VIRUSMYTH HOMEPAGE

A CRITICAL ANALYSIS OF THE EVIDENCE FOR THE EXISTENCE OF HIV AND THE HIV ANTIBODY TESTS

Presentation 12th World AIDS Conference

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Geneva 28 June 1998

The only way to prove the existence of a virus is to isolate its particles. It is only by doing this that we obtain pure particles to inspect, and analyse, and to introduce into fresh cell cultures to prove particles make more of the same. After all, no matter how viral-like they may look, this is what particles must show us before they ever earn the title, virus.

Have HIV experts gone to all this trouble? No. The only reason we have HIV is antibodies. A few antibodies amongst the plethora in AIDS patients that react with a few proteins present in the lymphocyte cultures of AIDS patients. When it is all said and done, it's not just that antibodies are used to prove some individuals are infected with HIV. For the HIV protagonists, antibodies are the proof that they have isolated HIV.

Shortly, Eleni Papadopulos-Eleopulos will guide us through a close look at this proposition. As she does take special note of the history of the discovery and demise of the world's first human, leukaemia retrovirus, HL23V. As in the case of HIV, Gallo and his colleagues claimed that antibodies elevated certain culture proteins to the status of a virus. Their reasoning was then and still remains a scientific impossibility. When it was discovered that such antibodies were induced by a wide variety of stimuli that had nothing to do with viruses, and they occurred in far more healthy people than could have ever had the virus to cause leukaemia, HL23V disappeared from the annals of science. This is why most of you, and most HIV experts, have never heard of it. Yet in the AIDS era we have the same method used to prove the existence of HIV, and a large number of instances of HIV antibodies where there is no HIV. But we still believe in HIV.

The message for us tonight is plain and simple. HIV might exist but there is no proof that it does exist. As you listen to what is still the best evidence for HIV, imagine it is 1983, you are the consummate, disinterested scientist, living in Paris, working at the Pasteur Institute and charged with the task of discovery. Try to decide, each one of you, what you discovered. Was it a retrovirus HIV or have you let down your guard and allowed the immune system to trick you once again with antibodies which mean something entirely different but which you have mistakenly, once again called a retrovirus and HIV?

As far back as 1974 German researchers knew that:

The proteins of molecular weights between 10,000 and 30,000 of retroviruses of a given species share antigenic determinants.

In fact they knew that the proteins of molecular weights of about 30,000 from retroviruses of many species, including monkeys, also shared antigenic determinants.

This means that the *gag* and *pol* proteins of retroviruses and antibodies directed against them are non-specific. In other words, finding an antibody to one or more of these proteins is not proof that a particular retrovirus is present. This is acknowledged by the well known HIV/AIDS expert William Blattner.

> "Viral proteins, particularly the polymerase and *gag* proteins may be highly conserved between subtypes of viruses."

Nowadays we know that antibodies to "retroviral" proteins are widespread in human populations even when there are no retroviruses present.

In the mid 1970's Robert Gallo and his associates claimed proof for the existence of the first human retrovirus, HL23V.

They found reverse transcriptase activity in the uncultured lymphocytes of a patient with leukaemia.

When the cell free fluids from lymphocyte cultures were banded in sucrose density gradients at the density of 1.16g/ml they found:

1. Retrovirus like particles
2. Reverse transcriptase activity
3. Proteins which reacted with the patient's sera

The HL23V antibodies were confirmed by Reinhard Kurth and Robin Weiss, who concluded:

> "The serological studies presented have and by others provide indirect evidence that the infectious mode of transmission remains a real possibility in humans, and suggest that infection with an oncovirus [retrovirus] may be extremely widespread"

However, by 1980, two research groups, one from the National Cancer Institute, and the other from the Sloan-Kettering Cancer Center had shown that "the antibodies in question are elicited as a result of exposure to many natural substances possessing widely cross-reacting antigens and are not a result of widespread infection of man with replication competent oncoviruses."

In 1981 Gallo accepted that the antibodies were not HL23V antibodies.

This finding had such a profound significance in regard to the existence of HL23V, that today nobody, not even Gallo, claims that HL23V ever existed.

Thus, before the AIDS era ample evidence existed revealing it is impossible to prove the existence of or infection with a retrovirus by an antibody test. The question then is: how is it possible that millions of people around the world have been told that they are infected with HIV on the basis of an antibody test?

To perform an HIV antibody test one needs:

- HIV proteins, that is HIV antigens,
- a blood sample,
- and a gold Standard.

The latter is necessary because finding a reaction between the HIV proteins and antibodies present in the blood does not prove that the blood contains HIV antibodies, that is, antibodies specifically induced by HIV.

There is a very good reason for this. Antibodies directed against antigen other than the HIV proteins, may also react with the HIV proteins.

This so called cross-reactivity, is especially likely to happen when the sample is from AIDS patients and those at risk, since their blood is known to have antibodies against many infectious agents as well as auto-antibodies.

Because of this, the test specificity must be determined by using a gold standard, that is, by comparing the results of an antibody-antigen reaction with the ability to isolate HIV.

If there are such entities as HIV antibodies they will manifest if and only if it is also possible to simultaneously isolate HIV from the patients' cultures.

The procedure for isolating (purifying) retroviruses is well known:

Layers of sucrose solution (bands) of known density are set up in a centrifuge tube.

A sample from a culture is gently placed on the top of the solution and the tube spun at high speed for many hours whereupon retroviral particles will descend though the sucrose solution until their progress is arrested by a layer of the same density.

By definition retroviruses are arrested, that is, they band in a layer where the density of sucrose is 1.16 gm/ml.

To claim isolation of HIV by this method one must show:

1. Firstly, that the culture cell-free fluids contain particles having morphological characteristics of retrovirus the principle of which are:

    1. a diameter of 100-120nm
    2. condensed inner cores
    3. surfaces studded with spikes

2. When these fluids are banded in sucrose gradients the particles accumulate, that is, band, at the density of 1.16g/ml.

3. At that density there is nothing but retroviral-like particles.

Once this is proven, one proceeds to characterise the particle proteins and nucleic acids. If the particles are retrovirus, they should contain only RNA and not DNA, as well as an enzyme, reverse transcriptase, that copies RNA into DNA.

Because not all particles which look like retrovirus are infectious, that is are viruses, one must then prove that the particles which band at 1.16g/ml are indeed infectious.

5. When the particles are introduced into secondary cultures:

    1. the particles are taken up by the cells
    2. the entire RNA is reverse transcribed into cDNA
    3. the entire cDNA is inserted into cellular DNA
    4. the DNA is transcribed into RNA which is translated into proteins

6. The cells in the secondary cultures release particles into the culture medium.

7. The particles in the secondary cultures have exactly the same characteristics as the original particles, that is, they must have identical morphology, band at 1.16 gm/ml and contain the same RNA and proteins.

Indeed this is the method which Luc Montagnier and his colleagues claimed to have used to isolate HIV and thus to prove its existence.

The May 1983 study by Montagnier and his colleagues was entitled:

> "Isolation of a T-Lymphotropic Retrovirus from a patient at Risk of Acquired Immune Deficiency Syndrome (AIDS)"

They had a patient, BRU, a gay man who had lymphadenopathy.

Lymphocytes from BRU's lymph nodes were cultured. To the culture they added many stimulating agents.

They claimed that after a few weeks in the culture they could find the enzyme reverse transcriptase, RT.

In fact, they did not prove the existence of the enzyme, but of reverse transcriptase activity. That is, the ability of the culture to make DNA from RNA.

They put in a culture a long stretch of synthetic RNA, An, an RNA template. To one end of this they attached a small piece of synthetic DNA, dT15, a primer.

The outcome of this was that a long stretch of DNA was found to have been reverse transcribed from the RNA.

This was interpreted as proof that the culture and thus BRU's lymphocytes were infected with a retrovirus.

In a second experiment they added BRU's lymph node cells to a stimulated culture of lymphocytes from a healthy blood donor.

In this co-culture, they also found reverse transcriptase activity. This was interpreted as proof for the transmission and isolation of a retrovirus.

In yet another experiment cell-free fluids from the co-culture were added to cultures of umbilical cord T-cells.

In this culture they found:

1. RT activity
2. "typical type-C" retroviral particles

These findings were interpreted as proof that the retrovirus from BRU was transmitted to the donors T-cells and then to the umbilical cord T-cells.

There are many problems with this interpretation:

1. The enzyme RT is not specific for retroviruses. Indeed, since in 1972 Gallo showed that reverse transcriptase is found in normal non-infected cells, and in 1973 Barre-Sinoussi and Chermann acknowledge this fact, it is curious that ten years later they claim that the RT in this case proved HIV infection.

2. Furthermore, they did not prove the existence of an RT enzyme but only the ability of the cultures to reverse transcribe, that is, RT activity. However, the template primer An.dT15 can be transcribed not only by RT but by the normal cellular DNA polymerases.

3. Not all particles which look like retrovirus are viruses.

The important point is that Montagnier's experiments do not prove the existence of a retrovirus because, as Montagnier himself admits, all the phenomena he described are non-specific for retroviruses, or even for viruses.

Suffice it to say that two of the best known retrovirologists say:

> "Particles and proteins, could reflect non-viral material altogether."

> "Release of virus-like particles morphologically and biochemically resembling type-c virus but apparently lacking the ability to replicate have been frequently observed from leukaemic tissue."

Proof for the existence of a retrovirus requires purification (isolation) of particles, analysis of their constituents, introduction of pure particles into uninfected cells followed by a second analysis to prove that the particles produce identical progeny (see introduction). Only then can one claim to have proven a virus exists.

In the interview Montagnier gave to the French Journalist Djamel Tahi at the Pasteur Institute in July 1997 he was asked why he did not use immortal cell lines. His answer was that in such immortal cell lines such as MT2, MT4 (and thus HUT-78) "You have a mix of HIV and HTLV. It is a real soup."

But Montagnier may also have had a soup. There is a very good reason for this. Long before the AIDS era, retrovirologists knew that there was a significant difference between retroviruses and all other viruses. Unlike other viruses, retroviruses, especially in culture and under the conditions used by Montagnier, could arise even if the cells were not infected to begin with or if no retrovirus was introduced from outside. These retroviruses had become known as endogenous retroviruses.

Some of the best evidence for the existence of endogenous retroviruses comes from reproductive tissues including placenta and umbilical cord.. By using umbilical cord Montagnier had a soup of a different flavour, but still a soup.

Even if Montagnier had proof for the existence of a retrovirus in his umbilical cord lymphocytes, the retrovirus could not have originated in BRU. There are two very good reasons for this:

1. All cells, infected or not, release retrovirus particles in cultures, especially when the culture conditions are those used by Montagnier et al. As far back as 1976, eminent retrovirologist wrote, "the failure to isolate endogenous viruses from certain species may reflect the limitations of in vitro cocultivation techniques". In other words, given sufficient time and ingenuity, a researcher can produce retroviral particles from any cell.

According to all HIV experts, for the HIV particles to be infectious, gp120 is absolutely necessary.

gp120 is present only in the particles knobs.

However: To date, nobody, not even Hans Gelderblom, has found proof for the existence of these knobs in the cell-free HIV particles.

Since, into the umbilical cord lymphocyte culture Montagnier and his colleagues introduced cell-free fluids from the co-culture. It means that even if the cell-free fluids contain particles they could not have been infectious.

According to Montagnier and his colleagues, definite proof that they have a retrovirus was obtained by isolating the particles and showing that in sucrose density gradients the particles band at the density of 1.16g/ml.

In fact Montagnier and many other researchers since then have claimed that the 1.16g/ml band contained "pure virus", that is particles isolated from everything else, as the title of his paper proclaims.

However, neither Montagnier nor anybody else, up till 1997 had published any electron micrographs to prove that the 1.16g/ml band contained purified virus particles or indeed any particles, viral or otherwise.

The reason for this, at least in Montagnier's case, became obvious in his interview with Djamel Tahi in July 1997. Montagnier was asked, "Why do the EM photographs published by you [in 1983] come from the culture and not the purification?". His reply was, "There was so little production of virus it was impossible to see what might be in a concentrate of the virus from the gradient ["pure virus"]. There was not enough virus to do that. Of course one looked for it, one looked for it in the tissues at the start, likewise the biopsy. *We saw some particles but they did not have the morphology typical of retroviruses.* They were very different. Relatively different. So with the [unpurified] cultures it took many hours to find the first pictures. It was a Roman effort!…Charles Dauget [an EM expert] looked at the plasma, the concentrate, etc…he saw nothing major" (italics ours).

When asked if Gallo had purified HIV he replied: "I don't know if he really purified. I don't believe so."

In this case, this should have been the beginning and the end of HIV.

If there are not even retrovirus-like particles in the "purified virus", there can be no HIV and thus:

1. No HIV RNA and thus no probes and primers for hybridisation and PCR studies.
2. No viral load measurements.
3. No HIV proteins and thus no antibody test and no sero-epidemiology.

According to Montagnier on the one hand: "Analysis of the proteins of the virus demands mass production and prufication."

but on the other...

"I repeat, we did not purify!"

Thus Montagnier did not meet his own criteria for defining the HIV proteins.

Instead, Montagnier extracted the proteins from the umbilical cord lymphocytes and placed them on a strip according to their molecular weight. He then exposed the strip to BRU's serum as well as serum from a healthy donor and normal goat serum and from a goat immunised with HTLV-1. Many proteins reacted.

He repeated the experiment with the proteins which banded at 1.16g/ml. Only p24, p41, p80 reacted. He made no comment on p80, but Gallo later showed it to be a cellular protein.

Because p41 (which Montagnier called p45) reacted with all the sera and because a ubiquitous cellular protein, actin, has a molecular weight of 41,000, he said that p41 was actin which "contaminated" his "purified virus".

Because they found p24 to react only with BRU's serum, they assumed that p24 is an HIV protein and the antibodies specifically induced by HIV.

However, from an antibody-antigen reaction is impossible to determine the origin of even one reactant. Yet Montagnier claims to have determined the origin of both.

There is ample evidence which shows that the 1.16g/ml material contains both actin an another ubiquitous cellular protein, myosin, whose light chains have molecular weight of 24,000.

Montagnier himself showed that AIDS patients and those at risk have high levels of antibodies to both actin and myosin.

There is ample evidence that p24 reacts even more often than p41 with sera from healthy individuals and patients who are not at risk of AIDS.

For example: According to the AIDS vaccine clinical trials group the p24 band was common among young healthy volunteers.

In 1988 Lundberg showed that a p24 band was present in 163/1306 (12%) and for p41 129/1303 (10%) of individuals.

But only 25% of AIDS patients have a p24 on the HIV Western blot.

Thus by Montagnier's own criteria if p41 is a cellular protein, then p24, must be also.

It is significant that up to 1987 anybody, and up to 1990 any of the 5,000 gay men participating in the MACS study who had even a single p24 band in the Western Blot, was said to be infected with HIV.

By this criterion 4% of healthy individuals, 60% of hepatitis B sufferers and 39% of patients with biliary diseases would be said to be infected.

There is ample evidence to show that not only p24 and p41, but all the other proteins which are said to be HIV, are cellular proteins.

For example, in the well known Bess and co-authors 1997 *Virology* paper, the authors had 3 HUT-78 cultures, two infected and one uninfected control. (The HUT human cell line is that used by Gallo to "isolate" HIV in 1984 and since). The proteins from the banded material from all cultures including the control (lane A), which they called "mock virus", were compared using electrophoresis. They stated that the only difference between the 3 strips was that the infected strips contained major bands of p24, p17 and p6/7 and called them HIV proteins. But these same bands, although weaker, can also be seen in the "mock" virus protein strip whereas, to be HIV proteins, requires them to be present exclusively in the "infected" strips. When asked for proof that p24 etc in the strips B and C were HIV proteins their answer was that the labels were added for the reader's convenience at the suggestion of the reviewers.

So Bess and his colleagues have shown that the same proteins are present in the pure HIV and "mock" virus. (*Virol*. 230:134-144)

In their effort to develop a vaccine, and because humans cannot be injected with either HIV or "mock" virus, Bess and his colleagues first injected macaques with the "mock" virus. (This is culture fluids from the uninfected H9 clone of the human HUT78 cell line "purified" as it would be to obtain "HIV" or "SIV"). After the initial immunisation, the animals were given boosters at 4, 8 and 12 weeks. At fourteen weeks, the the monkeys were challenged with intravenous SIV prepared from the same human cells as "mock" virus and then monitored for seroconversion with the SIV Western blot. According to the authors, the animal immunised with "mock virus" "did not seroconvert to viral proteins after intravenous challenge with SIV", and "These results are the first demonstration that immunisation with purified cellular protein can protect from virus infection...It has recently been suggested that immunisation with alloantigens might serve as a vaccine to protect against HIV infection. Our demonstration...support this concept".

The underlying principle of immunisation is its specificity. That is, to protect against microbe 'X', the person or animal must be exposed to material from 'X' in order that the immune system generates specific antibodies. For example, immunisation with hepatitis vaccine does not protect agains poliomyelits. Since monkeys immunised with proteins derived from uninfected human cells are protected from infection with 'SIV' prepared from the same uninfected human cells, "mock" virus and "real" SIV must be identical. If such "mock" virus and "SIV" are one and the same we would expect that when "SIV" is prepared in antigenically different cells, for example, monkey cells, there will not be "protection". This is in fact what Bess and his colleagues proved in another experiment. The only logical explanation of these data is that they reflect immune responses to cellular proteins. Thus and SIV proteins, and thus by inference, HIV proteins, are nothing else but cellular proteins.

Interestingly, so far the only evidence of an animal model for AIDS has been obtained by allogenic stimulation, a procedure which leads to the appearance of "type C particles". Since individuals belonging to the AIDS risk groups are repeatedly subjected to alloantigenic insult, one would expect these individuals to have a positive antibody test, and not be surprised if they developed AIDS, without ever coming in contact with a retrovirus, HIV.

To claim that a stretch of RNA is the genome of a retrovirus it is absolutely necessary but not sufficient to prove:

1. The existence of a unique molecular entity
2. The molecular entity originates from a retroviral particle

Instead, Montagnier and his colleagues added to the 1.16g/ml material in which there were no retroviral-like particles, a short synthetic DNA primer and the substrates needed for DNA synthesis.

They claimed the detection of a DNA which was the result of reverse transcription of an RNA rich in adenine and concluded that the RNA was the HIV RNA and the DNA the HIV provirus.

There are many problems with this interpretation including the following:

Regardless of how or what RNA or DNA is obtained, without proof for the existence of retrovirus-like particles as 1.16 gm/ml it is impossible to claim that such RNA or DNA is the genome of a retrovirus.

To do so would be like conducting a paternity suit where the sample for DNA used for testing may not have even come from a man, any man at all, not to mention the defendant.

RNA rich in adenine is not specific to retroviruses. Any RNA can be reverse transcribed into DNA and reverse transcription is not specific to retroviruses.

There is no data in the Montagnier experiments to exclude the RNA being cellular.

As far as the sequences are concerned, the HIV genome is reported to have immense variations, while for all other RNA viruses, the variation is about 1%.

According to Peter Duesberg...

> "There is a range, in which you can mutate around without too much penalty, but as soon as you exceed it you are gone, and you are not HIV any longer, or a human any longer... then you are either dead or you are a monkey, or what have you."

By comparison, two RNA containing viruses (polio and influenza, the latter after 27 years of dormancy,) vary by less than 1% as do RNA molecules self-assembled in test tubes denied the organising propinquity of living cells.

To date, no two, same HIV genomes have been reported.

While the difference between the human and the chimpanzee's genome is no more than 2% the difference in the "HIV genomes" may exceed 40%.

In fact, the vast majority, more than 99% of the HIV genomes, are defective, that is, they lack part of whole of one or more genes.

The first, absolutely necessary step in proving that the RNA and thus the complementary DNA are the genome of a retrovirus which infects the T-cells of AIDS patients and those risk, is to have positive hybridisation results.

Again, for some unknown reason, Montagnier did not report such data.

Gallo did and Gallo's results were negative.

The HIV genome was resurrected by the polymerase chain reaction.

However, in addition to the many other problems mentioned so far:

1. With PCR one detects only small fragments of nucleic acid sequences which means that a positive PCR is not proof for the existence of the whole genome.
2. PCR reproducibility and specificity have not been determined.

To date only one study, by French researchers, has been published where such effort was reported.

These authors did not use the only scientifically valid gold standard, that is HIV isolation. Instead, as a gold standard, they used the antibody test. Even so they found that:

> "false-positive and false-negative results were observed in all laboratories (concordance with serology) ranged from 40% to 100%"

The same data revealed that in the seven French laboratories with extensive experience in PCR detection of "HIV DNA", of 138 samples shown to contain "HIV DNA", 34 (25%) did not contain "HIV antibodies" while of 262 specimens that did not contain "HIV DNA", 17 (6%) did contain "HIV antibodies".

Even with PCR nobody has ever presented proof for the existence of the whole HIV genome in even one, single cell of even one AIDS patient.

In fact, even a part of the "HIV" genome cannot be detected in all patients.

A striking feature of the results obtained so far [1990] is the scarcity or apparent absence of viral DNA in proportion of patients.

The HIV genome has undergone a further resurrection by the development of tests which claim to quantify the HIV in plasma, that is the viral load. There are 3 main tests and they suffer from all the limitations mentioned so far.

For example, in a study by French researchers they cultured 15 HIV-1 strains. Strain zero, which is said to be the most diverse, was not included. They tested samples of cell free fluid containing the same load of HIV as quantified by p24 measurements. (*Journal of Acquired Immune Deficiency Syndromes and Human Retrovirology* 15:174.)

If the tests were true measurement of HIV RNA the results should have been the same for all strains in a given test and all tests for a specific strain.

As can be seen, this is not the case. If this test measured the "viral load" accurately, every number to the right of the first column should be identical.

One wonders how such tests can be used to measure anything, much less to quantify a deadly virus.

According to the HIV theory of AIDS, increase in viral load leads to AIDS, but what has been found is that increase in "viral load" follows does not precede disease. Resolution of disease decreases "viral load".

The current anti-HIV drugs are said to act by preventing fresh cycles of infection, that is by preventing the formation of new proviral DNA in previously uninfected cells, which in turn prevents the formation of new RNA within these cells and the production of new viral particles.

Thus drugs that decrease the viral load, that is, RNA, should first decrease the proviral DNA, that is, the "viral burden".

But what is found is that the viral load decreases to undetectable levels while there is no significant decrease in DNA.

Thus either "HIV RNA" and "HIV DNA" or both are not HIV, or RT and protease inhibitors do not have anti-HIV effects.

They only prohibit the measurement of RNA.

If the former is the case then there can be no HIV genome and thus no anti-HIV drugs.

As the title of Montagnier's paper reveals: HIV and thus HIV RNA, HIV DNA, HIV proteins and HIV antibodies, that is, the existence of HIV, is based upon claims of the isolation of a unique infectious retrovirus.

Such proof does not exist.

Thank you very much for your attention tonight. *

This is the second edited version of the presentation made by Eleni Papadopulos-Eleopulos on behalf of the International Forum for Accessible Science (IFAS) at the XIIth International AIDS Conference Geneva, 28th June 1998.

This session was conducted live by satellite from Perth, Western Australia.Many thanks to Michael Baumgartner for inviting and organising the Perth group's participation.

Revised 28/9/98 and 16/10/98 VFT

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