ROGUE CELLS TO THE JC VIRUS

We have returned to the problem of the nature and cause of these cells in a serious way in the past half-dozen years. Because the simian polyoma virus 40 (SV40) had been shown to produce similar cytogenetic damage in cultured human fibroblasts [Nichols et al., 1985; Ray and Kraemer, 1993; Ray et al., 1990, 1992; Stewart and Bacchetti, 1991], the possible role in this phenomenon of two well-known human polyoma viruses, the JC virus (JCV) and the BK virus (BKV), was investigated [Neel et al., 1996]. In a collaboration with Drs. Eugene Major and Thomas Glover, it was demonstrated that antibody titers against these two viruses were significantly elevated in persons in whom rogue cells were detected, the anti-JCV titers more so than the anti-BKV titers. Furthermore, inoculation of cultured human fetal brain cells with JCV produced chromosomal damage in the early post-inoculation cell divisions similar to that produced by SV40 in the early divisions of inoculated cultured human fibroblasts. (JCV has been demonstrated to be the agent responsible for the progressive multi-focal leucoencephalopathy of the acquired immunodeficiency syndrome (AIDS), and cultured human fetal brain cells were employed in this study because this preparation is currently the substrate of choice in the culture of JCV.) On the basis of these observations, we hypothesize that a newly acquired infection with JCV (or, less likely, BKV) or a flare-up of an existing infection, was at least one cause of the appearance of rogue cells in the peripheral circulation [Neel et al., 1996]. (Throughout the rest of this presentation, whenever I postulate effects for JCV, those effects, for all that is known, usually might equally well be characteristic of BKV.)

ROGUE CELLS AND "SIMPLE" CHROMOSOME DAMAGE

The typical rogue cell is chromosomally so complexly damaged that it could not be expected to complete a successful mitotic cell division very often. Most rogue cells must be essentially dead-end cells. However, from the first we have been interested in the possibility that "simple" chromosome damage was also elevated in the lymphocytes of persons exhibiting rogue cells. Such simple damage consists of stable translocations and inversions, and unstable multicentric chromosomes, free fragments, centric and acentric rings, and double minutes, and when observed usually involves only one or two chromosomes per cell. There are extensive reports of such simple stable chromosomal rearrangements resulting in clones that play a significant role in onco-

genesis [reviews in Heim and Mitelman, 1995; Rowley, 1996; Sandberg, 1990]. In our own past studies, the evidence that an elevation of this baseline might be characteristic of persons exhibiting rogue cells has been somewhat erratic. Thus, in the original Yanomama study, $4.10 \pm 0.28\%$ of all non-rogue cells scored (200 in 4,875 cells) showed simple damage of the types enumerated, whereas in follow-up studies in one of the same villages 2 years later, when the frequency of rogue cells had fallen to 0.01% (one in 9,849 cells), the corresponding percentage was $1.30 \pm 0.13\%$ (128 in 9,849 cells) [Bloom et al., 1970, 1973]. Although this difference is highly significant ($\chi^2 = 117.62$, P (one-tailed) < 0.0001), the validity of the comparison is diminished by the 2-year interval between the observations. Among the eight persons exhibiting rogue cells in a Ukrainian village, the frequency of simple damage was $1.52 \pm 0.30\%$ (24 among, 1,580 cells scored), whereas in the 16 persons not found to exhibit rogue cells, the corresponding figure was $1.03 \pm 0.18\%$ (33 of 3,200 cells scored) ($\chi^2 = 2.14$, P (one-tailed) = 0.095) [Neel et al., 1992]. Most recently, with the collaboration of Dr. A.A. Awa, we have re-examined the data from a study on Japanese residing in Hiroshima, whose original objective was to determine the cytogenetic effects of the atomic bombs [Awa et al., 1971, 1978, 1987]. Among a total of 1,835 persons examined, there were 45 exhibiting one or more rogue cells. A total of 179,599 cells was scored for simple chromosomal damage. In the exposed and the control populations, there was an absolute increase of approximately 1.5% in the frequency of simple chromosomal damage in those persons exhibiting rogue cells when compared with the frequencies observed in those not exhibiting rogue cells, a statistically quite significant difference [Neel, 1998].

Two observations by other groups are important in this respect. From the various studies of Tawn and associates on English persons [Tawn, 1987; Tawn and Binks, 1989; Tawn et al., 1985], one can conclude that in the control individuals in their series, the frequency of cells with asymmetrical exchange-type aberrations (i.e., dicentrics and centric rings) was 12 in 1,141 non-rogue cells in individuals in whom rogue cells were observed (three persons), but 25 in 16,550 individuals in whom rogue cells were not observed (114 persons) ($\chi^2 = 41.4$, d.f. = 1, $P < 0.001$). Lazutka [1996], in a study of chromosome aberrations in persons residing in Lithuania who had been involved in the Chernobyl clean-up operation, plus suitable controls, found that for the total sample, the frequency of simple damage (dicentrics, rings, translocations, inversions, and chromatid breaks) was $3.57 \pm 0.39\%$ in the 31 persons exhibiting rogue cells but $3.40 \pm 0.16\%$ in the 179 persons in whom rogue cells were not observed, the difference clearly non-significant but in the same direction as in the other studies. The relatively high frequency of cells with damage in both groups reflects the radiation exposures sustained by the clean-up workers. Considering the consistency of the finding, and its magnitude in some populations, we conclude that simple chromosomal damage is increased in the non-rogue cells of persons exhibiting rogue cells. In this connection, I note that

the so-called big T antigen produced by JCV has high homology with the big T antigen of SV40, and the latter is well known to function as a helicase [reviewed in Fanning and Knippers, 1992]. Thus, there is an established basis for the postulated clastogenic effects of JCV.

THE FREQUENCY AND GEOGRAPHY OF JCV AND BKV INFECTION

Numerous studies have shown that in urbanized, industrialized populations, significant hemagglutination antibody titers to JCV and BKV (titers $\geq 1/40$) are observed in some 80% of all adults [reviewed in Walker and Frisque, 1986]. In contrast, in isolated and unacculturated populations, as of Amerindians, the frequency of seropositives is much lower, in some tribes being zero [Brown et al., 1975; Candeias et al., 1977]. Among the Yanomama, in whom we first detected rogue cells, the frequency of positives was 29.5% in a sample of Indians with an estimated average age of about 20 years, whereas in a sample of young, urban Japanese, average age 23.9 ± 4.5 years, the frequency of positives was 62.0%. The frequency of positive responders appears lower in the least acculturated South American Indians as compared with the most acculturated (E.O. Major and J.V. Neel, unpublished manuscript), and it may be that the relatively high frequency of rogue cells encountered in the Yanomama reflects the impact of viral activity on a relatively virgin population.

EPIDEMIOLOGY AND LIFE HISTORY OF JCV

The epidemiology of JCV is still very poorly understood. On the basis of the sequencing of a 610-bp sequence from the VT-intergenic regions of the virus, JCV can be subdivided into nine major subtypes and many more minor types [Sugimoto et al., 1997]. Comparing these types in parents and children, Kitamura et al. [1994] and Kunitake et al. [1995] conclude that in Japan, viral transmission is from parent to child in approximately half of the infections, the other half of the infections usually originating outside the nuclear family. From the failure to detect the JCV subtypes that comprise the majority of infection in Americans in Okinawans born during the occupation of Okinawa by U.S. troops, Kato et al. [1997] conclude that JCV "is rarely transmitted between human populations." (I would modify "rarely" to "not easily," since the conditions on Okinawa were certainly not those characterizing an integrated population.) The symptomatology, if any, that accompanies the acquisition of seropositivity is unknown.

There is evidence that the type of JCV one contracts in youth tends to persist throughout life [Kunitake et al., 1995]. There is also reason to believe that a latent infection may periodically, throughout life, become active, much as is the case for the herpes virus. For instance, we have observed in Japanese subjects rogue cells with equal frequencies in a group of adults and their children [Neel, 1998], and anti-JCV titers were

quite similar in the older and younger subjects studied in Japan [Neel et al., 1996].

TISSUE DISTRIBUTION OF THE VIRUSES

Systematic studies of the tissue distribution of the viruses are only now being undertaken. Up to the present time, the presence of the virus in at least four cell/tissue/organ systems seems to have been established. First, the human cell best known for its sensitivity to JCV infection is the oligodendrocyte, viral activity in which results in the progressive multifocal leukoencephalopathy (PML) encountered in the immunosuppressed, especially those with AIDS. In such patients, viral DNA can also be demonstrated in bone marrow, liver, spleen, and lung [Grinnel et al., 1983]. Recently, Rencic et al. [1996] have described the presence of JCV DNA in an oligoastrocytoma from an immunocompetent person. Second, the presence of viral DNA in the circulating lymphocytes of AIDS patients with PML and in HIV-positive persons without AIDS is well established, and JCV DNA has been demonstrated by the polymerase chain reaction (PCR) techniques in a small fraction of the lymphocytes of apparently normal persons as well as in hematopoietic stem cells [Tornatore et al., 1992]. It is presumably some fraction of this small fraction of the lymphocytes in which activation of the virus results in the rogue cells we have described. Third, Coleman et al. [1980] first detected viral DNA in the urine of some 2% of pregnant English women, this finding presumably reflecting the mild immunosuppression that occurs in pregnancy. However, in Japan, with a similar frequency of JCV seropositives, virus was even more frequently detected in the urine of non-immunosuppressed individuals, the fraction JC-positive increasing from five in 38 (13.2%) in the 0 to 29 age group to 20 in 44 (45.5%) in the age interval 60 to 89 [Kitamura et al., 1990]. JC sequences were subsequently recovered from the normal renal medulla of 40.6% (13/32) of individuals undergoing surgery for renal cancer [Tominaga et al., 1992]. Whether the virus is normally resident in renal cellular tissue, or whether its presence in urine and renal medulla is the result of contamination of the kidney by virus-bearing lymphocytoid cells, is not yet clear. Fourth and finally, Dr. Richard Boland's group has recently reported the presence of JCV DNA in normal and abnormal colon cells [Laghi et al., 1996]. DNA was isolated from 37 resected colon cancers and matched normal tissues and examined for the presence of three JCV T antigen sequences by the PCR technique. Sequences were found in 73% of normal samples and 97% of colon cancers. That this viral presence is not (always) due to transitory cells of the lymphocytoid line is strongly suggested by the fact that viral fragments were detected in five of ten colon cancer xenografts. A systematic study of the presence of JCV DNA in other tissues is clearly in order.

JCV AND ONCOGENESIS

So now in concluding let me try to weave these observations, together with those of several other inves-

tigators, into a consistent hypothesis. What I have to say is highly influenced by the extensive studies on SV40, whose DNA structure is highly homologous to that of JCV and BKV. I suggest that JCV (and perhaps BKV) may be important players in human oncogenesis. This possibility was raised shortly after the two viruses were discovered, largely on the basis of the homology of the virus with SV40, but has not been pursued vigorously in recent years [references in Walker and Frisque, 1986]. These recent data on cytogenetic effects force a reconsideration of that hypothesis. I postulate that in many tissues of the body—the exact tissue distribution yet to be established—the presence of the virus is sporadically associated with the generation of both simple and complex chromosomal damage. So far as is known, this damage is at random. The damage is a low-frequency event, but given episodic activity of the virus in the millions of cells of an infected tissue, there will during an individual's lifetime be thousands and thousands of damaged cells in any tissue. Many of the cells with simple damage, and most rogue cells, will undoubtedly be quickly lost. However, it just requires one translocation of the correct type to play a role in a clonal malignancy. The argument is that JCV (and possibly BKV) are the “machines” that drive some (considerable?) fraction of the chromosomal damage that characterizes so many malignancies. If we may argue by analogy with the clastogenic effects of ionizing radiation, the lag period between the initial chromosomal insult and the diagnosis of a malignancy may require some 20 to 30 years [Schull and Weiss, 1992]. This long lag period, coupled with the probability of periodic viral activity, presents serious problems in establishing epidemiological relationships.

Thus far this presentation has emphasized the clastogenic possibilities of infection with the human polyoma viruses, presumably primarily through helicase activity on the part of the large T antigen, as documented for the large T antigen produced by SV40 [reviewed in Shah, 1996; Walker and Frisque, 1986]. However, the T antigen of SV40 has also been shown to complex with no less than seven proteins in host human cells, including two proteins playing key roles in oncogenesis, namely, p53 and the pRb family of proteins [reviewed in Fanning and Knippers, 1992]. The highly homologous T antigen of BKV has also been shown to react with both p53 and the pRb family of proteins as well as several other cellular proteins [references in Dyson et al., 1990; Harris et al., 1996]. Although studies of JCV T antigen have not been as numerous, recent experiments are demonstrating in the JCV T antigen functional domains similar to those in BKV T antigen (and SV40 T antigen) [references in Major et al., 1992; Swenson et al., 1996]. Because of the key role of p53 and the pRb family of proteins in oncogenesis in general [reviewed in Vogelstein and Kinzler, 1998], it seems to be assumed this complexing might play a role in oncogenesis. However, to be effective in oncogenesis, these complexes would seem to have to tie up those key proteins that, in effect, functional nulls are created, that have to be recreated each cell generation. While this possibility cannot be denied, I put my primary money on the clastogenic potential of this vi-

rus, since the critical translocation, deletion, or duplication need happen only once and then is indefinitely propagated.

In closing, I thank you again for the honor, and the accompanying opportunity to tell this little story about how one unexpected observation can lead to another and still another, a surprising and exhilarating sequence of events, until finally one has a full-blown hypothesis. Let me be very careful—I did not claim to have found *the* cancer virus, but only one possible player in the complex of events that constitute oncogenesis. I also want to recognize how difficult it will be to establish this hypothesis in a way that will satisfy Koch's postulates but suggest that the techniques of molecular genetics have much to offer in the future.

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REFERENCES

- Awa AA, Honda T, Neriishi S, Sofuni T, Shimba H, Ohtaki K, Nakano M, Kodama Y, Itoh M, Hamilton HB. 1987. Cytogenetic studies of the offspring of atomic bomb survivors. In: Obe G, Basler A, editors. Cytogenetics: basic and applied aspects. Berlin: Springer-Verlag. p 166–183.
- Awa AA, Honda T, Sofuni T, Neriishi S, Yoshida M, Matsui T. 1971. Chromosome-aberration frequency in cultured blood-cells in relation to radiation dose of a-bomb survivors. *Lancet* 2:903–905.
- Awa AA, Neel JV. 1986. Cytogenetic “rogue” cells: what is their frequency, origin, and evolutionary significance? *Proc Natl Acad Sci USA* 83:1021–1025.
- Awa AA, Sofuni T, Honda T, Itoh M, Neriishi S, Otake M. 1978. Relationship between the radiation dose and chromosome aberrations in atomic bomb survivors of Hiroshima and Nagasaki. *J Radiat Res (Tokyo)* 19: 126–140.
- Bloom AD, Neel JV, Choi KW, Iida S, Chagnon N. 1970. Chromosome aberrations among the Yanomama Indians. *Proc Natl Acad Sci USA* 66:920–927.
- Bloom AD, Neel JV, Tsuchimoto T, Meilinger K. 1973. Chromosomal breakage in leukocytes of South American Indians. *Cytogenet Cell Genet* 12:175–186.
- Bochkov NP, Katosova LD. 1994. Analysis of multiaberrant cells in lymphocytes of persons living in different ecological regions. *Mutat Res* 323:7–10.
- Brown P, Tsai T, Gajdusek DC. 1975. Seroepidemiology of human papovaviruses. *Am J Epidemiol* 102:331–340.
- Candeias JAN, Baruzzi RG, Pripas S, Iunes M. 1977. Prevalence of antibodies to the BK and JC papovaviruses in isolated populations. *Rev Saude Publica* 11:510–514.
- Coleman DV, Wolfendale MR, Daniel RA, Dhanjal NK, Gardner SD, Gibson PE, Field AM. 1980. A prospective study of human polyoma virus infection in pregnancy. *J Infect Dis* 142:1–8.
- Dyson N, Bernards R, Friend SH, Gooding LR, Hassell JA, Major EO, Pipas JM, Vandyke T, Harlow E. 1990. Large T antigens of many polyomaviruses are able to form complexes with the retinoblastoma protein. *J Virol* 64:1353–1356.
- Fanning E, Knippers R. 1992. Structure and function of simian virus 40 large tumor antigen. *Annu Rev Biochem* 61:55–85.
- Fox DP, Robertson FW, Brown T, Whitehead AR, Douglas JDM. 1984. Chromosome aberrations in divers. *Undersea Biomed Res* 11:193–204.
- Grinnel BW, Padgett BL, Walker DL. 1983. Distributions of nonintegrated DNA from JC papovavirus in organs of patients with progressive multifocal leukoencephalopathy. *J Infect Dis* 147:669–675.

- Harris KF, Christensen JB, Imperiale MJ. 1996. BK virus large T antigen: interactions with the retinoblastoma family of tumor suppressor proteins and effects on cellular growth control. *J Virol* 70:2378–2386.
- Heim S, Mitelman F. 1995. *Cancer cytogenetics*. 2nd edition. New York: John Wiley & Sons. p x, 536.
- Kato A, Kitamura T, Sugimoto C, Ogawa Y, Nakazato K, Nagashima K, Hall WW, Kawabe K, Yogo Y. 1997. Lack of evidence for the transmission of JC polyomavirus between human populations. *Arch Virol* 142: 875–882.
- Kitamura T, Aso Y, Kuniyoshi N, Hara K, Yogo Y. 1990. High incidence of urinary JC virus excretion in nonimmunosuppressed older patients. *J Infect Dis* 161:1128–1133.
- Kitamura T, Kunitake T, Guo J, Tominaga T, Kawabe K, Yogo Y. 1994. Transmission of the human polyomavirus JC virus occurs both within the family and outside the family. *J Clin Microbiol* 32:2359–2363.
- Kunitake T, Kitamura T, Guo J, Taguchi F, Kawabe K, Yogo Y. 1995. Parent-to-child transmission is relatively common in the spread of the human polyomavirus JC virus. *J Clin Microbiol* 33:1448–1451.
- Laghi L, Chauhan DP, Marra G, Major EO, Neel JV, Boland CR. 1996. Amplification of JC virus (JCV) sequences from human colorectal cancers. *Gastroenterology* 110:A548.
- Lazutka JR. 1996. Chromosome aberrations and rogue cells in lymphocytes of Chernobyl clean-up workers. *Mutat Res* 350:315–329.
- Major EO, Amemiya K, Tornatore C, Houff SA, Berger JR. 1992. Pathogenesis and molecular biology of progressive multifocal leukoencephalopathy, the JC virus-induced demyelinating disease of the human brain. *Clin Microbiol Rev* 5:49–73.
- Neel JV. 1998. An association, in adult Japanese, between the occurrence of rogue cells among cultured lymphocytes (JC virus activity) and the frequency of “simple” chromosomal damage among the lymphocytes of persons exhibiting these rogue cells. *Am J Hum Genet* 63:489–497.
- Neel JV, Awa AA, Kodama Y, Nakano M, Mabuchi K. 1992. “Rogue” lymphocytes among Ukrainians not exposed to radioactive fall-out from the Chernobyl accident: the possible role of this phenomenon in oncogenesis, teratogenesis, and mutagenesis. *Proc Natl Acad Sci USA* 89:6973–6977.
- Neel JV, Major EO, Awa AA, Glover T, Burgess A, Traub R, Curfman B, Satoh C. 1996. Hypothesis: “rogue cell”-type chromosomal damage in lymphocytes is associated with infection with the JC human polyoma virus and has implications for oncogenesis. *Proc Natl Acad Sci USA* 93:2690–2695.
- Nichols WW, Girardi AJ, Bradt CI, Hill R, Cody C. 1985. Cytogenetic changes induced in human diploid fibroblasts by tsA58 SV40 at permissive and restrictive temperatures. *Mutat Res* 150:327–332.
- Ray FA, Kraemer PM. 1993. Iterative chromosome mutation and selection as a mechanism of complete transformation of human diploid fibroblasts by SV40 T antigen. *Carcinogenesis* 14:1511–1516.
- Ray FA, Meyne J, Kraemer PM. 1992. SV40 T antigen induced chromosomal changes reflect a process that is both clastogenic and aneuploidogenic and is ongoing throughout neoplastic progression of human fibroblasts. *Mutat Res* 284:265–273.
- Ray FA, Peabody DS, Cooper JL, Cram LS, Kraemer PM. 1990. SV40 T antigen alone drives karyotype instability that precedes neoplastic transformation of human diploid fibroblasts. *J Cell Biochem* 42:13–31.
- Rencic A, Gordon J, Otte J, Curtis M, Kovatich A, Zoltick P, Khalili K, Andrews D. 1996. Detection of JC virus DNA sequence and expression of the viral oncoprotein, tumor antigen, in brain of immunocompetent patient with oligoastrocytoma. *Proc Natl Acad Sci USA* 93:7352–7357.
- Rowley JD. 1996. Leukemias, lymphomas, and other related disorders. In: Rimoin DL, Connor JM, Pyeritz R, editors. *Principles and practice of medical genetics*. London: Churchill Livingstone. p 1687–1701.
- Salomaa S, Sevan'kaev AV, Zhloba AA, Kumpusalo E, Mäkinen S, Lindholm C, Kumpusalo L, Kolmakow S, Nissinen A. 1997. Unstable and stable chromosome aberrations in lymphocytes of people exposed to Chernobyl fallout in Bryansk, Russia. *Int J Radiat Biol* 71:51–59.
- Sandberg AA. 1990. *The chromosomes in human cancer and leukemia*. New York: Elsevier Science Publishing Co. p 1315.
- Scheid W, Weber J, Petrenko S, Traut H. 1993. Chromosome aberrations in human lymphocytes apparently induced by Chernobyl fallout. *Health Phys* 64:531–534.
- Schull WJ, Weiss KM. 1992. Radiation carcinogenesis in humans. *Adv Radiat Biol* 16:215–258.
- Sevan'kaev AV, Tsyb AF, Lloyd DC, Zhloba AA, Moiseenko VV, Skrbabin AM, Klimov VM. 1993. “Rogue” cells observed in children exposed to radiation from the Chernobyl accident. *Int J Radiat Biol* 63:361–367.
- Shah KV. 1996. Polyomaviruses. In: Fields BN, Knipe DM, Howley PM, Chanock RM, Melnick JL, Monath TP, Roizman B, Straus SE, editors. *Field's virology*. Philadelphia: Lippincott-Raven. p 2027–2043.
- Stewart N, Bacchetti S. 1991. Expression of SV40 large T antigen, but not small T antigen, is required for the induction of chromosomal aberrations in transformed human cells. *Virology* 180:49–57.
- Sugimoto C, Kitamura T, Guo J, Al-Ahdal MN, Shchelkunov SN, Otova B, Ondrejka P, Chollet J-Y, El-Safi S, Ettayebi M, Grésenguet G, Kocagöz T, Chaiyarasamee S, Thant KZ, Thein S, Moe K, Kobayashi N, Taguchi F, Yogo Y. 1997. Typing of urinary JC virus DNA offers a novel means of tracing human migrations. *Proc Natl Acad Sci USA* 94:9191–9196.
- Swenson JJ, Trowbridge PW, Frisque RJ. 1996. Replication activity of JC virus large T antigen phosphorylation and zinc finger domain mutants. *J Neurovirol* 2:78–86.
- Tawn EJ. 1987. The frequency of chromosome aberrations in a control population. *Mutat Res* 182:303–308.
- Tawn EJ, Binks K. 1989. A cytogenetic study of radiation workers: the influence of dose accumulation patterns and smoking. *Radiat Protect Dosis* 28:173–180.
- Tawn EJ, Cartmel CL, Pyta EMT. 1985. Cells with multiple chromosome aberrations in control individuals. *Mutat Res* 14:247–250.
- Tominaga T, Yogo Y, Kitamura T, Aso Y. 1992. Persistence of archetypal JC virus DNA in normal renal tissue derived from tumor-bearing patients. *Virology* 186:736–741.
- Tornatore C, Berger JR, Houff SA, Curfman B, Meyers K, Winfield D, Major EO. 1992. Detection of JC virus DNA in peripheral lymphocytes from patients with and without progressive multifocal leukoencephalopathy. *Ann Neurol* 31:454–462.
- Verschaeve L, Domracheva EV, Kuznetsov SA, Nechai VV. 1993. Chromosome aberrations in inhabitants of Byelorussia: consequences of the Chernobyl accident. *Mutat Res* 287:253–259.
- Vogelstein B, Kinzler KW. 1998. *The genetics of cancer*. New York: McGraw-Hill. p xix, 731.
- Walker DL, Frisque RJ. 1986. The biology and molecular biology of JC virus. In: Salzman NP, editor. *The papovaviridae*. Vol. 1: the polyomaviruses. New York: Plenum Press. p 327–377.