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The Origin of HIV-1, The AIDS Virus

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Abstract—This article proposes a series of experiments to determine if cows and sheep could be used as animal models for HIV-1, the AIDS virus. To justify this effort, a substantial case is presented that HIV-1 is a natural recombinant of Bovine Leukemia Virus (BLV) and Visna Virus. This natural recombinant may have been inadvertently transferred to humans through the Intensified Smallpox Eradication Program conducted in sub-Saharan Africa in the late 1960s and most of the 1970s.

Introduction

The purpose of this article is to propose a series of experiments to determine if cows and sheep could be used as animal models for Acquired Immune Deficiency Syndrome (AIDS). This would be of assistance in searching for a cure or a vaccine. To justify these experiments, a chain of reasoning is presented that the Human Immunodeficiency Virus Type 1 (HIV-1) is a natural recombinant of Bovine Leukemia Virus (BLV) and Visna Virus, also known as Bovine Immunodeficiency-like Virus (BIV) (1). This natural recombinant was inadvertently transferred to humans through contaminated vaccine used to inoculate millions of Africans against smallpox during the smallpox eradication program conducted in sub-Saharan Africa in the late 1960s and most of the 1970s. These facts have serious implications not only for AIDS research but also for the new technology of 'pharming' (2, 3), whereby genetically altered cows, sheep, goats and pigs are used to obtain human hormones, proteins, and other drugs.

First, the remarkable correlation between the distribution of AIDS cases in the world and the vaccination program that eradicated smallpox will be discussed. Then facts will be presented regarding BLV, Visna, HIV-1, and the recombination of retrovirus genomes which suggest that the smallpox vaccine manufacturing procedure could have created HIV-1 and transmitted it to humans. Finally, the conclusions describe the experiments to establish whether or not cows and sheep could be used as effective animal models for AIDS research.

The Intensified Smallpox Eradication Program and distribution of AIDS cases

From 1967 to 1979 the World Health Organization conducted the Intensified Smallpox Eradication Program (ISEP) (4) resulting in the formal declaration of worldwide smallpox eradication on May 8 1980. During the ISEP, 370 506 000 doses of smallpox vaccine were distributed in 69 countries (5).

About one-quarter of these doses, 97 290 000, were distributed in 7 sub-Saharan African countries—Tanzania, Rwanda, Burundi, Uganda, Zaire, Malawi and Zambia (5). The incidence of HIV-1 infections in these countries today is the highest in the world (6).

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The *New York Times* of September 16 1990 reported that in urban areas of Malawi, Rwanda, Uganda and Zambia, more than 20% of the sexually active adult population was infected with the AIDS virus. This same article noted that in the adult population of Tanzania, Burundi and Zaire, the rate of AIDS infection ranges from 5–20% and is growing. The *Pretoria News* (8 December 1988) reported that one out of ten of a population of 5 million people are infected in Burundi, 50% of the population in Uganda is expected to be HIV positive by the turn of the century, more than 23% of the population in Zambia is estimated to be HIV positive and 8% of the blood donors in Kinshasa, Zaire, are HIV positive. The *Detroit Free Press* revealed on November 26 1991 that so many people are dying in Malawi that group funerals are now necessary.

As of May 1991 the World Health Organization estimated (7) that in sub-Saharan Africa there are as many as 6 million HIV-1 infections and 800 000 cases of full-blown AIDS in adults. This is in addition to an estimated 900 000 HIV-1 infections and 500 000 cases of AIDS in infants and children. Clearly, something extraordinary occurred in sub-Saharan Africa in order for these massive numbers of infections to appear in such an explosive manner since the late 1970s and early 1980s.

Furthermore, many of the original cases of AIDS can be traced back to sub-Saharan Africa. Bygbjerg (8) describes in detail the case of Dr Margarethe Rask, a surgeon who worked in rural northern Zaire from 1972–1975 and in Kinshasa, Zaire from 1975–1977. She died in Copenhagen in December 1977 after suffering from a disease with all the symptoms of AIDS—persistent diarrhea, fatigue, weight loss, generalized lymphadenopathy, candidiasis and *Pneumocystis carinii* pneumonia. Other authors (9–13) have documented early cases of Africans, particularly from Zaire, who presented themselves at hospitals in Brussels and Paris for treatment of clinical symptoms suggestive of AIDS. In 1985 Belgium was reported to have the highest incidence of AIDS in Europe due to Africans from Zaire (the former Belgian Congo) coming for medical treatment. Of the 99 cases reported in Belgium as of August 30 1985, 73 were from Central Africa, mostly Zaire (13). There is one unconfirmed report that Gaetan Dugas, the so-called 'Patient Zero' in the United States, became infected with AIDS by means of sexual contact with Africans (14).

Biggar (15) argues persuasively that these cases, as well as reports from clinicians working in Africa, point strongly to a fairly recent sub-Saharan origin for HIV-1. He notes a contrary theory (16–19) that HIV-1 has been endemic for many years in rural Africa, and its abrupt appearance around 1980 was due to people migrating from rural to urban areas. This theory proposes that the virus was then propagated through changing sexual mores, increased use of blood products in the practice of medicine and increased airplane travel. This seems unlikely, however, since the rural to urban migration in Africa has been going on for over 30 years. If HIV-1 had been endemic in rural areas, the disease should have appeared well before 1980 in urban areas, and it didn't. The clinicians Biggar (15) interviewed do not believe they could have failed to recognize AIDS as a distinct clinical entity if it had existed prior to the late 1970s or early 1980s.

The growth of HIV-1 infections worldwide and in sub-Saharan Africa since the late 1970s and early 1980s has been exponential (20) (Fig. 1). Thus, because of the long incubation period of HIV-1, as long as 10 years in some cases (21), we know that the virus had to have been infecting our species sometime in the decade before exponential growth began, i.e. the late 1960s and most of the 1970s. This is precisely the time period of the Intensified Smallpox Eradication Program.

The Haitian connection

Numerous authors have documented the high incidence of AIDS in Haiti and the possible transmission of some cases of AIDS into the United States by means of sexual contact with Haitians (19, 22–28). This is associated with Zaire, because there were thousands of Haitians in Zaire providing technical and teaching assistance to help the French-speaking people of Zaire after the Belgians left (23). There is one unconfirmed report that many of these Haitians were vaccinated against smallpox (29) simply because they were present during the time the smallpox eradication program took place. In any case, the time of appearance of AIDS in Haiti corresponds with the return of these professionals to Haiti (29, 30).

The American Red Cross, sub-Saharan Africa, and Haiti

The American Red Cross is also aware there was something about those sub-Saharan African countries and Haiti that led to a high incidence of AIDS. For many years in the 1980s, the Red Cross had the following question on the form to be signed before donating blood, '*Have you been to Haiti or Africa since 1977?*' This question was used until a new form was published in September 1990 which asks '*Have you been outside the United States in the last 3 years?*' If you answer yes, the follow-up questions concern

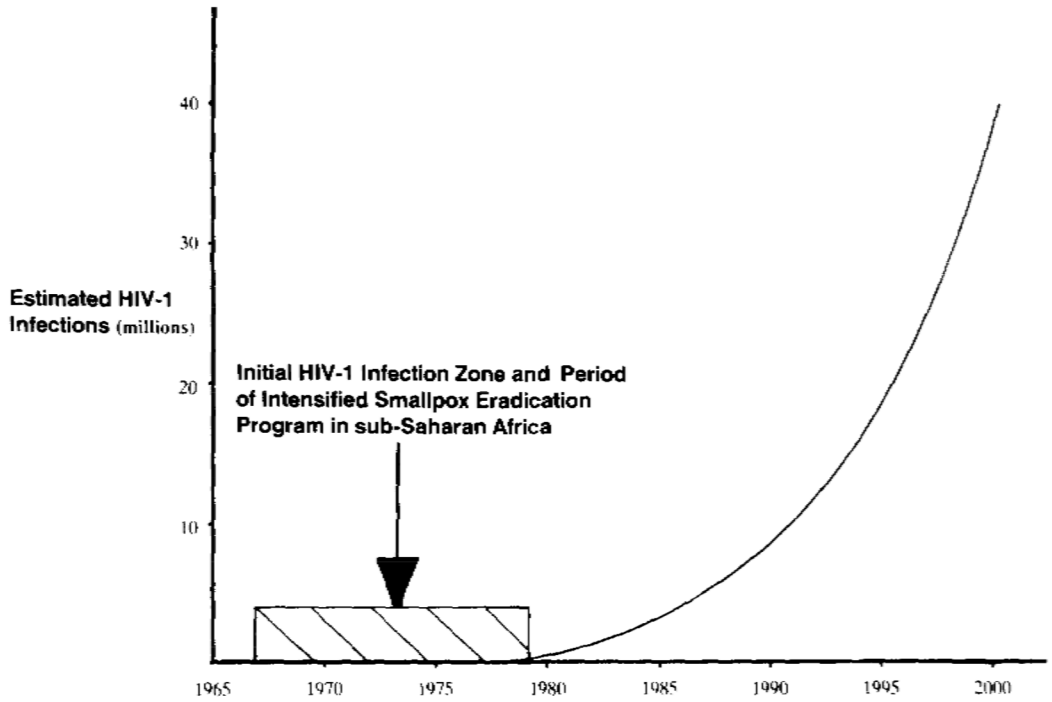

Fig. 1 Estimated worldwide HIV-1 infections versus time. Data from (6, 7).

travel to Haiti or Africa. Also, in the pamphlet entitled *What You Must Know Before Giving Blood*, November 1986, which was used until August 1988, there is the following statement, '*You are at risk of getting AIDS and spreading the AIDS virus if you are a native of Haiti, Burundi, Kenya, Rwanda, Tanzania, Uganda or Zaire who entered the United States after 1977*'. That sentence was changed in November 1987 and the new pamphlet substituted the words '*...if you are a native of Haiti, Sub-Saharan Africa or any island close to Sub-Saharan Africa*'. The newest form, dated February 1992, removes all references to sub-Saharan Africa and Haiti. Consequently, even though the actual names of these countries no longer appear on Red Cross forms, it is clear that the Red Cross and the Food and Drug Administration, which supplied the wording for the Red Cross forms (31), knew as early as the mid-1980s that the incidence of AIDS in sub-Saharan Africa and Haiti was exceptionally high. This, while not conclusive, is further indication that the origin was sub-Saharan Africa.

Bovine Leukemia Virus

Bovine Leukemia Virus (BLV) is an RNA retrovirus (32) which causes an immunosuppressive infection of cattle and sheep by replicating in the cells of the immune system (33). Its provirus is 8714 base pairs long (34). Normally, it takes 4–5 years (32) for symptoms of the disease to manifest themselves from the date of the infection, but the virus can be transmitted at any time after it enters the animal's bloodstream. Because it is slow acting and there are no visible signs of the disease, many animals remain in the preclinical stage for their entire lifetimes (32). Diagnosis before the onset of symptoms is by means of an ELISA (enzyme-linked immunosorbent assay) test (35) for antibodies to BLV similar to one of the tests used for detecting antibodies to HIV-1 in humans.

When the infection finally appears, two of the most common symptoms are lymphadenopathy – enlargement of the superficial lymph nodes – and a chronic, persistent diarrhea (32). After that, various cancers and diseases can infect the digestive tract, the heart, the nervous system, and other organs of the animal's body. This is similar to AIDS in humans.

The major modes of BLV transmission (36) are contaminated hypodermic needles used in blood transfusions and vaccinations, ear-tag applications and dehorning using the same equipment, rectal palpation using the same polyethylene sleeve from animal to animal, horseflies and colostrum from a seropositive mother to its offspring. In short, any mechanism that can transfer blood or any lymphocyte containing bodily fluid into the blood stream of another cow or sheep can transmit this disease. Johnson (36) has shown conclusively that as little as 1 μl of blood (a fraction of a drop) from a seropositive cow injected into calves intramuscularly, intravenously, subcutaneously or intradermally causes se-

roconversion within as little as 4 weeks after inoculation. In other words, a small blood to blood transfer or, more precisely, a small infected lymphocyte to blood transfer is enough to transmit BLV.

In 1985, Ratner et al (37) reported a nucleotide sequence homology of 62% between the long terminal repeat (LTR) sequences of BLV and HIV-1. Also in 1985, Sodroski et al (38) reported that HIV-1 shares with BLV a phenomenon connected with these LTR sequences, known as *trans*-acting transcriptional regulation. They believe this greatly speeds up the rate of gene replication which may account for the virulence of HIV-1 infection.

Visna virus

Visna is a slow acting, pathological disease first reported in sheep in Iceland in 1935 (39). (The word Visna is an Icelandic word meaning wasting or shrinking.) It is caused by an RNA retrovirus of the subfamily *Lentivirinae* or slow-acting viruses (40), and its provirus is 9202 base pairs long (41). This disease was first shown to infect cattle by Van der Maaten, Boothe and Seger in 1972 and 1974 (42, 43). Gonda et al, in 1985 and 1987 (1, 44, 45), clearly demonstrated a genetic, structural and serologic relationship between HIV-1 and Visna. Many authors (41, 46–52) have confirmed this close relationship between HIV-1 and Visna, and HIV-1 is now considered to be a member of the lentivirus subfamily (41, 53).

Like BLV and HIV-1, Visna may take years of incubation (32, pp805–807, 54) before symptoms of the disease appear, and many infected animals with no conspicuous clinical signs are slaughtered (55). When symptoms do occur, two of the most common in sheep are a chronic persistent pneumonia and significant weight loss (39, 40). Both of these symptoms are common in human patients with AIDS. Like HIV-1 (56), Visna crosses the blood-brain barrier and is found in brain cells and cerebral spinal fluid (57, 58). At autopsy, CNS lesions of chronic meningitis and demyelination are observed over the spinal cord and portions of the brain (40). Like BLV and HIV-1, tests for antibody to Visna must be performed by the veterinarian to confirm the existence of this disease prior to the onset of clinical symptoms (40).

The transmission of Visna has been studied less than that of BLV, but it is felt that hypodermic needles contaminated by using one needle for multiple injections is one cause (54). Another is viral infected lymphocytes in the colostrum being passed to the newborn (40). Again, the transfer of even small amounts of contaminated blood or lymphocytes to the bloodstream of another animal is suspected of transmitting this virus.

In 1985 Gonda (44) reported that 35% of the genome of HIV-1 was heteroduplexed (essentially identical) with Visna, a degree of homology he called 'striking'.

Prevalence of BLV and Visna

BLV is widespread in the United States and around the world (59). In Florida, antibodies to BLV were detected at a level of 48% in 7768 dairy cattle and 7% in 4911 beef cattle older than 18 months. In Michigan in the early 1980s, the prevalence rate varied from 24 – 36% in 3000 dairy cattle from 82 herds. Visna (BIV) antibodies were reported in 4% of 1997 cattle tested in eight states in 1989 (60). Van Der Maaten and Whetstone stated their belief that Visna (BIV) infections are widely distributed among US cattle, and they suspect Visna-BLV coinfections also exist (61). Amborski and her colleagues at Louisiana State University demonstrated in 1989 (62) that BLV and Visna can and do coinfect cattle, although no survey has been done to determine the percent of animals so coinfected.

BLV, Visna, and HIV-1

Thus, we have two animal diseases whose clinical symptoms are essentially identical to the clinical symptoms of AIDS. The T-cell (immune cell) affinity that AIDS demonstrates corresponds to the immunosuppressive characteristics of BLV. The dementia associated with AIDS corresponds to the ability of Visna to enter the brain (56), and the weight loss and pneumonia associated with AIDS correspond with the weight loss and pneumonia of Visna. Additionally, both retroviruses show a significant degree of nucleotide sequence homology to HIV-1 and, structurally, HIV-1 is indistinguishable from the lentivirus subfamily to which Visna belongs (44).

Consequently, it is reasonable to conjecture that HIV-1 was formed by BLV and Visna combining together in some manner and being transferred to humans. How could this have occurred? The answer lies in the rapid rate of genetic divergence of RNA viruses and in the production method for Vaccinia, the vaccine used to immunize humans against smallpox.

Recombination of RNA retrovirus genomes

The rate of genetic divergence of RNA viruses in general is very rapid (63). Studies have shown that the rate of divergence of RNA viral genomes at the nucleotide level is as high as 0.03–2.0% per year (64). This is in striking contrast to the divergence rate in the DNA genomes of animals and humans, approximately

1% per million years (65). In other words, some RNA viruses diverge as much as a million times faster than human DNA. James and Ellen Strauss (66) have pointed out that this rapid RNA sequence divergence rate has important implications for RNA viruses as disease pathogens.

One of the ways in which this divergence manifests itself is in recombination—the exchange of gene segments between two parental viruses during a mixed infection to form offspring with characteristics from both parents (66). It has been shown in the laboratory that true recombination occurs among the picornaviruses and the coronaviruses (63). Hahn, Lustig, E. Strauss and J. Strauss (67) have shown that western equine encephalitis virus (WEEV), a 'successful' widespread virus which causes encephalitis in humans, is a natural recombinant of eastern equine encephalitis virus (EEEV) and a Sindbis-like virus. RNA recombination is now considered to be an important force in the evolution of all types of RNA viruses (63, 67).

In the special case of retroviruses which contain two molecules of single-stranded RNA and reverse transcriptase (RT) in their core, it has been shown (68) that they undergo the highest rate of recombination of any biological system known. (BLV, Visna, and HIV-1 are such retroviruses.) A typical experiment is to infect a cell culture with two genetically different retroviruses and then examine the offspring after a few replication cycles. As many as 40% of the progeny can be found as recombinants (69), in that all contain nucleotide sequences derived from each parent. Genetic experiments with two avian retroviruses (70, 71) have shown that the frequency of recombinant virus from mixed infections exceeds 10%. Hunter (70) proposed the formation of a heterozygote virus from a coinfected cell which then would produce the recombinant virus during the second cycle of infection as shown in Figure 2.

The exact mechanism by which this recombination occurs is subject to some debate (68, 70, 71), but the fact that recombination does occur from retroviral coinfected cells after multiple cycles of infection is not in dispute.

Amborski's demonstration (62) that BLV and Visna coinfect cattle in nature does not prove that these viruses coinfect the same cells, but it does strongly suggest the possibility. If BLV and Visna coinfect the same cells, HIV-1 would be a possible recombinant.

This is supported in addition by the following facts:

1. The genomes of all three are approximately the same size (BLV–8714 nucleotides, Visna–9202, and HIV-1–9749 (37)).

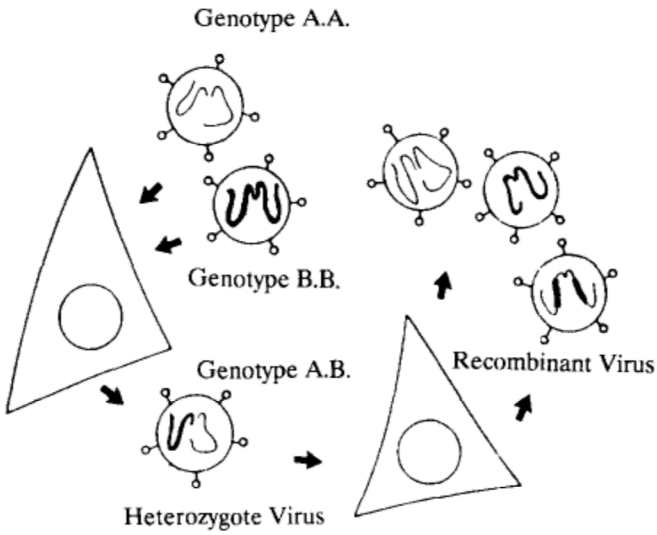

Fig. 2 Proposed two cycles of infection required for recombination between two genetically different RNA retroviruses. Adapted from (70). Used with permission.

2. HIV-1 contains sequences from both BLV and Visna as a natural recombinant would.

3. All three are members of the same retroviral family, *retroviridae* (72).

Smallpox vaccine manufacture

It is well known that Vaccinia virus replicates in humans, cows and sheep. The antibodies produced against this virus protect humans against Variola, the smallpox virus. In order to manufacture smallpox vaccine, the following steps are generally followed (73, 74):

1. Shave the skin of a cow or a sheep, called the vaccinifer, and disinfect the skin with soap and water and/or quarternary ammonium salt or alcohol.

2. Scarify the skin with a scarifying instrument which looks like a narrow ice scraper with 7 or 8 sharp needles on the end with the points approximately 5 mm apart, so that when drawn across the skin it leaves 7 or 8 parallel cuts in the skin roughly 5 mm apart. This scarification is made horizontally, vertically and then diagonally over the same area. Successive areas approximately a foot square are usually done on the abdomen and flanks.

3. The seed lot Vaccinia virus is thoroughly rubbed into the scarified areas and allowed to grow for 4 days at which time the skin is again thoroughly washed with soap and water to remove any bacteria. The animal is then killed humanely and the blood washed from its body in order to obtain water clear vaccine.

4. The vaccine 'pulp' is harvested by using a scoop shaped instrument similar to a sharpened spoon to

scoop out the infected areas from the scarified portions of the vaccinifer's epidermis.

5. This pulp is then processed by homogenizing it with a fluorcarbon in a blender, centrifuging it to form a clear aqueous phase, and treating it with phenol to kill any bacteria which might have been picked up in the process. The resulting clear 'lymph' is then ready to be freeze-dried or used as the vaccine.

Practically all of the vaccine used in the Intensified Program was freeze-dried for shipment, reconstituted in the field, and injected into humans (4). This injection was a lymphocyte to blood transfer from cows and sheep to humans.

Proposed method for this procedure transferring HIV-1 to humans

Simple mechanical transfer is a means by which the above procedure could transmit HIV-1 to the vaccine and then to humans. HIV-1 has been isolated from epidermal Langerhans cells of human AIDS patients (75), and Langerhans cells, which are an important part of the immune system (76), are common to the epidermis of mammals (77). Consequently, if HIV-1 was already in the vaccinifer, some of it would be in its epidermis. Vaccinia virus was grown in the epidermis of the vaccinifer, and even if the Vaccinia virus did not coinfect a cell already infected with HIV-1, some HIV-1 contaminated cells would be picked up during the 'scooping out' of the infected portions of the vaccinifer's skin and transferred to the vaccine. It was standard procedure (74) to pool individual harvests from one animal with harvests from others to achieve a minimum volume for freeze-drying, and 400 doses of vaccine (0.0025 ml each) could be obtained from 1 ml of vaccine using bifurcated needles (4, p569). Thus, a significant amount of vaccine could have been contaminated from even one diseased animal.

Enhancement of the amount of HIV-1 in the epidermis of the vaccinifer

If Vaccinia virus coinfects a cell already infected with HIV-1, there is a possible mechanism by which the amount of HIV-1 in the epidermis of the vaccinifer might be increased before harvest. This mechanism is the integration of HIV-1 into the Vaccinia genome.

HIV-1 (78), upon entering the host cell, has its RNA transcribed by RT into proviral DNA which exists in three forms—a linear form with one long terminal repeat (LTR) sequence at each end, a circular or plasmid form with two LTRs and a circular or plasmid form with one LTR (Fig. 3). This proviral DNA is then integrated into the host cell's nuclear DNA where it directs viral replication to continue the life cycle of the virus.

Brown et al (79) show that this integration is dependent on the LTR seqeunces of the proviral DNA, and it is quite possible that the circularized (plasmid) 2-LTR form of the proviral DNA is the immediate precursor to the integrated DNA. It is important to note that a significant amount of unintegrated DNA remains in the cytoplasm of the host cell in all three forms—linear with one LTR at each end, 2-LTR circular, and 1-LTR circular (78). It should also be noted that this mechanism of infection, life cycle, and retention of proviral DNA in the cytoplasm is identical to that of Visna virus (80, 81).

It is also known that Vaccinia virus can coinfect cells already infected with RNA viruses (82), and that Vaccinia's transcription and translation take place totally in the cytoplasm of the host cell (82, 83). The first step after infection is the uncoating of the Vaccinia virus core (84). Thus, if Vaccinia coinfected an already HIV-1 infected cell during vaccine manufacture, the uncoated Vaccinia virus DNA core would then be in the cytoplasm of the HIV-1 infected cell along with the 2-LTR circularized form of proviral HIV-1 DNA. This 2-LTR circularized HIV-1 DNA could then integrate itself into the Vaccinia genome by the same procedure it used to integrate itself into the host cell's genome originally.

This would be analogous to the work of Fenner, Mackett, Smith, Moss and Paoletti et al (83–90) who have shown the ability of Vaccinia virus to serve as a foreign gene expression vector. By constructing plasmids containing a foreign gene under the control of a Vaccinia virus promoter flanked by Vaccinia virus DNA, many foreign genes have been inserted in to Vaccinia and expressed from Vaccinia virus recombinants (82–86). Panicali, Paoletti, and Perkus (87–90) have shown that there are at least 13 sites on the Vaccinia genome where genes can be inserted without interfering with the ability of Vaccinia to replicate. They also showed that this integration of foreign genes into Vaccinia and their subsequent expression can be achieved without directly engineering Vaccinia promoters. It is, therefore, reasonable to conclude that the 2-LTR sequences on the plasmid form of HIV-1 could serve as the promoter for the integration of the entire 9749 bp HIV-1 genome into the Vaccinia virus DNA. Because the Vaccinia virus genome is large, about 187 000 base pairs (82), it has the capacity to accept genes containing as many as 25 000 base pairs (91). Thus, there is more than adequate room for this integration to take place. Once it does, HIV-1 could be expressed in that cell or by the next cell that the Vaccinia/HIV-1 recombinant infects.

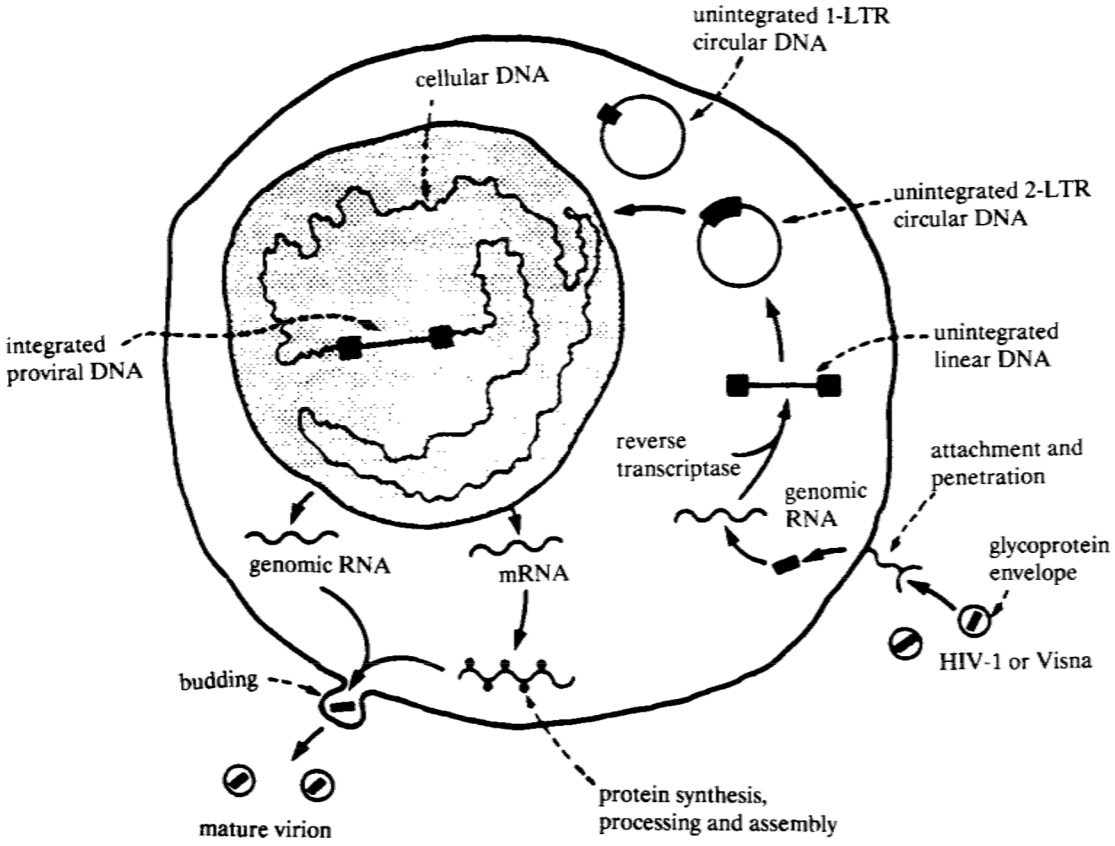

Fig. 3 The life cycle of HIV-1 and Visna. Adapted from Fauci A S. The Human Immunodeficiency Virus: Infectivity and Mechanisms of Pathogenesis. Science 1988; 239: 617–622. Used with permission.

This suggested mechanism would increase the concentration of HIV-1 in the epidermis of the vaccinifer beyond its concentration in the absence of the Vaccinia infection.

How AIDS was created

We now have the following theory for how AIDS was created: at least one of the cows and sheep used in the manufacture of smallpox vaccine, unknown to those conducting the procedure, was coinfected with BLV and Visna at the stage in their development when visible symptoms of the associated diseases were not yet present. The animal or animals would have appeared healthy, but the BLV and Visna had already naturally recombined into HIV-1. The concentration of HIV-1 in the epidermis of the vaccinifer may or may not have been increased during the 4 days of Vaccinia infection by the mechanism discussed in the previous section. In either case, the HIV-1 was then transmitted to the vaccine along with the Vaccinia virus and subsequently to humans during vaccination.

What does this theory explain?

1. It explains why we have no immunity to HIV-1. It is basically an animal disease.

2. It explains why the disease in Africa occurred with approximately equal frequency in men and women (92, 93), since there were roughly equal numbers of men and women in the vaccination lines.

3. It explains why the disease was so prevalent in Haiti but not in the Dominican Republic, which shares the same topography, climate and island.

4. It explains the complexity of the AIDS virus without it having gone through thousands of years of evolution, an issue raised by Montagnier (17).

5. It explains why the decade of the 1970s was critical for the introduction of this virus into humankind. This was the time period of the smallpox inoculations.

6. It explains why some countries such as Ethiopia and the Sudan that participated in the Intensified Eradication Program did not have a problem with AIDS. Only a part of the smallpox vaccine was contaminated. The manufacturing process is a batch process, and we can conclude that only some of the batches of vaccine that went to Zaire, Zambia, Rwanda, Burundi, Malawi, Tanzania and Uganda were contaminated.

Weak points of this theory

1. It does not explain some of the early reported cases of AIDS, especially the case of Robert R, a St Louis teenager who died in 1969 of a disease that was identical to or at least closely related to HIV-1 (94, 95). The early date of this case suggests a conflict with this theory, but this is not clear. We do not know enough about Robert R's medical history. There have also been reports of positive antibody tests to HIV-1 in frozen blood samples from Africa prior to 1969 (96). However, this is not conclusive since these test results may follow from prior exposure to malaria or to human leukocyte antigen due to pregnancy (97–100). Additionally, the positive test results reported in one study came from children who had been vaccinated against smallpox in Upper Volta (96).

2. It does not explain the origin of HIV-2, another human retrovirus capable of causing AIDS that is more homologous to simian retroviruses than HIV-1 (19, 101). At this time we can offer only the following note that may eventually lead to a resolution of this issue: the smallpox vaccination program in approximately 20 countries of Central and Western Africa (102) at the beginning of the Intensified Program from 1967 – 1971 used vaccine supplied and manufactured under bilateral agreements with the United States Agency for International Development (103). Today the prevalence of HIV-2 is highest in those Central and West African countries where these bilateral programs took place (104).

3. It does not explain why AIDS has appeared now, and not previously. We have, after all, used this same method of vaccinating against smallpox for almost 200 years. This is perhaps the strongest argument against this theory. However, the creation of AIDS was a small likelihood event. It required the presence of both BLV and Visna in one animal, and it required the use of that animal as a vaccinifer for the production of Vaccinia virus. Furthermore, since the veterinary literature does not define BLV as a known virus until the late 1960s and early 1970s (105–109), since BLV is exogenous to cattle (110), and since the first report of Visna in cattle did not occur until 1972, it may be that BLV and Visna simply were not present in the animals used as vaccinifers.

On the other hand, it is entirely possible that other human diseases might have been created in this manner, and we are not aware of it. For example, one author (57) has noted some similarities between Visna and human demyelinating diseases such as multiple sclerosis, and the National Multiple Sclerosis Society has supported research on Visna virus (111). Other authors have noted an evolutionary relationship between BLV and human T-cell leukemia viruses (112).

Summary

HIV-1 is a natural recombinant of BLV and Visna virus inadvertently transferred to humans through the procedure by which Vaccinia virus, grown in coinfected cows and sheep, was used to manufacture the vaccine that inoculated millions of Africans against smallpox in the late 1960s and the 1970s.

Conclusions

A low cost, plentifully available animal model for AIDS would be of great assistance in developing a cure or a vaccine. The Gibbon ape and chimpanzee (53, 113, 114) are infectable with HIV-1. However, neither of these animals is plentiful enough for the experimental work that needs to be done, and even those infected years ago do not develop clinical symptoms (115). More recently, rabbits (116) and pigtail macaques (117) have shown promise as animal models for defining the initial stages of HIV-1 infection, but it is not yet clear that either of these animals will serve as a means of study to reduce or eliminate the disease in infected persons. To do that, it would be helpful if the animal would develop clinical symptoms after seroconversion (118).

It is proposed that a study be undertaken to determine whether cows and sheep can be used as animal models for HIV-1. (To the author's knowledge, this has not yet been done.)

This would involve screening a variety of species of cows and sheep to certify them free from BLV and Visna and dividing them into three groups. Group 1 would be the control group. Group 2 would be directly injected with human HIV-1 virus. Perhaps these animals would develop antibodies to HIV-1 and subsequent symptoms of AIDS.

Group 3 would be used in an experiment which essentially reverses the methodology of making smallpox vaccine described above. Human tissue culture known to contain HIV-1 would be deliberately infected with Vaccinia and allowed to incubate for 4 days. This mixture would then be directly injected to see if antibodies to HIV-1 and subsequent AIDS symptoms develop. This latter procedure also could be tried with smaller animals such as shrews, mice, hamsters, guinea pigs and various monkeys which have not seroconverted after direct injection of HIV-1 blood components (119, 120). If successful, these experiments would yield an animal model for HIV-1 which will assist AIDS research.

Pharming is a new process using genetically altered cows, sheep, goats and pigs to produce human hormones and other drugs from their milk or blood. In the light of the danger of this technology generat-

ing another unknown human disease, any animal used for pharming should be thoroughly examined for the presence of retroviruses such as BLV and Visna.

This is an urgent matter. People are dying and new infections are occurring everyday. We must explore every avenue which offers any hope of success.

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