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LECTURE

EVOLUTION AND MODES OF TRANSMISSION OF RNA TUMOR VIRUSES

Evolution and Modes of Transmission of RNA Tumor Viruses

Parke-Davis Award Lecture

George J. Todaro, MD

Most vertebrates contain sets of gene sequences (virogenes) which are an integral part of the chromosomal DNA and which can code, in some instances, for the production of Type C RNA tumor viruses. These genes are transmitted from parent to progeny along with other cellular genes, and their activation from a normally repressed state may be part of the mechanism by which RNA tumor viruses produce cancer. Isolates of endogenous genetically transmitted baboon Type C viruses are morphologically and biochemically related to other mammalian Type C viruses but can clearly be distinguished from the other groups (mouse, rat, cat, etc.) by immunologic and nucleic acid hybridization criteria. Within the primates, Type C viral gene sequences have evolved as the species have evolved, with virogenes from the most closely related genera and families showing the most sequence homology; all higher primates, including man, however, do have detectable virogene sequences in their normal tissues. Type C viruses have also been transferred under natural conditions between species only remotely related phylogenetically. The results show three clear examples where viral genes from one group of animals have become incorporated into the germ line of genetically distant groups of animals (inheritance of acquired genes). Infectious Type C viruses of primates, distinct from the endogenous primate virus group, have also been isolated (woolly monkey and gibbon isolates) and can be shown to produce tumors in other primates. Related viral information (nucleic acid sequences, enzymes, and antigens) have been reported in human tumors. The significance of infectious and/or genetically transmitted viruses in naturally occurring cancer is a major focus of current research. The presence of genetically transmitted viral genes in so many vertebrate species and the evidence that they have been conserved in several distinct vertebrate lineages suggests that they may provide some normal function(s) advantageous to the species carrying them and that their potential to cause cancers is a pathologic manifestation of normal, as yet undefined, physiologic processes. (Am J Pathol 81:590–606, 1975)

Transmission of Virogenes

In the course of studies on the development of continuous lines of mouse embryo cells in culture,[1,2] it was noted that certain of the spontaneously transformed cell lines began to release Type C RNA tumor viruses.[3] This finding, along with several observations from Dr. Robert Huebner's laboratory on the presence of virus-specific antigens in the embryos of several mouse strains and the knowledge that this group of viruses was capable of producing tumors in several species, led to the hypothesis

From the Viral Leukemia and Lymphoma Branch, National Cancer Institute, Building 37, Room 1B-22, National Institutes of Health, Bethesda, Maryland, 20014.

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(*virogene–oncogene hypothesis*) [4,5] that the information for the formation of such viruses was transmitted genetically from parent to progeny along with other cellular genes. Activation of this normally repressed, genetically transmitted, Type C endogenous virogene information, rather than infection from outside the animal, was proposed as the most common mechanism by which Type C RNA tumor viruses produce naturally occurring cancers.

Genetic transmission from parent to offspring rather than infection from animal to animal was postulated to be the primary means by which the genes have been maintained in animal populations. Much subsequent experimental work supports this, the most important being that "virus-free" cell cultures (Table 1) derived from chicken, mouse, hamster, rat, pig, cat, and baboon tissues (see Lieber and Todaro [6] for review) can begin to secrete, either spontaneously or after treatment with chemical inducing agents, typical complete Type C viruses.[7,8] Cocultivation of the virus-producing cell cultures with appropriate permissive cell lines from heterologous species has been needed to detect and amplify virus production in several of these systems. In general, avian and mammalian cells in culture have been resistant to superinfection by their own endogenous Type C viruses. The properties characterizing such endogenous mammalian Type C viruses which are products of the genetically transmitted virogenes are summarized in Table 2.

The endogenous Type C virogenes are those sets of sequences that are an integral part of the host species' chromosomal DNA and code for the production of Type C viruses. These gene sequences in normal cellular DNA related to Type C viruses (virogenes) should be distinguished from Type C viral DNA sequences which can be added to the animal's genome from the outside by "exogenous" viral infection and subsequent integration (*provirus formation*).[9] Endogenous Type C virogenes should also be distinguished (Table 3) from those gene sequences not originally present in the genome that are postulated to arise by gene duplication

Table 1—Species Where a Complete Virogene Is Known to be Present in Normal Cells

Chicken
Chinese hamster
Syrian hamster
Mouse (*Mus musculus*)
Mouse (*Mus caroli*)
Rat
Cat
Pig
Baboon

Table 2—Properties of Endogenous Type C Virogenes

1. DNA of all somatic and germ cells of all the animals in a species contain viral gene sequences.
2. Multiple related but not identical copies present in the cellular DNA, more than DNA from a heterologous cell that is actively producing virus.
3. Virus expression (RNA, gs antigen, polymerase, complete particles) under cellular control; expressed in certain tissues at certain times during development.
4. Clonal lines either spontaneously or after induction are capable of releasing complete virion.
5. Cells generally resistant to exogenous infection by the homologous endogenous virus.

and/or recombination during the lifetime of the animal mediated by the reverse transcriptase mechanism [10,11] (*protovirus formation* [12]).

The sets of virogenes that a particular species possesses are normally repressed, but they can be activated by a variety of intrinsic (genetic, hormonal) as well as extrinsic (radiation, chemical carcinogens, other infecting viruses) factors (Table 4). As cellular genes, Type C virogenes are subject to the pressures of mutation and selection; as such, closely related animal species would be expected to have closely related endogenous Type C virogenes. What is unique about Type C virogenes—as distinguished from all other known cellular genes—is that, at least in some species, they can give rise to the production of infectious Type C virus particles. Since endogenous Type C virogenes code for the production of particles secreted by the cell and contain specific viral proteins, a reverse transcriptase, and a high molecular weight RNA, they provide a very attractive system for the study of the genetic control of cellular genes.

While viruses have been known in several animal species for a long time, it has only been within the last year or two that endogenous Type C viruses have been successfully propagated from the primates, man's closest relatives. More than a dozen independent isolates of infectious baboon Type C virus have been obtained in this laboratory by cocultivation of normal kidney, spleen, lung, testes, and placenta from several different

Table 3—Major Differences Between Virogene and Protovirus Models

Virogene	Protovirus
1. Viral copies present in germ cells and somatic cells.	1. Germ cells lack virus information. Generated in rare somatic cells by chance.
2. Genes maintained in population by normal cellular replication. Reverse transcriptase *not required.*	2. Reverse transcriptase plays essential role in generating new viruses.
3. Transformation results from activation of normally latent cellular genes associated with and/or part of the viral gene sequences.	3. Transformation results from the generation of new gene sequences that do not preexist in normal cellular DNA.

Table 4—Implications of the Virogene-Oncogene Hypothesis

	Virogenes
1.	All somatic cells of a species have DNA homologous to Type C virus RNA of that species (virogenes).
2.	Type C viruses derived from closely related species should have closely related specific antigens, e.g., gs antigens, polymerase, and their nucleic acid sequences should be more related to one another than are those viruses released by distantly related species (virogene evolution).
	Oncogenes
3.	The transformation specific sequences of RNA tumor viruses should be present in normal cellular DNA (oncogenes).
4.	Spontaneous, chemically induced and virus-induced transformed cells and tumor cells should have RNA as well as DNA sequences homologous to the transforming specific sequences found in tumor viruses (oncogene expression).

species of baboon (*Papio hamadryas*, *P. papio*, *P. anubis*, and *P. cynocephalus*) with suitable permissive host cell lines. These isolates are all morphologically and biochemically typical of mammalian Type C viruses but are distinctly different, as characterized by immunologic and nucleic acid hybridization techniques, from all other previously studied Type C viruses.[13,14] Further, the isolates are closely related to each other by host range, viral neutralization and interference, and by immunologic and nucleic acid hybridization criteria. ^{3}H-DNA transcripts prepared from three of the baboon Type C virus isolates hybridize completely to DNA extracted from various tissues of several different normal baboons.[14] These data suggested that these Type C virus isolates were, indeed, endogenous viruses of baboons. The finding of DNA sequences in normal tissues is one of the strongest pieces of evidence that the viral information is maintained in the population as cellular genes.

If the baboon Type C viruses were truly endogenous primate viruses as we proposed [13] and had evolved as the species evolved, then it appeared reasonable to suspect that other old-world monkeys that are close relatives to the baboon would have related virogene sequences in their DNA. Those primate species less closely related to baboons taxonomically would be expected to have much more extensive mismatching of their virogene DNA sequences. This would be reflected in a decreased extent of hybridization and a lower thermal stability of the products formed when heterologous cellular DNA was hybridized to the baboon Type C viral complementary DNA probes.

The study of the evolutionary relationships of Type C viral gene sequences is especially favorable in primates since much is known about the evolutionary relationships between primates: the fossil record has been intensively analyzed, as *Homo sapiens* has been particularly interested in its own origins. The old-world monkeys (which include the baboon spe-

cies) have been separated from the great apes and man for 30 to 40 million years. The new-world monkey branch diverged from the common stem leading to both the apes and the old-world monkeys approximately 50 million years ago, while the prosimians evolved from primitive mammalian stock roughly 60 to 80 million years ago.

Hybridization studies employing a DNA copy of the baboon virus RNA were used to detect Type C viral nucleic acid sequences in primate cellular DNA. Sequences related to those of the baboon Type C virus are found in all other old-world monkey species and higher apes and are also found in man. The results establish that, within the primates, Type C viral genes have evolved as the species have evolved, with virogenes from more closely related genera and families showing more sequence homology than those from distantly related taxons. The ubiquitous presence of endogenous Type C virogenes among anthropoid primates and their evolutionary preservation for at least 30 to 40 million years clearly shows they have evolved in primates for at least this length of time as stable cellular elements.[15] The results suggest, but do not in themselves prove, that the transmitted viral genes may provide a selective advantage to the species possessing them.

Transmission of Type C Genes Between Distantly Related Species

Type C viruses have also, under natural conditions, been transferred between species that are only remotely related phylogenetically. One example involves the transfer of an endogenous primate Type C virus into the germ line of the ancestor of the domestic cat.

RD-114 virus in domestic cats was characterized following studies involving the heterotransplantation of human RD tumor cells into fetal cats, some of which developed disseminated rhabdomyosarcomas with the human karyotype of the original RD cell line. Although the original RD cell line did not have detectable Type C viruses, electron microscopic studies of two of the cat tumors and a cell line established from one of the tumors revealed typical Type C virions.[16] Also, further investigations have shown that clones of cat cells from the CCC line of fetal kidney fibroblasts spontaneously released viruses found to be closely related to RD-114. Subsequent studies establish that RD-114/CCC viruses are not human in origin but belong to a distinct group of endogenous feline Type C virus.[17,18]

In comparing the endogenous primate viruses to the RD-114/CCC group of viruses, we found that they are related to each other but can be distinguished by biologic and immunologic criteria and by partial nucleic acid sequence homology. Virogene sequences in the DNA of old-world

monkeys and domestic cats show a degree of relatedness not shared by the DNA of these species. Genes related to the nucleic acid of an endogenous domestic cat Type C virus (RD-114) are, nevertheless, found in the cellular DNA of anthropoid primates, while at the same time many members of the cat family Felidae lack these sequences (Table 5). Endogenous viruses from one group of mammals (primates) are thus concluded to have infected and become a part of the germ line of an evolutionary distant group of animals, the ancestors of the domestic cat.[19,20] From the relatives of the domestic cat that have RD-114 viral genes and from those that did not acquire them, we have concluded that the infection occurred 3 to 10 million years ago, somewhere in Africa or in the Mediterranean Basin region. Because of the stability of the viral gene sequences when they are incorporated into cellular DNA, events that have occurred millions of years ago still can be recognized by examining the genetic information of the virus and that of the host cell.

More recently, our laboratory has found two additional examples of gene transmission between species, in one case from a rat ancestor to the domestic cat ancestors and their close relatives, and in the other, from an ancestor of the mouse to an ancestor of the domestic pig.

Experiments have shown that, besides the RD-114/CCC cat viruses which were transmitted from primates to cats (as described above), another distinct class of Type C RNA virus was acquired by cats and is now present in their germ line. These feline leukemia viruses (FeLV) were transmitted from an ancestor of the rat to ancestors of the domestic cat and their close relatives.[21] The relationships observed between FeLV and the endogenous viruses of rodents are quite similar to those with endogenous feline viruses of the RD-114/CCC group and endogenous primate Type C viruses. FeLV-related gene sequences are found not only in the

Table 5—Relationship Between Cat and Baboon Endogenous Type C Virus

1. The cat (RD-114/CCC) and baboon virus groups are *related,* but are distinct from one another by:
   a. Viral DNA-RNA hybridization,
   b. Inhibition of polymerase activity by antibody,
   c. Antigenicity of the p30 protein,
   d. Viral interference,
   e. Viral neutralization.
2. Cat and baboon unique sequence DNA markedly different, species diverged from one another over 80 million years ago.
3. Cat (RD-114/CCC) virus DNA transcripts hybridize to the DNAs of *all* old-world monkeys and apes, and to the DNAs of domestic cats and certain other *Felis* species.
4. Baboon (M7/M28) virus DNA transcripts hybridize to the DNAs of *all* old-world monkeys, higher apes, and man, and to DNAs of those *Felis* species which contain RD-114 related sequences.

cellular DNA of domestic cats but also in the DNA of three other closely related Felidae (*Felis sylvestris, F. margarita, F. chaus*). More distantly related *Felis* species lack FeLV-related virogenes, while the cellular DNA of rodents, in particular rats, contains related virogene sequences. This suggests that FeLV-related genes were introduced into the *Felis* lineage following trans-species infection(s) by Type C viruses of rodent origin. It is interesting that cats which contain sequences related to RD-114/CCC genes also contain FeLV-related genes, while other *Felis* species lack both sets of sequences. Both groups of viral genes appear to have been introduced to the cat germ cells from distinctly different groups of animals (rodents and primates).[21]

The third example of gene transmission between species is that from an ancestor of the mouse to an ancestor of the domestic pig. Pig cell cultures produce Type C viruses [22-25] that can be shown to be genetically transmitted and present in all pig tissues in multiple copies in the cellular DNA.[25,26] Close relatives, such as the European wild boar and the African bush pig, have closely related viral genes in their DNA. It can be shown that this virus was acquired by an ancestor of the pig from a small rodent related to the mouse.[27] The nucleic acid homology between the endogenous pig Type C viral RNA and murine cellular DNA suggests that the endogenous virus had a common origin. From the extent of hybridization of the pig Type C viral DNA probes to rodent cellular DNA, it can be seen that the Type C virogenes in the pig were transmitted from members of the family Muridae after the mouse had separated from the rat but before the different species of mice had diverged from each other. Rodent viral genes thus gave rise to infectious particles that became incorporated into the porcine germ line.

The data, as summarized in Table 6, demonstrate that viral genes from one group of animals can give rise to infectious particles that not only can integrate into the DNA of animals of another species, but can also be incorporated into the germ line (germ line inheritance of acquired virus

Table 6—Examples of Transmission of Type C Virus Genes Between Species

Donor	Recipient	Genetically transmitted in recipient
Primate (old-world monkey)	*Felis* (ancestor of the domestic cat)	Yes
Rodent (mouse ancestor)	Pig ancestor	Yes
Rodent (rat ancestor)	*Felis* (ancestor of domestic cat)	Yes (but also horizontally transmitted in *Felis catus*)

genes). Clearly, if viral gene sequences can be acquired in this way, it is possible that Type C viruses have served to introduce other genes from one species to another, and they may provide an important mechanism by which species stably acquire new genetic information.

Origin of the Infectious Primate Type C Virus Group

So far we have focused on trans-species infections where viral information genetically transmitted in one group of animals becomes successfully incorporated into the germ line of another species. The endogenous viruses of one species clearly have the capacity to infect cells of distantly related species. Infectious primate Type C viruses have been recovered from several colonies of gibbon apes [28] and from one woolly monkey.[29,30] These viruses, GALV (gibbon ape leukemia virus) and SSV-SSAV (simian sarcoma virus–simian sarcoma-associated virus) have been isolated from primates; they spread from animal to animal under natural conditions and they are able to induce tumors when inoculated into other primates.[30-32] These viruses are related to one another by several immunologic criteria and by viral interference, and they are known to contain related RNA genomes.[33] Gene sequences homologous to those of the RNAs of GALV and SSAV have not been detected in the cellular DNA of normal primates studied thus far.[34,35] These woolly monkey and gibbon viruses appear to be infectious—horizontally transmitted from animal to animal—and *not* endogenous primate viruses like the baboon virus group.

In studying the relationships between the various mammalian Type C viruses using nucleic acid hybridization it was noted that the infectious primate viruses, GALV and SSAV, share partial sequence homology with endogenous Type C viruses from the laboratory mouse, *Mus musculus*.[36] These unexpected findings suggested the possibility that the infectious viruses of the GALV-SSAV group may have originated by trans-species infection of certain primates by an endogenous rodent Type C virus.

As noted above, most of the isolates of infectious primate Type C viruses have been obtained from gibbons in colonies both in the United States and in Southeast Asia. Naturally infected, as well as experimentally infected, animals in these colonies have been found that have antibodies directed against the infectious primate Type C viruses.[32,37] For these reasons we chose to study Type C viruses from several feral Asian subspecies of *Mus musculus*. *M. m. molossinus* from Japan and *M. m. castaneus* from Thailand were both found to have endogenous viruses closely related to laboratory strains of *Mus musculus*.[38] However, a Type C virus was also induced from a cell culture of the distantly related Thai mouse

species *Mus caroli*. The latter virus, unlike the isolates from the *Mus musculus* subspecies, was found to be closely related antigenically to the gibbon and woolly monkey Type C viruses and only weakly related to any of the previously described "mouse" Type C viruses. It shares cross-reactive reverse transcriptase and p30 antigens and cross-interferes with the infectious primate Type C viruses.[39] These results lead to the conclusion that the infectious, horizontally transmitted primate Type C viruses have originated by trans-species infections of primates with an endogenous Type C virus from *Mus caroli* or another closely related species. This trans-species infection appears to be a relatively recent event, with the viruses not yet being incorporated into the genomes of the recipient primate species.

The Type C viruses of the GALV-SSAV group are poorly controlled by the primate host and appear readily capable of producing neoplastic disease. Infection by such viruses can cause local epidemics of lymphoproliferative tumors in infected gibbon colonies.[37] The ability of investigators to isolate viruses from gibbons, however, is not restricted to animals with tumors. Recently, three isolates have been obtained from animals without tumors—the brains of normal gibbons which came from a single colony in the United States.[33] Based on immunologic assays and interference tests, the group of infectious Type C viruses of primates contains many members, all partially related to one another. Based on DNA-RNA hybridization which shows extensive mismatching of the gene sequences when the different gibbon isolates were compared to one another and to SSAV,[33] the infectious primate Type C viruses at present can be classified into four distinct subgroups (Table 7). It is probable that additional subgroups will be defined as new isolates are obtained.

Type C Viruses in Primate Populations

Since the horizontally transmitted primate viruses described above are infectious for and can cause tumors in primates, the possibility of the involvement of this group of viruses in human tumors is a subject of considerable interest. This possibility is supported by data obtained by different experimental procedures in a number of laboratories. An enzyme

Table 7—Infectious Primate Type C Viruses: Isolation and Partial Characterization

	Proposed subgroup	Isolates	Reference
A	Woolly monkey	SSV/SSAV	Wolfe *et al.*[30]
B	Gibbon Type 1	GALV-1	Kawakami *et al.*[25]
C	Gibbon Type 2	GALV-SEATO	Kawakami and Buckley[37]
D	Gibbon Type 3	GBr-1, GBr-2, GBr-3	Todaro *et al.*[33]

with biochemical properties related to those of Type C viruses and with antigenic properties similar to polymerases of the woolly monkey Type C virus (SSAV) and the gibbon ape leukemia virus (GALV) has been detected in human acute leukemia cells.[40,41] The DNA products of endogenous reactions from the "virus-like" partiulate fraction of acute leukemia cells hybridize preferentially to viral RNA from SSAV and GALV.[42,43] Using radioimmunoassays, antigens related to the major structural proteins (p30) of Type C viruses have been detected in peripheral white blood cells from 5 patients with acute leukemia.[44] These results suggest that viruses of this group, known to be infectious and tumorigenic in other primates, may also be associated with acute leukemia in man.

Quite recently, several laboratories have reported the isolation of complete infectious Type C viruses from human materials.[45-48] The significance of these reported isolations, however, requires further evaluation. The isolate designated HL-23 (from a cell culture derived from a woman with acute myelogenous leukemia) has been shown to be closely related, if not identical to, the woolly monkey virus, SSAV[49] and thus belongs to one of the four previously described subgroups of infectious primate viruses. The extremely close relationship between HL-23 and SSAV raises the possibility that the virus may have been inadvertently introduced into this leukemic cell culture at some time during its cultivation *in vitro*. The careful characterization of additional isolates made by other laboratories from human tissues and cell cultures is awaited with keen interest.

Primates, including man, are known to contain endogenous Type C viral sequences in their genome which are related to those found in endogenous baboon viruses.[15] Endogenous virogenes may be partially expressed in humans and other primates as evidenced by the detection of RNA sequences [50] and antigens related to the p30 proteins [50,51] of endogenous baboon viruses. The expression of endogenous viral related antigens is found in carcinomas and lymphomas [51] as well as in leukemias; [44] viral p30 antigen expression has also been reported in certain normal human tissues.[52]

Whether infectious viruses or genetically transmitted viruses are more likely to be involved in the generality of cancer is a major focus of investigative research and is, as yet, unresolved. The infectious, horizontally transmitted primate viruses spread from animal to animal and are completely unrelated by molecular and antigenic criteria to endogenous, genetically transmitted primate viruses. The properties of infectious viruses traveling from animal to animal can become rapidly altered, thereby obscuring their origins. For example, the ability of viruses to continually change is a well-established property of the influenza virus group. In

contrast, with genetically transmitted viruses, the species from which they originated can be precisely determined. In this case, genetically transmitted viruses have remained stable enough to make it possible to detect events which occurred millions of years ago.

If infectious Type C viruses are an important cause of human cancer, it is critical to know how they are transmitted and how they are maintained in the population. Is there an animal reservoir which harbors them, or is their spread solely from primate to primate? Finding this reservoir(s), if it exists, provides a chance of disrupting the process. If human leukemia involves the spread of an infectious agent from individual to individual, as is clearly shown to be the case for cat leukemia [53] and bovine leukemia,[54] then identification of the agent and its mode of spread would provide one set of approaches to prevention of the disease. If, on the other hand, activation of genetically transmitted virus by extrinsic (chemical and physical agents) as well as by various intrinsic factors leads to tumor development and there is no contagious virus involved, the approaches to the prevention of disease would be quite different.

Possible Normal Functions of Type C Viruses

The presence of genetically transmitted viral genes in so many vertebrate species and the evidence that they have been conserved through evolution in several distinct vertebrate lineages suggest that they may provide some normal function(s) advantageous to the species carrying them. One may speculate on the possible normal functions they might provide (Table 8). The first suggested role, derived from studies on the expression of viral antigens during the course of development, was that activation of certain virus-specific information during the early stages of differentiation was a normal part of the developmental process.[4] If this were the case, the expression of cancer genes later in life would be an inappropriate manifestation of a normal developmental function. Should the viral genes provide a function critical for normal development they clearly would be conserved during evolution.

Table 8—Possible Functions of Genetically Transmitted Virogenes in Normal Cells

1. Activation of oncogenic information, while inappropriate in adult tissue, plays a normal role during differentiation and development.
2. The integrated virus serves to protect the species against related, more virulent infectious Type C viruses.
3. Virus activation, being linked to transformation, protects the animal by altering the cell membrane. The released virus could alert the immune system making the transformed cells more susceptible to immunologic control.
4. They may have had an evolutionary role as conveyors of genetic information not only within a species but also between species. Only this group of viruses has been shown to transmit genes between germ cells of different species under natural conditions.

The acquisition of viral genes by cats from both primates and rodents, and by pigs from rodents, along with the fact that they have been maintained for millions of years suggests the possibility that the newly acquired viral genes, once integrated, might have been beneficial to the recipient species if they were able to prevent infection by more virulent related viruses. The animals that successfully integrated the genomes would have been at a selective advantage relative to those that did not, if the integrated genome protected against infection, and if infection led to cancer or other type C virus-mediated diseases. Genes that provide protection against disease, and especially against epidemic diseases, would be at a strong selective advantage in natural populations, and this may well explain the success of the transmissions between species described above. For example, in our laboratory we have shown that those species of the genus *Felis*, including the domestic cat, that have acquired primate Type C viral genes are resistant to infection by the endogenous baboon viruses, while those *Felis* species that have not acquired the viral information are still susceptible to baboon viral infection.

A third possible role for endogenous viruses would result if viral activation were closely linked to the expression of the transformed state in the cell. Virus expression in normal situations might, then, be a result of transformation rather than its cause. The activated virus could function to alter the cell membrane and by so doing alert the host immune system. Viewed in this light, a normal function of the virogene system and its activation might be to convey information to the immune system on the number and location of transformed cells in a region of the body. In support of this possibility is the observation that transformed cells in culture—whether transformed spontaneously, by chemical carcinogens, or by other viruses—release their endogenous Type C viruses more readily than do their normal, untransformed counterparts.[55-57] Transformed cells that are releasing high titers of Type C virus have been reported to be much less able to produce tumors when inoculated into immunocompetent animals of the same species.[58] Partial viral expression where viral antigens are introduced into the cell surface may well be sufficient to alter the antigenicity and decrease the possibility that the transformed cell would escape immune control.

One final possibility that should be considered is that Type C viruses have played an important evolutionary role as transmitters of genetic information, not only between cells of an animal and animals of a species, but also between species. That viruses can transmit themselves between the germ cell DNAs of very different species has been established as a result of experiments in the past year. That they can recombine with

cellular gene sequences and transmit these genes to new cells of a different species also has been clearly demonstrated.[59,60] That this transmission of cellular gene information between species has been a major force in evolution, however, remains a speculation.

This suggestion that viruses may have had a major role in evolution is not a new one.[61] Viruses are unique in that they can carry information between genetically isolated species. Classic Darwinian evolution deals with changes which occur by mutation and selection, duplication and rearrangements; the genetic information of a species can be changed and rearranged but not added to from the outside. Viruses, however, offer the possibility of additions of new gene sequences to a species. The Type C viruses as a group are uniquely suited for this role since they must incorporate into the cellular DNA in order to replicate,[12] and they also do not kill the cells that they infect. Each time they move from cell to cell, they have the possibility of carrying with them host cell genes. They thus provide a means of communication between cells of different species and different phyla; they serve to keep a species in contact or in communication with its neighbors—ecologic neighbors as well as genetic neighbors.

Of course, they can transmit information that may disrupt normal cellular control, and by so doing, lead to the development of cancer in the individual. Instances of genetic significance, however, occur when the new genes are incorporated into the germ line. From this perspective, the fact that these viruses cause cancer would then be viewed as a pathologic manifestation of normal processes. While the viral genes may well be etiologic agents in cancer causation, either as exogenous or endogenous viruses, and this may be of profound significance to the affected individuals, these relatively rare and sporadic cases may not be of great evolutionary significance.

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