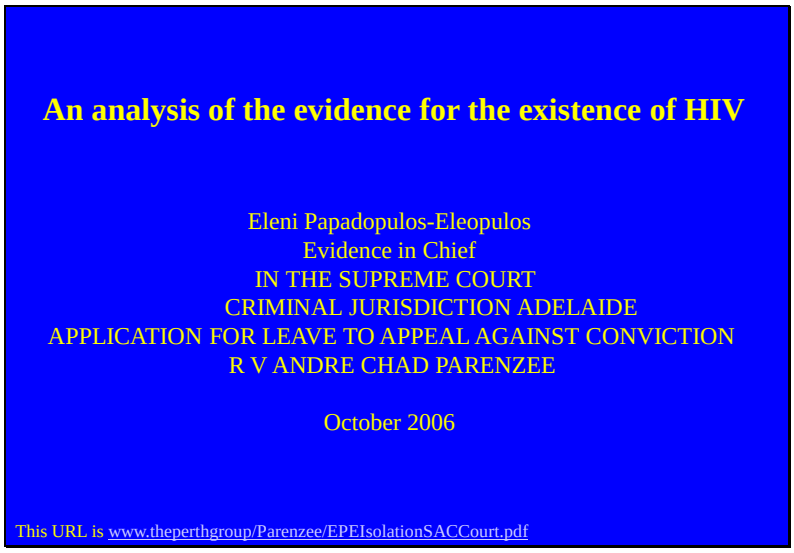
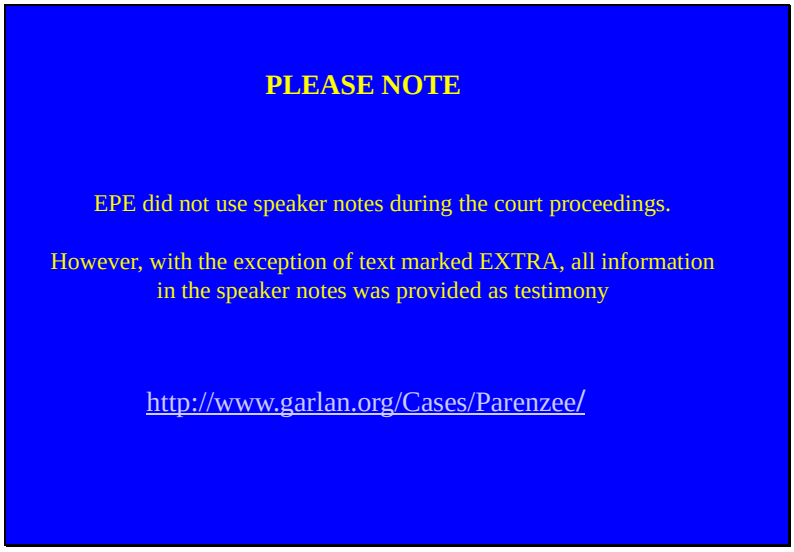
Slide 1 BACK



Slides 1-18 of this presentation cover some background material in regard to cells, viruses, proteins, DNA, RNA and enzymes.

Slide 2



Court transcripts of the prosecution witnesses' testimony can be found at

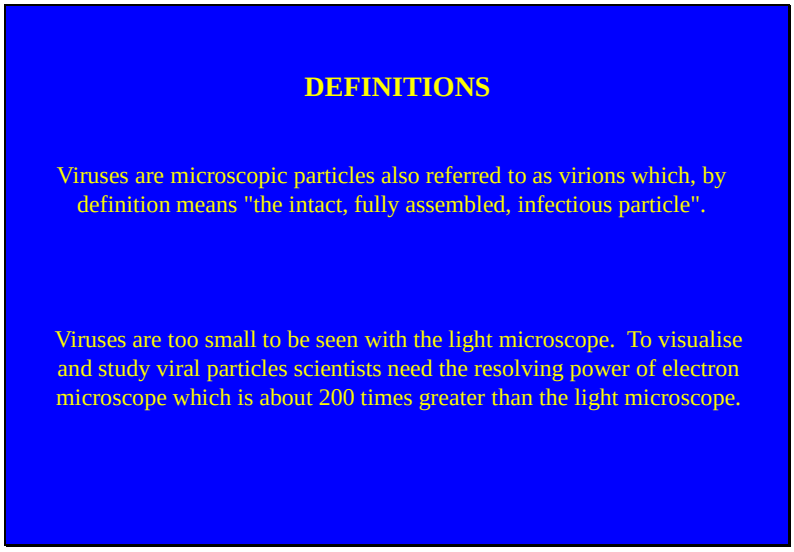
http://www.garlan.org/Cases/Parenzee/

T followed by a number refers to the page of the trial transcript

This presentation can be read in conjunction with our analysis and commentary on Montagnier's scientific research at

www.theperthgroup.com/montagniernobel.html

Slide 3

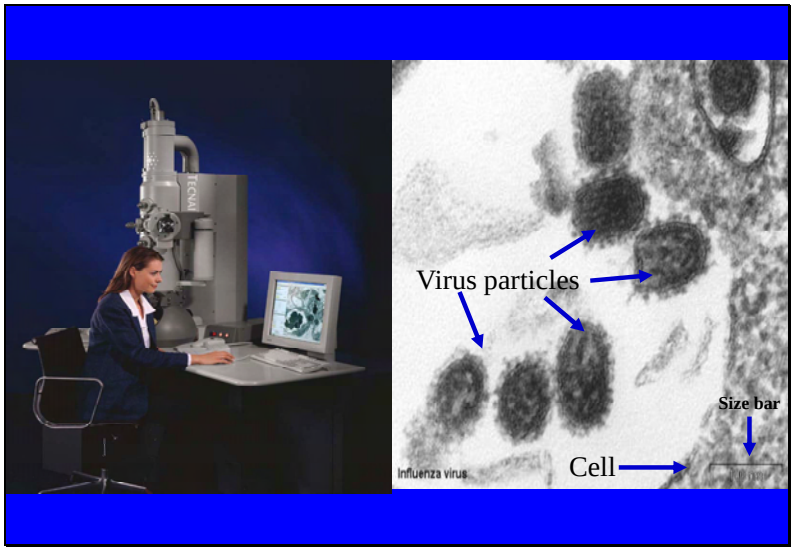


The main property of viruses is their being microscopic particles of particular sizes and shapes and endowed with other defining features, collectively referred to as morphology. Virus particles are too small to be seen with the light microscope. Hence to study their morphological features scientists must use the electron microscope which is capable of enlarging objects 200 times smaller than those that can be seen with the light microscope.

EXTRA
Most virus particles have diameters between 10 and 300 nanometres (nm). The particle claimed to be HIV has a diameter of 100 nanometres. This means that 20,000 could line up across the diameter of the head of a sewing pin (2 mm). Millions could be placed on the head of the pin.
In fact the HIV particle is only 1000 times larger than a hydrogen atom.

For further information on the sizes of biological objects see
http://en.wikibooks.org/wiki/Biology_Cell_biology_Introduction_Cell_size

Slide 4



On the left is an electron microscope and on the right, an example of what it can reveal. In this case six influenza virus particles. Notice the particles lie outside the cell which is many times larger than the particles. This is why in this magnification we see only a small portion of the cell.

Also notice in the bottom right hand corner a thin, dark, horizontal line called the size bar.

The size bar is placed in the electron micrograph so the scientist or technician reporting the EM can determine the dimensions of particles or other structures. The size bar is essential because dimension is one of the key classifying elements in determining what species of viral particle is present.

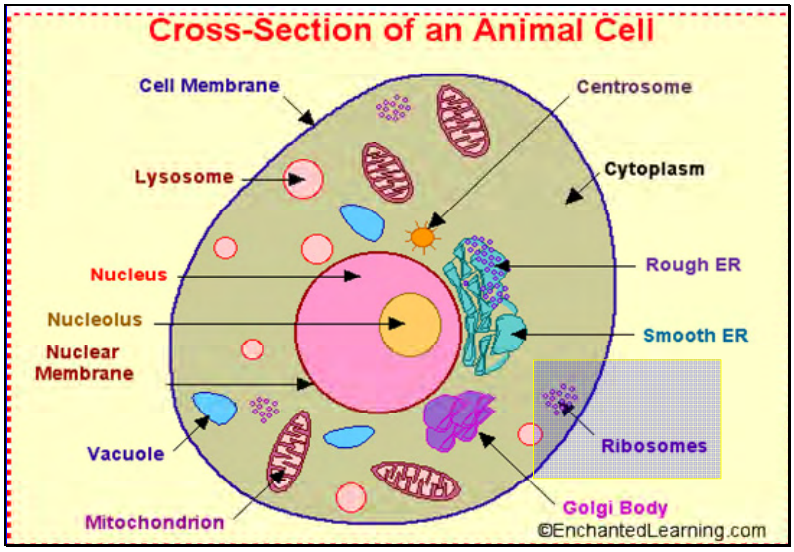
EXTRA
One should note that many images of particles claimed to be HIV are not electron micrographs but artists' renditions done on a computer. Many images of HIV do not have size bars.

For example, the National Institute of Allergy and Infectious Diseases has a website called "Focus on the HIV/AIDS connection".
http://www3.niaid.nih.gov/news/focuson/hiv/resources/default.htm

This website has a link to "Electron micrographs and other images of HIV".
http://www.virology.net/Big_Virology/BVretro.html

Of the 25 images of HIV at this site 17 are artists' renditions and 8 are electron micrographs. None of the images has a size bar. No professional electron microscopist would report on such images in the absence of a size bar or without noting the approximate magnification.

Slide 5



This is a diagram of a cell. Cells are much larger than virus particles and are readily seen with the light microscope.

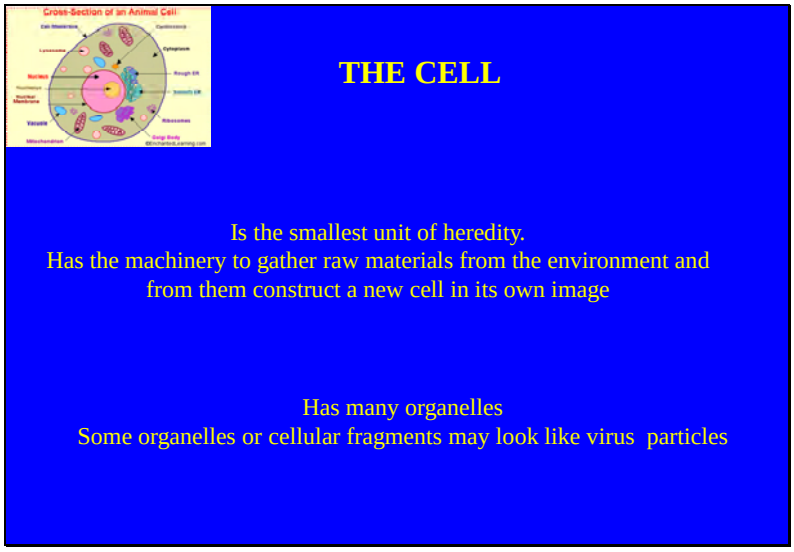
A typical human cell has a diameter between 10-30 micrometres µm.

Cells consist of a nucleus surrounded by a nuclear membrane. The nucleus houses the genetic material of the cell in the form of DNA.

Outside the nucleus lies the cytoplasm, a colloid-like material containing water, electrolytes, proteins and many other biochemical constituents.

Also in the cytoplasm are a range of different structures including those known as organelles ("little organs"). These include lysosomes and mitochondria.

Slide 6



A cell is the smallest unit of heredity.

The cell has the metabolic machinery required to turn raw materials gathered from its environment into an identical copy of itself. That is, replicate. Cells do this by dividing in two (binary fission).

It is important to note that some cellular organelles may on cursory examination of electron micrographs look like virus particles.

The same can be said of cellular fragments which are parts of dead or dying cells, or of cells that have been deliberately disrupted during the processing of a cell culture experiment.

Most significantly, viruses are made principally of proteins and nucleic acids and these are also principal components of cells. In other words, viruses do not have "special" biochemical constituents that do not occur in cells which would otherwise identify their source as a virus*.

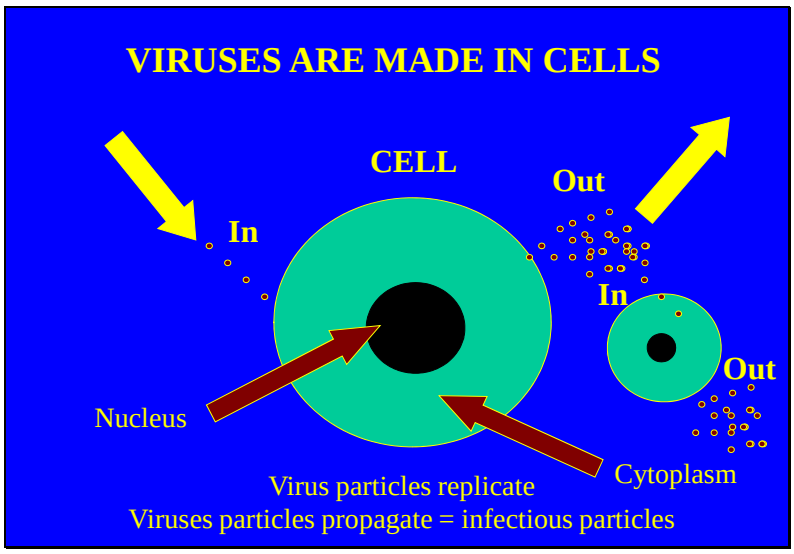
EXTRA

*This is salutary lesson. Over fifty years ago scientists believed that an enzyme called ATPase, which is a protein, was unique to retroviruses. "The ATPase was considered so consistent a component of virions" that is, the retrovirus particles, that it was used to both detect and quantify the number of retrovirus particles [1]. When it was realized that ATP and ATPases were found in all cells and that its presence in the oncovirus [=retrovirus] particles depended "upon cell-specific, not virus-specific factors", the enzyme ceased to be used for the detection and quantification of the oncoviral particles [2].

1. Mommaerts EB, Sharp DG, Eckert EA, Beard D: Virus of avian erythromyeloblastic leukosis. I. Relation of specific plasma particles to the dephosphorylation of adenosine triphosphate. J Nat Cancer Inst 1954, 14:1011-1025

2. Bader JP. Reproduction of RNA Tumor Viruses. In: Fraenkel-Conrat H, Wagne RR, eds. Comprehensive Virology. Vol. 4. New York: Plenum Press, 1975:253-331.

Slide 7



Cells (which include bacteria) replicate provided they have an adequate supply of raw materials. On the other hand, no amount of raw materials can induce a viral particle to replicate because it lacks the requisite metabolic machinery.

Hence to replicate viruses are obliged to parasitise living cells. For a virus to multiply it must invade a suitable host cell and hijack the cell's metabolism. Once inside the cell the viral genetic material (RNA or DNA depending on the type of virus) takes over the cell's metabolism to synthesise the viral proteins and nucleic acids. These are then assembled within the cell following which the complete virus particle exits the cell. In doing so some viruses destroy the cell while others, such as retroviruses, exit the cell membrane in a process referred to as "budding". According to Gallo, this leaves "holes" in the cell membrane which can directly cause death of the cell. One cycle of virus replication cycle is followed by another, and then another, as shown on the right hand of the slide.

It bears repeating that not every particle that looks like a virus is a virus. What makes a particle a virus is its ability to replicate, and in doing so,

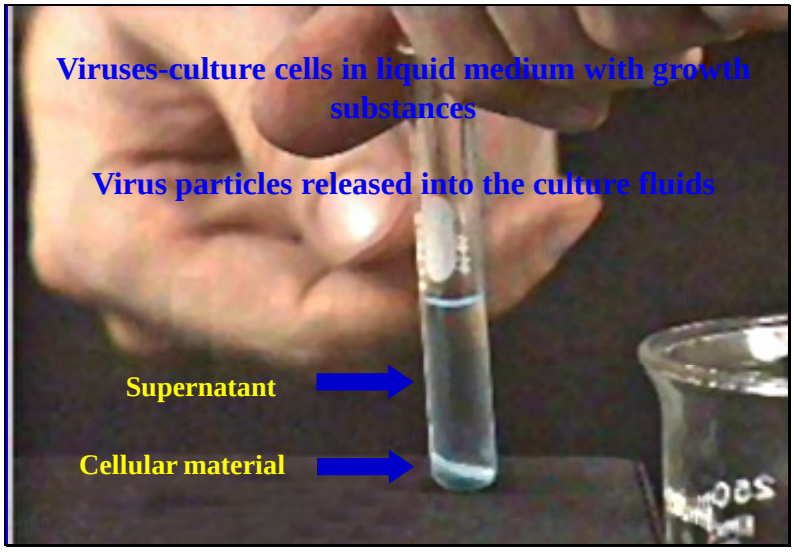
passing from cell to cell, and person to person. Without proof of replication a particle cannot be called a virus and should be referred to as "virus-like".

This can be summed up by the statement that a virus is an infectious particle. And this is how viruses are spread. As HIV expert Simon Wain-Hobson says "…a virus's job is to spread. "If you don't spread you're dead" [1]. You are inanimate, you are not a virus. Even if you look like you are.

From this it is easy to appreciate there is more to proving the existence of a virus than producing an electron micrograph of a virus-like particle. Electron microscopy is an extremely important step, it is a necessary but not sufficient step to prove the existence of a virus.

1. Weiss R. New Strain of AIDS Virus Discovered in Africa. *The Washington Post, 1998:A02.*

Slide 8

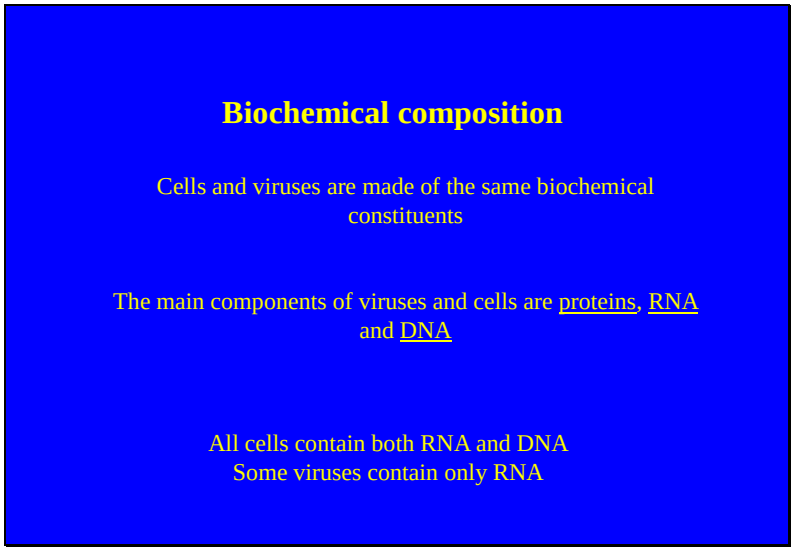


So we can summarise by saying that to obtain viruses you must culture the cells you think are infected with a virus (T-lymphocytes in the case of AIDS), in the presence of various nutrients, growth factors and other chemicals which decades of research have shown are necessary for cells to survive and grow outside the body.

The cultures are tended daily, adding a fresh supply of nutrients and after a period of time, usually 1-3 weeks, if the cells are infected with a virus, the virus particles will be released from the cell into the culture fluids.

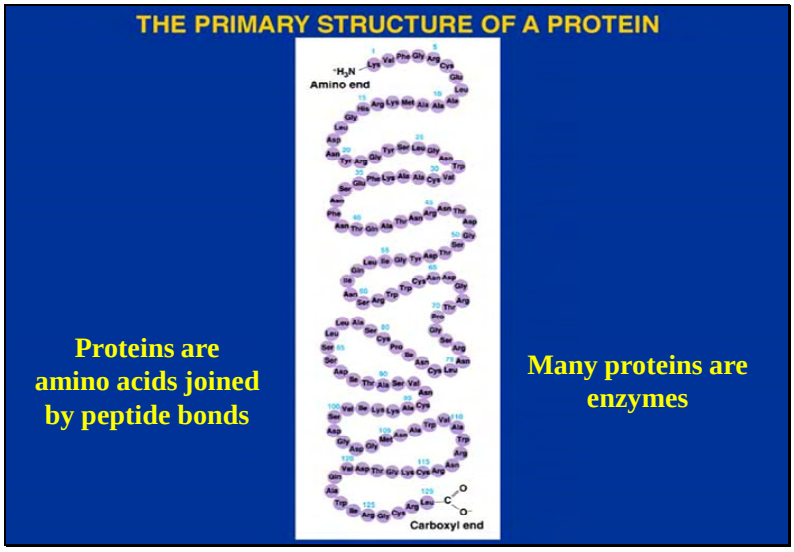
If the test tube is centrifuged for ten or so minutes the larger and heavier cellular material will be forced to the bottom of the tube leaving a clearer, liquid part on top. This is fluid is known as the culture supernatant. This is where viral particles will be found, albeit mixed up with some cellular material and debris from dead and dying cells. Of course if the cells are not infected with a retrovirus, that is, if the hypothesis is wrong, there will be no retroviral particles in the supernatant.

Slide 9



Cells and viruses are made of a several different biochemical molecules which include proteins and the nucleic acids RNA and DNA.

Slide 10

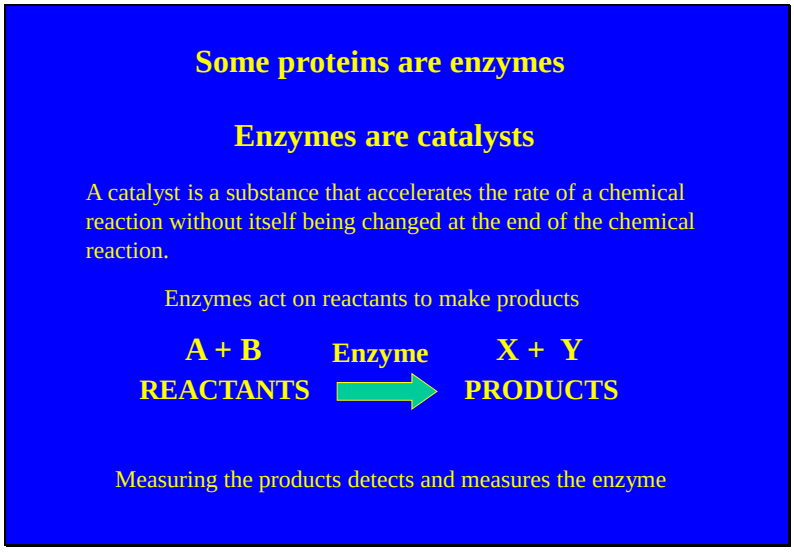


Proteins are polymers made up of 20 different chemical building blocks known as amino acids. Each circle with a three letter annotation is one particular amino acid. For example, Lys is the amino acid lysine.

Within the protein molecule the individual amino acids are joined together by chemical bonds known as peptide bonds. The number of amino acids in a protein is typically a hundred to thousands.

Proteins serve many functions. Some, such as actin and myosin, are structural components of cells, bacteria and viruses. Some such as insulin and oxytocin are hormones. Antibodies are all proteins. Many proteins are enzymes.

Slide 11



An enzyme is a biological catalyst and a cell's complement of enzymes undertakes the myriad of chemical reactions essential for cellular metabolism and life. It is from these reactions that the cell extracts energy from its foodstuffs without which the cell will cease to function and ultimately die.

A catalyst is a substance that increases the rate at which a chemical reaction occurs. That is, the rate at which reactants such as A and B unite to form a different substance or substances such as the products X and Y. However, at the completion of the reaction, the catalyst is chemically unchanged and is present in the original amount.

Within limits the more enzyme there is available the greater the amount of products produced. If there is only a tiny amount of enzyme there will not be enough to go round. With more enzyme the amount of products will increase.

In theory, to measure the quantity of a particular enzyme, or indeed to determine if there is any enzyme present, the enzyme is extracted from the cell or the reaction mixture and its amount determined. That is, the protein

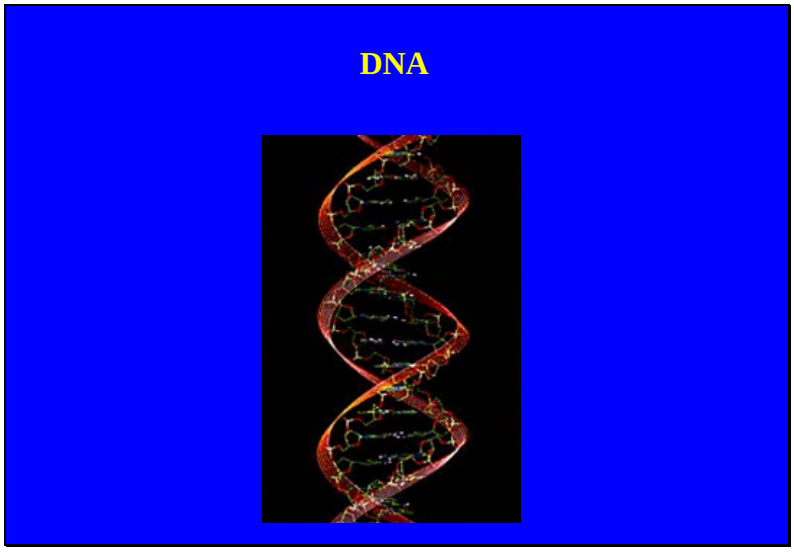
that is the enzyme is purified and weighed. However, the much easier and usual way is to infer the presence and amount of the enzyme present by measuring the amount of products it produces. This is sometimes reported as the enzyme "activity" but it must be remembered that this is an indirect way of measuring the amount of enzyme present. It goes without saying this method will produce unambiguous results if and only if that enzyme is the only enzyme present that catalyses a particular reaction.

EXTRA

It has long been taught that one enzyme catalyses one particular chemical reaction but no others. However, scientists are increasingly appreciating that the same enzyme may catalyse more than one reaction [1].

1. James LC, Tawfik DS: Catalytic and binding poly-reactivities shared by two unrelated proteins: The potential role of promiscuity in enzyme evolution. *Protein Science* 2001, 10:2600-2607.

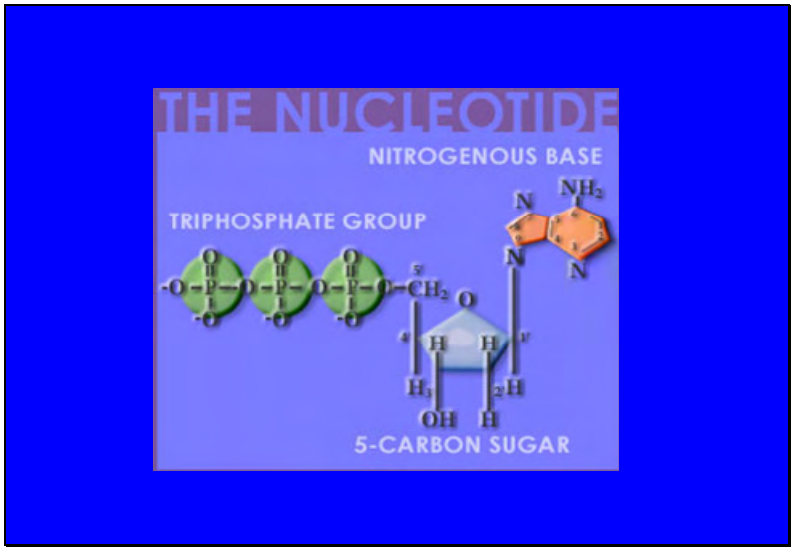
Slide 12



Deoxyribonucleic acid or DNA is one of the two nucleic acids found in cells. It is a large molecule consisting of two intertwined chains that carry the genetic information required to produce all the cell's proteins.

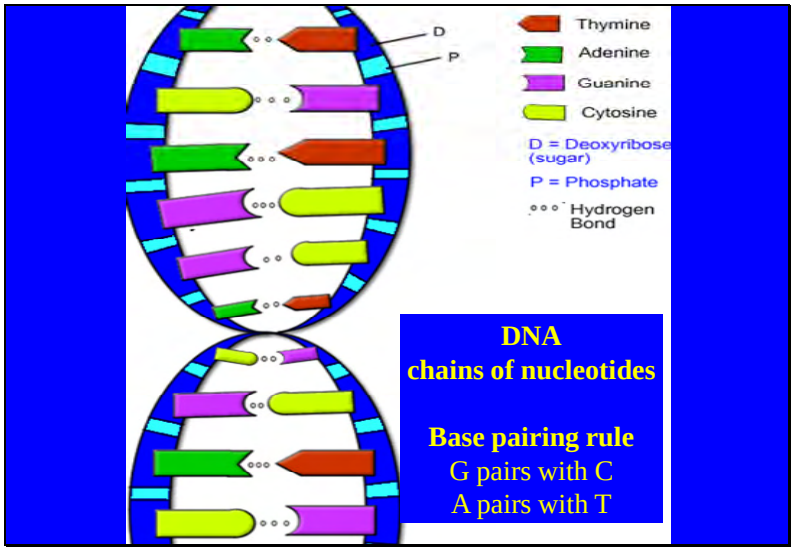
DNA is made of four chemical building blocks called nucleotides.

Slide 13



A nucleotide consists of a nitrogenous base, a 5 carbon sugar (deoxyribose or ribose) and three phosphate groups.

Slide 14



The two nucleotide chains are twisted around each other in an arrangement resembling a spiral staircase with two handrails. Each hand rail consists of a continuous chain of alternating sugar (deoxyribose) and phosphate groups. The half steps (imagine each step cut in half and then glued back together) are made of four nitrogenous bases called guanine, cytosine, adenine and thymine (G, C,A,T). One part of the each base is attached to the sugar/phosphate chain and another part is linked to a base on the opposite chain by chemical bonds known as hydrogen bonds (imagine these bonds are the glue joining the cut, half steps together, the '0's in the picture between opposite bases). The base pairing is such that G on one chain always pairs C on the other and A always pairs with T. This is known as the base pairing rule.

Because hydrogen bonds are relatively weak the two chains that make up DNA can be prized apart. For example, if the DNA molecule is heated. This property is used in the technique known as the polymerase chain reaction (PCR), which is frequently used by HIV/AIDS experts to detect and quantify DNA they claim is that of "HIV".

Also, because of the base pairing rules, if the sequence of one chain is known, the other is automatically determined. For example, if one chain is GCAT the other is CGTA.

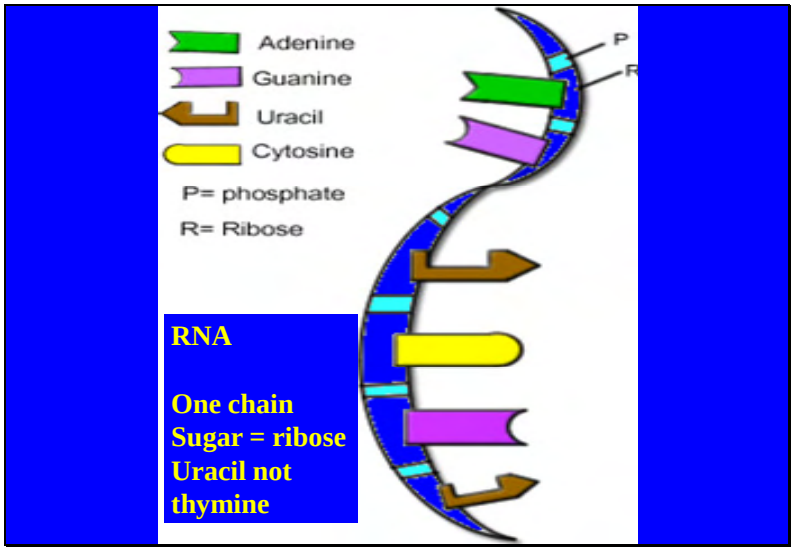
DNA chains are typically thousands of nucleotides long.

EXTRA
Many biologists, scientists and doctors think of particular DNA sequences as "genes". However, defining a gene appears to be an impossible task.

In 1982 EPE corresponded with Sir Gustav Nossal in regard to her theory of antibody diversity. There she wrote "the dogma 'one gene/one protein chain' is fundamentally flawed". Twenty four years later one reads in *Nature* "In classical genetics, a gene was an abstract concept – a unit of inheritance that ferried a characteristic from parent to child. As biochemistry came into its own, those characteristics were associated with enzymes or proteins, one for each gene. And with the advent of molecular biology, genes became real, physical things – sequences of DNA which when converted into strands of so-called messenger RNA could be used as the basis for building their associated protein piece by piece…This picture is still the working model for many scientists [including the "HIV experts and dissidents]. But those at the forefront of genetic research see it as increasingly old-fashioned – a crude approximation that, at best, hides fascinating new complexities and, at worst, blinds its users to useful new paths of enquiry...Without a clear definition of a gene, life is also difficult for bioinformaticians who want to use computer programs to spot landmark sequences in DNA that signal where one gene ends and the next begins. But reaching a consensus over the definition is virtually impossible, as Karen Eilbeck can attest. Eilbeck, who works at the University of California in Berkeley, is a coordinator of the Sequence Ontology consortium. This defines labels for landmarks within genetic-sequence databases of organisms, such as the mouse and fly, so that the databases can be more easily compared...Eilbeck says that it took 25 scientists the better part of two days to reach a definition of a gene that they could all work with. "We had several meetings that went on for hours and everyone screamed at each other," she says. The group finally settled on a loose definition that could accommodate everyone's demands".

Pearson H: Genetics: what is a gene? *Nature* 2006, 441:398-401.

Slide 15

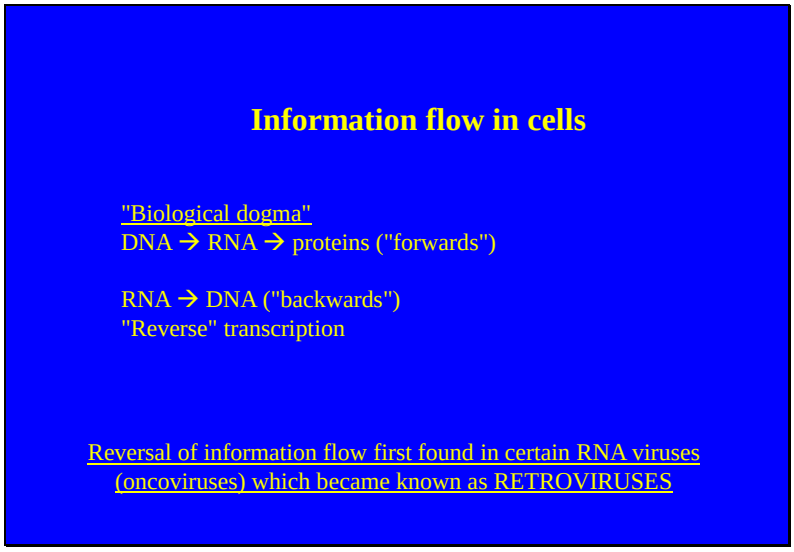


RNA is the other nucleic acid found in cells and in some but not all viruses.

RNA has much the same composition as DNA but with three significant differences.

- RNA is a single chain
- Its sugar is ribose rather than deoxyribose
- In RNA the base uracil takes the place of thymine that is contained in DNA.

Slide 16

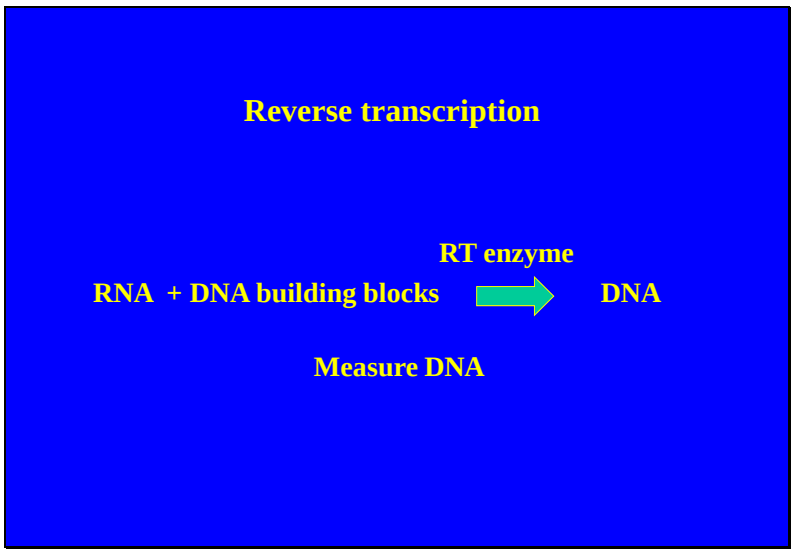


Soon after DNA was discovered in 1953, a theory was put forward that the information in cells flows exclusively in one direction. That is, from DNA to RNA and from RNA to proteins. This unidirectional flow is known as the 'biological dogma'. However, since 1970 it has been realised the dogma is wrong. Information may also flow at least partly in the opposite direction. That is, from RNA to DNA. In other words if DNA to RNA is considered "forwards" then RNA to DNA is a "backwards" or "reverse" information flow.

Since the process of producing RNA from DNA is known as transcription, the process of producing DNA from RNA is called reverse transcription and, like the majority of chemical reactions that occur in cells, reverse transcription requires an enzyme.

Enzymes able to perform this task were first discovered viruses which, until then, were known as an oncoviruses because oncoviruses were believed to cause tumours (cancers). (oncus=Greek for tumour). Nowadays oncoviruses are called retroviruses and the enzymes are known as reverse transcriptases.

Slide 17

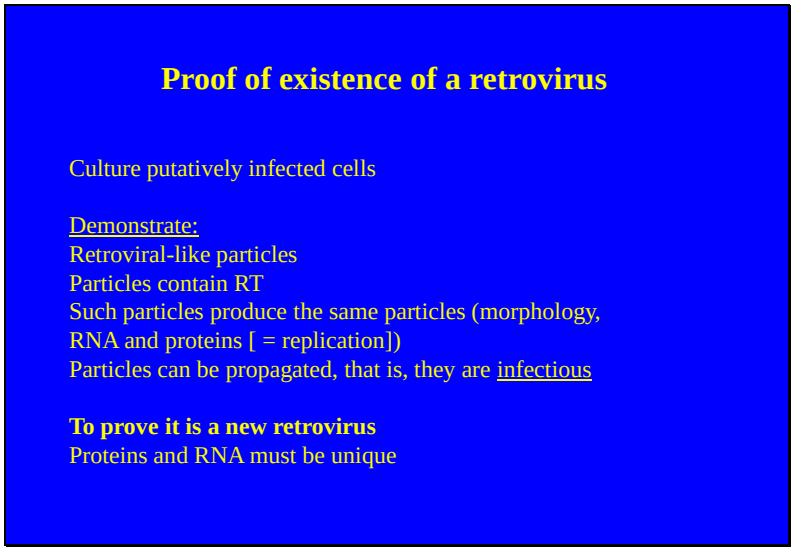


To prove that a cell culture contains an enzyme capable of undertaking reverse transcription, a scientist introduces a piece of RNA called the template (the piece of RNA to reverse transcribe) and the four DNA nucleotides from which DNA is made (G, C, A and T). At one end of the RNA template is attached a small piece of DNA which is called the primer. (Without the addition of this primer the reaction will not start). The entire RNA-DNA molecule is referred to as the "template-primer".

Sometime later the culture is tested for the presence of DNA. If DNA matching the temple-primer RNA is found this infers the presence of a reverse transcribing enzyme whose purpose it is to catalyse this chemical reaction.

The amount of DNA found is a measurement of the amount of enzyme present and is referred to as the reverse transcriptase "enzyme activity" (RT activity).

Slide 18

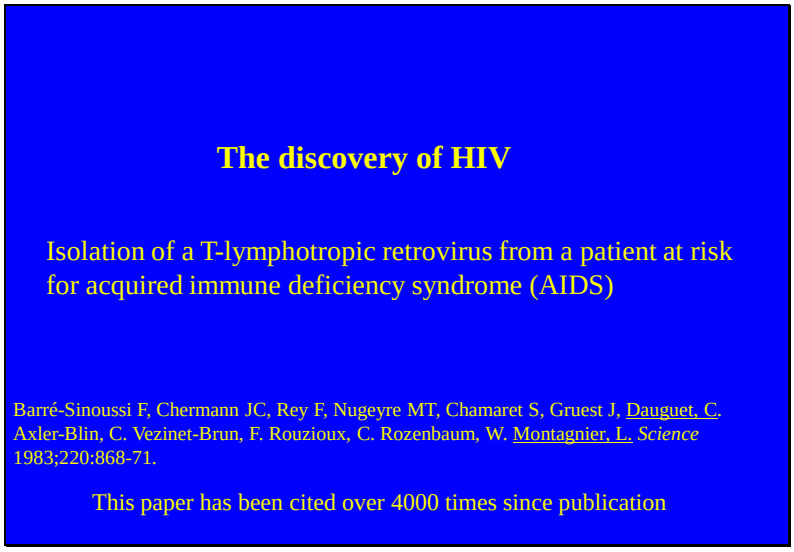


Given the definition of a retrovirus, that is, an infectious particle of particular morphology containing the enzyme reverse transcriptase, here is the experiment a scientist must perform in order to prove the existence of a new retrovirus. (Virologists may use the term "novel" in place of "new". It means the same thing).

1. Culture the cells in which it is hypothesised a retrovirus infection is present.
2. Demonstrate that after several days or weeks the culture supernatant (into which putative viral particles are released) actually contain such particles (particles bearing the correct morphology of retroviruses).
3. Verify that the particles contain a protein that causes reverse transcription.
4. Prove the particles are infectious, that is, the particles can be transmitted. To prove this the particles are introduced into an uninfected cell culture whereupon particles appear which have the same morphology and the same proteins and RNA as the original particles.
5. Prove the retrovirus is new by showing that the particles consist of proteins and RNA which are unique to them, that is, they are not found in any other retrovirus particles.

Bearing this in mind we can now ask what is the evidence for the existence of HIV.

Slide 19



Everyone in the scientific community accepts that Professor Luc Montagnier from the Pasteur Institute and 11 colleagues, including Francoise Barre-Sinoussi and Jean Claude Chermann, are the scientists who proved the existence of HIV. According to the scientific community this proof appeared in 1983 in a paper entitled "Isolation of a T-lymphotropic retrovirus from a patient at risk for acquired immune deficiency syndrome (AIDS)". In this paper and subsequently this patient is referred to as BRU.

Everybody cites this paper as the first where the existence of AIDS has first been reported. Since 1983 it has been cited in over 4,000 publications.

EXTRA
During cross-examination by Mr. Kevin Borick QC, Prosecution witness Professor Elizabeth Dax was asked about a citation of the Montagnier paper she made in one of her own publications [reference 6 in 1 below]. She first read the relevant sentence from her paper:

A…Anti-HIV immunoassays…were developed and first implemented in HIV soon after the discovery of HIV-1 as the aetiological factor for the development of acquired immunodeficiency syndrome AIDS, yes.

Q. Would you agree that you cited Montagnier's 1983 paper in support of that claim.
A. If that's what reference No. 6 says, yes. Yes, that's the Science paper in 1983, yes.
Q. What evidence in the Montagnier paper, which you have cited, convinced you that Montagnier had proved that HIV is the cause of AIDS.
A. Well, this is the accepted reference, this is the accepted seminal reference on the isolation of HIV and it refers to the isolation of a virus, 'the virus' that is associated with people who had acquired immunodeficiency disease. So I think that's an accepted reference of the basic isolation. I could have cited many others but when you produce a scientific paper you try and go back to the fundamental reference and this is the accepted fundamental reference". T898

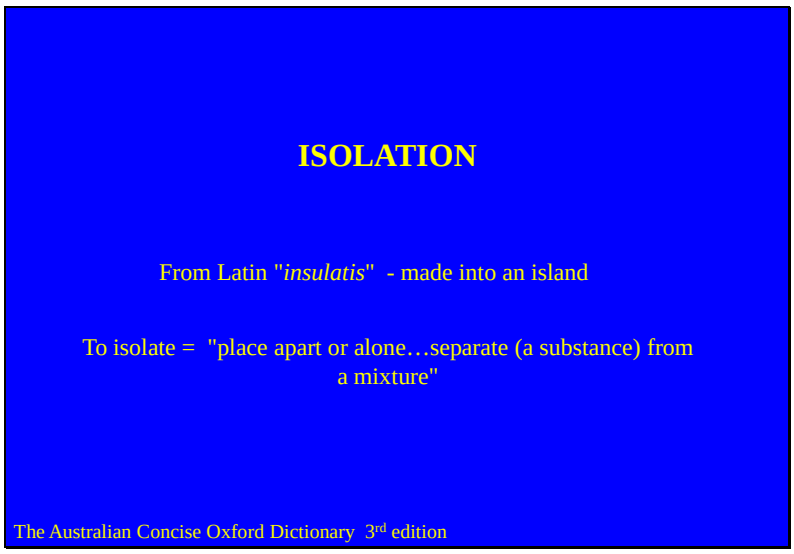
Hence if HIV exists this "fundamental" paper should leave the reader in no doubt this is a fact.

Note Professor Dax did not answer Mr. Borick's question.

Also note that neither Montagnier in 1983 nor the Nobel Committee in 2008 claimed that in 1983 Montagnier proved HIV is the "aetiological factor for the development of acquired immunodeficiency syndrome AIDS".

1. Dax EM, Arnott A. Advances in laboratory testing for HIV. *Pathology* 2004; 36:551-60.

Slide 20



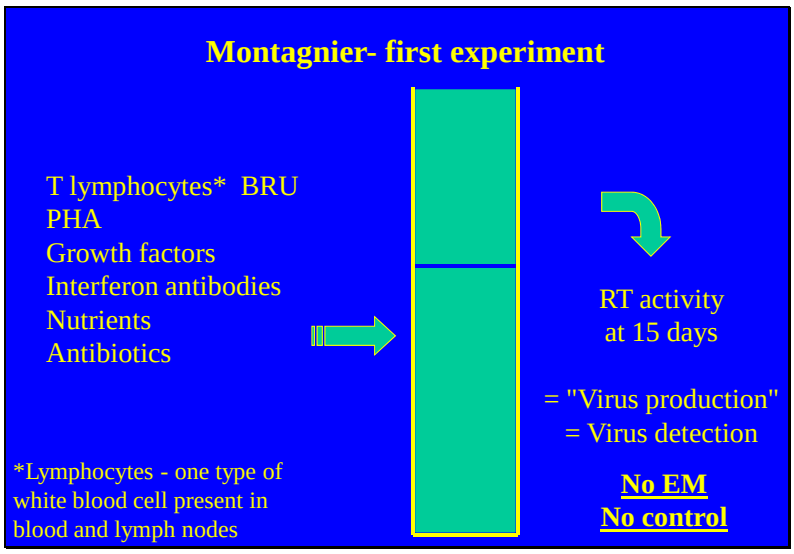
Montagnier and his colleagues claimed to have proven the existence of HIV by isolating it. In fact "Isolation" is the first word in the title of Montagnier's paper.

According to the dictionary, "isolation" comes from the Latin word "*insulatis*" meaning "made into an island". Isolation means to place apart or alone, or to separate a substance in a mixture from everything else in that mixture.

If Montagnier and his colleagues had isolated a retrovirus-like particle which they proved infectious and hence a virus, and it was a new virus, then the claim for the existence of HIV can be justified.

However, this is not what Professor Montagnier and his colleagues meant by isolation and it certainly is not what their experiment achieved.

Slide 21



Montagnier and his team performed three main experiments and we will examine each in some detail.

In the first experiment Montagnier extracted lymphocytes from the lymph nodes of BRU and put them into culture with a number of chemical agents. One of the chemicals was a substance called phytohaemagglutinin (PHA), a protein extracted from kidney beans which has the property of inducing cell division (mitosis), also referred to as "stimulation". In other words, PHA is a mitogenic agent.

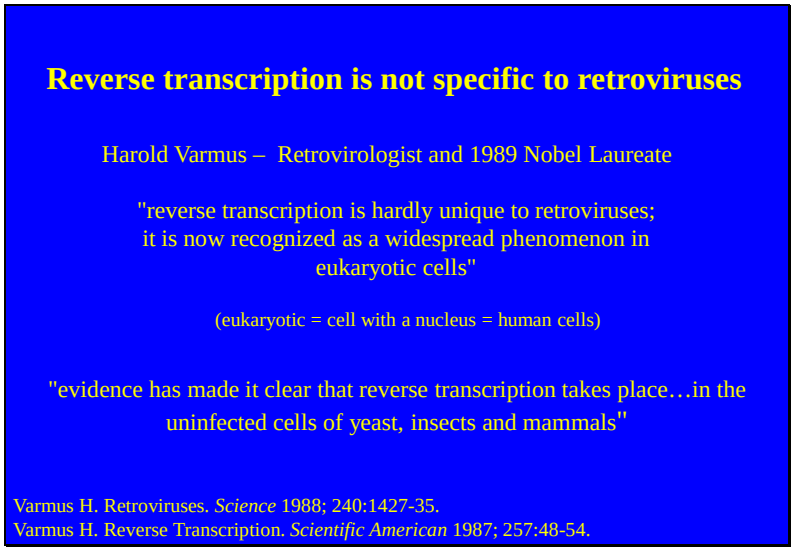
After 15 days of culture Montagnier could detect reverse transcription in the culture supernatant. From this he concluded he had demonstrated "Virus production" and "Virus detection".

As said earlier, the main characteristic of viruses is being particles which can be visualised using the electron microscope. But Montagnier did not publish any pictures of what, if anything, he had in the way of particles in the first experiment. There were no EM of the culture supernatant or even the culture. The claim of virus production and detection was based on nothing more than the detection of a reverse transcriptase activity.

Now, it is not difficult to appreciate that the detection of a reverse transcriptase activity is not isolation of anything, let alone a virus. For example, when a doctor orders a blood test on a patient with chest pain, he is looking for evidence of enzymes that leak out of damaged heart muscle. And if such enzymes are present no one would consider calling this "isolation of the heart".

However, as distinct to isolation, reverse transcriptase activity could be proof of detection of a retrovirus but if and only if reverse transcriptase activity is not only a property of retrovirus particles but is also a unique property of retroviruses. This is not the case.

Slide 22



Reverse transcription is not specific to retroviruses.

Some of the best known retrovirologists such Harold Varmus affirm this fact.

Varmus is a biologist who received a Nobel Prize for research into cancer and viruses, in fact on oncogenes.

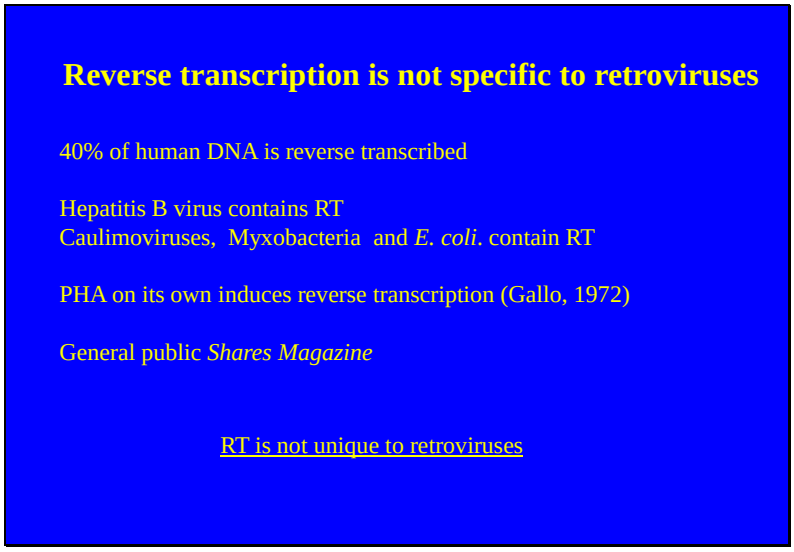
Varmus says "Although reverse transcription was first encountered in the retrovirus life cycle, it is hardly unique to retroviruses; it is now recognized as a widespread phenomenon in eukaryotic cells and viruses. Indeed, as much as 10% of the eukaryotic genome may be composed of products of reverse transcription".

and

"…evidence has made it clear that reverse transcription takes place…in the uninfected cells of yeast, insects and mammals"

So, according to Varmus, reverse transcription is not specific to retroviruses. Hence it follows that Montagnier was wrong to claim he had "Virus production" and "Virus detection" merely because in his culture he detected reverse transcriptase activity.

Slide 23



Scientists who study the origin of life claim that what came first was RNA, then DNA. And this DNA was made by reverse transcribing enzymes using RNA as a template. In fact, today many molecular biologists consider about 40% of our DNA originated at the behest of reverse transcription of RNA.

It is a known that bacteria and viruses other than retroviruses possess reverse transcription ability. These include hepatitis B virus which infects a very high percentage of gay men to which intravenous drug users are also prone. Bacteria too can reverse transcribe. Including the bacterium *E. coli* which is present in vast amounts in the human gastro-intestinal tract and is an extremely common human pathogen.

As far back as 1972 Gallo, who is the second researcher considered to have proven the existence of HIV, showed that normal, non-infected lymphocytes, cultured with PHA, reverse transcribe when using the same template-primers used in "HIV" research.

The non-specificity of reverse transcription has even appeared in the popular press. In 2001 the Australian magazine S*hares* published an

article about investing in biotechnology stocks which pointed out that reverse transcription is not specific to retroviruses [1].

So, we can conclude that reverse transcriptase activity is not specific to retroviruses and Montagnier had no basis to claim a retrovirus was being produced and detected in BRU's cell culture.

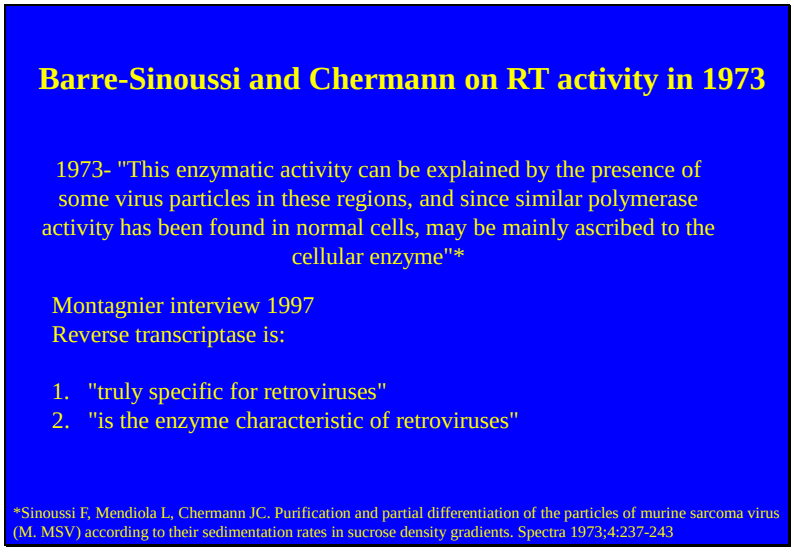
1. Pachacz M. No need to be phased. *Shares*, 2001:28-32. http://theperthgroup.com/POPPAPERS/SharesMagazine2001.pdf

For a more detailed discussion on reverse transcriptase see

Papadopulos-Eleopulos E, Turner VF, Papadimitriou JM. Is a positive Western blot proof of HIV infection? *Biotechnology* 1993;11:696-707.
http://www.theperthgroup.com/SCIPAPERS/biotek8.html

Papadopulos E, Turner V, Papadimitriou J, Page B, Causer D. A CRITICAL ANALYSIS OF MONTAGNIER'S 1983 "SEMINAL" PAPER "Isolation of a T-lymphotropic retrovirus from a patient at risk for acquired immune deficiency syndrome (AIDS)". *Science* 1983; 220:868-71.
www.theperthgroup.com/Nobel/Montagnier1983Paper.pdf

Slide 24



Barre-Sinoussi and Jean Claude Chermann are the first and second authors respectively of Montagnier's 1983 paper. However, a decade earlier, in 1972, these researchers organised a meeting on retroviruses held at the Pasteur Institute and the proceedings were published in a journal called *Spectra* [1].

Reporting on reverse transcriptase activity in an experiment involving the purification of murine (mouse) sarcoma virus, a retrovirus, they wrote "This enzymatic activity can be explained by the presence of some virus particles in these regions, and since similar polymerase activity has been found in normal cells, may be mainly ascribed to the cellular enzyme". ("regions" is a reference to fluids in a test-tube where retrovirus particles are not present but RT activity is found, see below and slide 64). What did they mean by this?

Polymerase is a general term for an enzyme that catalyses the formation of polymers, such as DNA or RNA. Since the reverse transcriptase makes DNA from RNA this enzyme falls into the general class of polymerases. What Barre-Sinoussi and Chermann's evidence proved is that a "similar polymerase…has been found in normal cells", that is, cells not infected with

a retrovirus. Hence in 1972, two authors of the Montagnier paper knew that retroviral RT is not the only enzyme capable of acting on an RNA template-primer to produce DNA. Hence not only was it wrong to conclude there was "Virus production" and "Virus detection", at least two of the authors of the Montagnier paper had proved it wrong.

In July 1997 Montagnier gave an interview to the French investigative journalist Djamel Tahi [2]. In response to the first question Montagnier said that reverse transcriptase is "truly specific for retroviruses". However, eight questions later Montagnier conceded that reverse transcriptase is only characteristic of retroviruses.

There is an enormous difference between being characteristic and being specific.

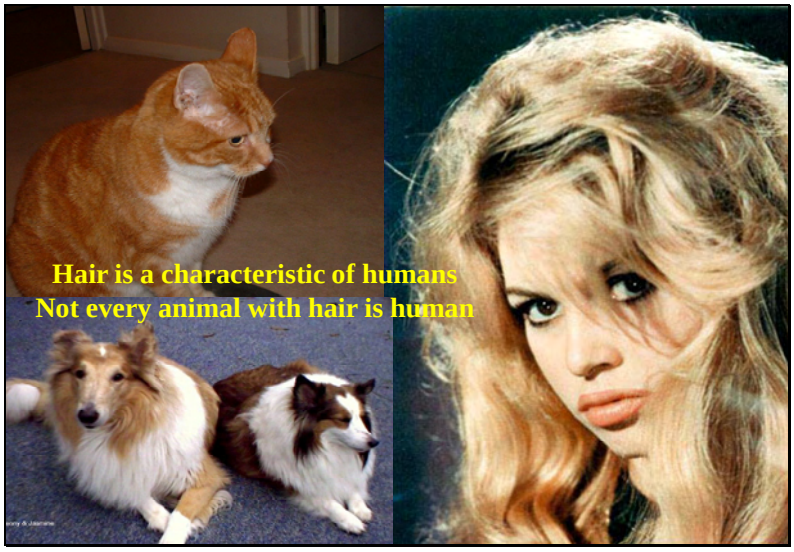
1. Sinoussi F, Mendiola L, Chermann JC. Purification and partial differentiation of the particles of murine sarcoma virus (M. MSV) according to their sedimentation rates in sucrose density gradients. *Spectra* 1973;4:237-243. http://theperthgroup.com/OTHER/Spectra.html

2. Tahi D. Did Luc Montagnier discover HIV? Text of video interview with Professor Luc Montagnier at the Pasteur Institute July 18th 1997. *Continuum* 1998;5:30-34. http://www.virusmyth.net/aids/data/dtinterviewlm.htm

EXTRA

The interview was recorded in July 1997 *en camera* and is the property of DJ Tahi dtahi@terraincognita.fr

Slide 25



For example, hair is a characteristic of human beings but hair is not specific because there are many other animals which have also have hair. Finding a hair in your soup doesn't tell you what kind of animal was sampling it.

EXTRA
During cross examination Professor Gallo was questioned by Kevin Borick QC regarding the specificity of reverse transcription.

Q. What did he [Montagnier] describe to you that convinced you this was a retrovirus, and apparently a unique retrovirus.
A... Montagnier had reverse transcriptase in a particle that has structural morphology, comparable, in my eyes, with a retrovirus... (T1312).

Further on:

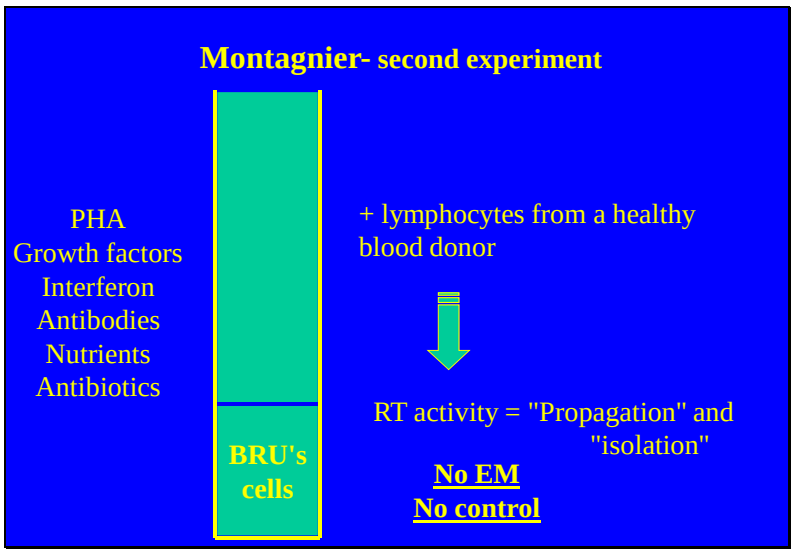
Q. ...Baltimore says that reverse transcriptase activity is not specific to retroviruses and he goes on--this is what I want to put to you--he, as I understand it, says about 50% of our DNA is obtained by reverse transcription of our RNA.

A. ...Reverse transcription is a process by which RNA is converted to DNA. Yes, it goes on. It is not released as a particle, that is point 1; point 2, the enzyme that does it, the enzymatic activity is quite distinct that only a fool would mistake for retrovirus reverse transcriptase (T1313).

Reverse transcriptases are proteins. Neither Montagnier in 1983, nor Gallo in 1984, or anybody else since, has proven the existence of a unique protein in the cell culture or in the material in the 1.16g/ml band [the "purified virus, see below] , much less in a "particle". In all research the evidence for the existence of the "HIV" reverse transcriptase is indirect. That is, by detecting reverse transcription of a synthetic RNA template, the same template which researchers including Gallo showed to be reverse transcribed by normal, non-retrovirus, infected cells including spermatazoa [1] and PHA stimulated lymphocytes [2,3,4].

1. Whitkin, S.S., Higgins, P.J. and Bendich, A. 1978. Inhibition of reverse transcriptase and human sperm DNA polymerase by anti‑sperm antibodies. *Clinical and Experimental Immunology* 33:244‑251.
2. Gallo, R.C., Sarin, P.S. and Wu, A.M. 1973. On the nature of the Nucleic Acids and RNA Dependent DNA Polymerase from RNA Tumor Viruses and Human Cells, p.13‑34. In: Possible Episomes in Eukaryotes. L.G. Silvestri (Ed.). North‑Holland Publishing Company, Amsterdam.
3. Sarngadharan MG, Allaudeen HS, Gallo RC. Reverse transcriptase of RNA tumor viruses and animal cells. *Methods in cancer research, 1976:3-47.*3.
4. Tomley, F.M., Armstrong, S.J., Mahy, B.W.J. and Owen, L.N. 1983. Reverse transcriptase activity and particles of retroviral density in cultured canine lymphosarcoma supernatants. *British Journal of Cancer* 47:277‑284.

Slide 26



In the second experiment Montagnier took some of BRU's cells and added lymphocytes obtained from a healthy blood donor. Again were added all the other substances shown on the slide including growth factors and PHA. Hence on this second occasion Montagnier had a co-culture, which is the term used when cells from two different sources are cultured together. Several days later reverse transcriptase activity was again detected in the culture. This "second appearance" of RT activity was now interpreted as proof of propagation and isolation of a retrovirus. Montagnier named this virus LAV, lymphadenopathy associated virus, which is now called "HIV". Still Montagnier did not publish any electron micrographs to prove particles were present in his cultures.

EXTRA
However, as has been pointed out already, detection of reverse transcription is not isolation of anything, including the enzyme which catalysed this process, let alone a virus. Since reverse transcription is non-specific to retroviruses and, given the right conditions, can be detected in non-infected cultures, finding evidence of reverse transcription, even in a thousand consecutive cultures, cannot be considered proof for transmission (propagation) of anything.

Even if reverse transcriptase activity was specific to retroviruses, since the co-culture contained not only lymphocytes from the healthy blood donor but also from BRU's lymph nodes, the detection of RT activity cannot be considered proof of viral transmission. The activity may have been solely due to the BRU cells.

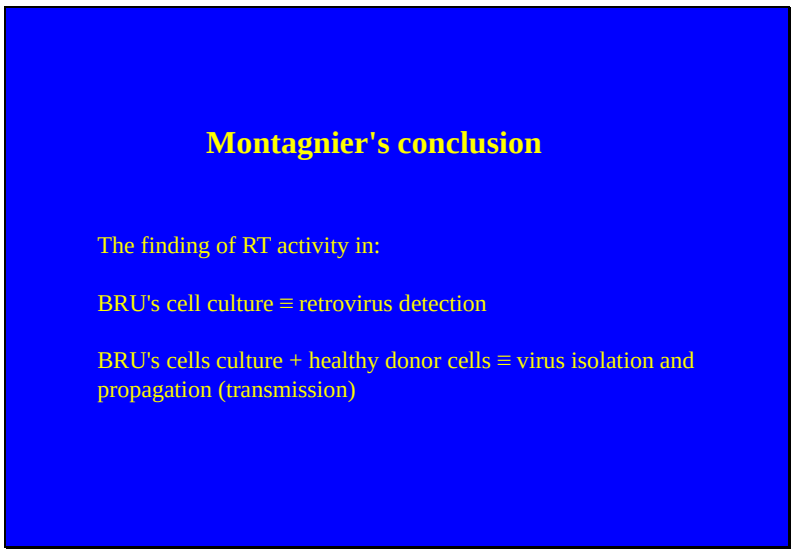
During the Parenzee hearing the following exchange took place between Kevin Borick and Gallo:

Q. In a paper published by you and others in 1976, 'Some evidence for infectious type C virus in humans', you said virus like-particles morphologically and biochemically resembling type C virus, but apparently lacking the ability to replicate, have been frequently observed.
A. By us? No. That's not true. You can't show me a paper I published in '76 that they aren't able to replicate-- found frequently in humans did I ever published, sorry. You are misunderstanding or misreading something.
Q. It is a paper published by you and someone called Wong-Staal and others, 'Some evidence for infectious type C virus in humans', Baltimore, D. Huang, AS, Fox CF, Animal Virology, New York, Academic Press, 1976 [1]..
A. You mean particles, not viruses. You said viruses before. Particles, yes you can find particles not released from the cells in some cases of human leukaemias that we were looking at. Those are likely to be endogenous retrovirus elements not forming full virus and certainly not transmitted in culture and certainly not infectious. ["Virus" is the title of the paper and type C particles lie outside the cell, that is, they are "released from the cells"].
Q. Does all this mean particles with the morphology of retroviruses which are reverse transcriptase activity are not necessarily retroviruses because they do not replicate.
A. Absolutely. If you have a virus that didn't replicate you couldn't call a virus, unless you transmitted it. Montagnier did succeed in his paper in transmitting it. Obviously he did. (T1308).

In the first and second experiment Montagnier did not have even evidence for particles let alone particles which were transmitted. Yet he interpreted his evidence as proving both virus isolation, production and propagation. And this was his claim despite his not publishing any electron micrographs of Part I and Part II of his experiments to show the existence of particles of any kind existed in his cultures.

1. Gallo RC, Wong-Staal F, Reitz M, Gallagher RE, Miller N, Gillespie DH. Some evidence for infectious type-C virus in humans. In: Balimore D, Huang AS, Fox CF, editors. *Animal Virology.* New York: Academic Press Inc., 1976:385-405.

Slide 27



Hence, based on two experiments showing nothing more than reverse transcriptase activity that is not a unique property of retroviruses, and without any electron microscopic evidence for the existence of particles of any kind, virus-like or not, from which this enzyme activity might have originated, Montagnier claimed that BRU was infected with a retrovirus which was transmitted to non-infected lymphocytes.

EXTRA
It is necessary to explain the phrase "No control" which appears in the slides summarising Montagnier's first and second experiments.

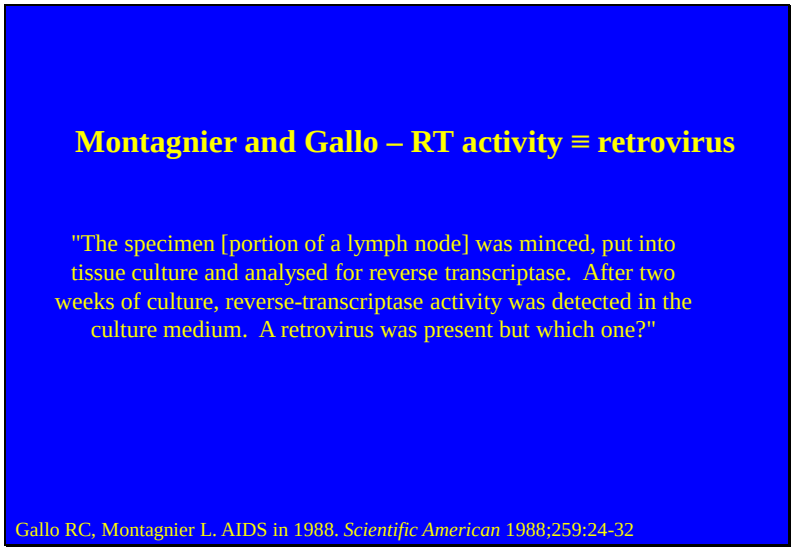
A control is an "Essential part of a scientifically valid experiment, designed to show that the factor being tested is actually responsible for the effect observed. In the control experiment all factors, apart from the one under test, are exactly the same as in the test experiments, and all the same measurements are carried out".
http://www.tiscali.co.uk/reference/encyclopaedia/hutchinson/m0024903.html

Hence a control culture is a culture run in parallel with the test culture and one that is treated in exactly the same manner as the test culture. Montagnier's first experiment should have included a culture which contained cells obtained from a patient similar to BRU but thought not to be infected with a retrovirus. In the second experiment the co-culture should have consisted of cells from similar sick individuals and not just healthy blood donors. The reason for controls is to make sure that the end result of the experiment, in this case RT activity, is not the result of unforseen factors which have nothing to do with a retrovirus infection. It is quite possible that the lymphocytes of patients similar to BRU, patients with the same gender, age, history and biochemical abnormalities, may have also reverse transcribed in cultures incubated with PHA. Recall Gallo proved that cultures of PHA stimulated normal lymphocytes reverse transcribe. In addition to avoiding bias, both experiments (test and control) must be performed blindly. That is, without the scientist knowing which is the test and which is the control.

Montagnier's omission of controls, a significant omission from virtually all HIV research, on its own negates any possibility of drawing definite conclusions from the experiment. In fact in all the experiments described in Montagnier's paper, no control data were reported.

On this basis alone it is very problematic that Montagnier's results were published.

Slide 28



Some may say we have misinterpreted Montagnier's findings, that is, Montagnier and his colleagues were fully aware of the non-specificity of reverse transcription and did not really interpret his reverse transcriptase activity in the manner we portray. However, we have not misinterpreted Montagnier. Five years after his paper was published, Montagnier and Gallo jointly published a paper in *Scientific American* in which they wrote, "The specimen [a portion of a BRU's lymph node] was minced, put into tissue culture and analysed for reverse transcriptase. After two weeks of culture, reverse-transcriptase activity was detected in the culture medium. A retrovirus was present but which one?"

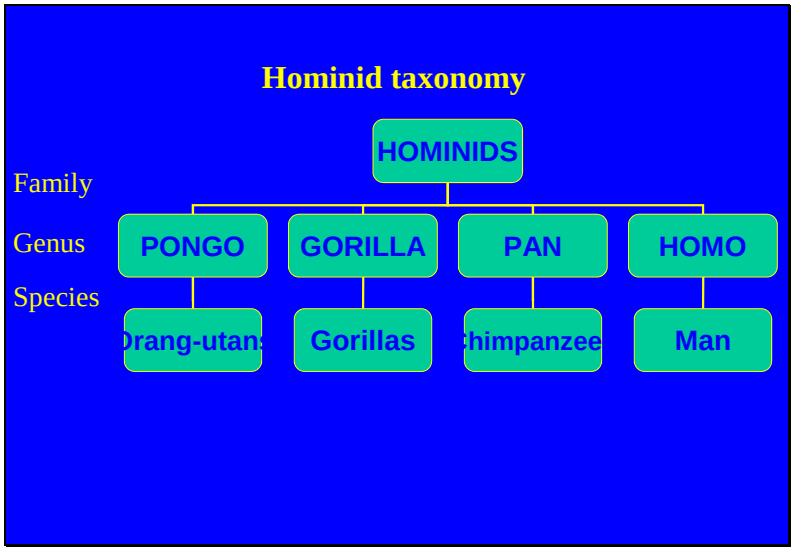
Hence despite being aware of the evidence to the contrary, both Montagnier and Gallo considered reverse transcriptase activity and this activity alone, proof that BRU was infected with a retrovirus and proof the virus was transmitted to healthy blood donor cells.

To answer the question "A retrovirus was present but which one?", Montagnier needed proof that not only had he isolated a retrovirus from BRU but this was a new retrovirus because, by 1983, Gallo had claimed

discovery of two human retroviruses known as human T-cell lymphotropic viruses type I and type II. (HTLV-I and HTLV-II).

What Montagnier wanted to show was that his discovery was different. Not one of the older human retroviruses but a new virus. What was his proof?

Slide 29

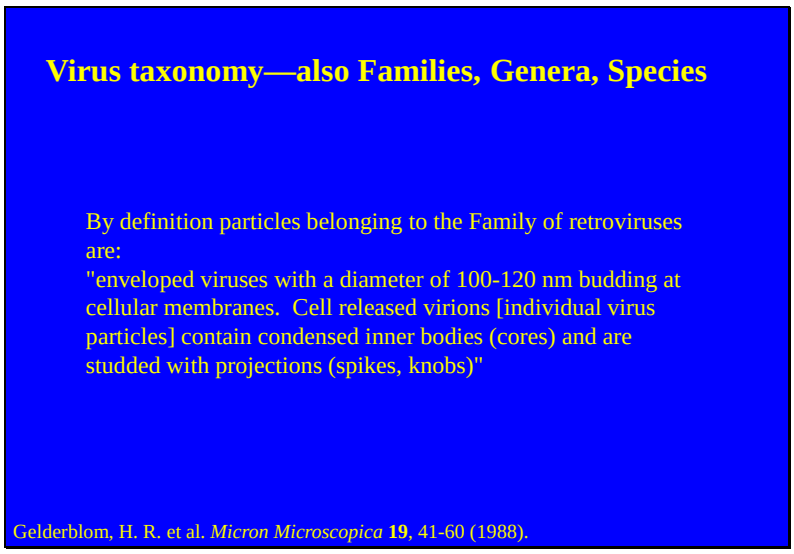


Before we can discuss this we need to make a brief digression into taxonomy.

Taxonomy is "the classification of organisms in an ordered system that indicates natural relationships" [Greek taxis, *arrangement*].

First of all, as we all know, the animal kingdom is divided into different categories. For example, humans belong to the Family called Hominids but since gorillas and chimpanzees share some features with us they too are included in this Family. But as we discern more detailed differences between humans, gorillas and chimpanzees, we see that each of these animals belongs to a different genus and a different species.

Slide 30



Retroviruses also are a Family. Not of animals of course but of viruses, the Family called *Retroviridae*. As expected, members of this Family share particular morphological features. These are particles:

- of diameter of 100-120 nm.
- possessing condensed inner bodies (cores).
- possessing an outer envelope in which there are embedded surface projections called knobs or spikes.

(Condensed means matter which is more highly aggregated and appears darker in EM pictures).

This classification is taken from a paper by Hans Gelderblom from the Robert Koch Institute in Berlin. He is one of the best known HIV researchers.

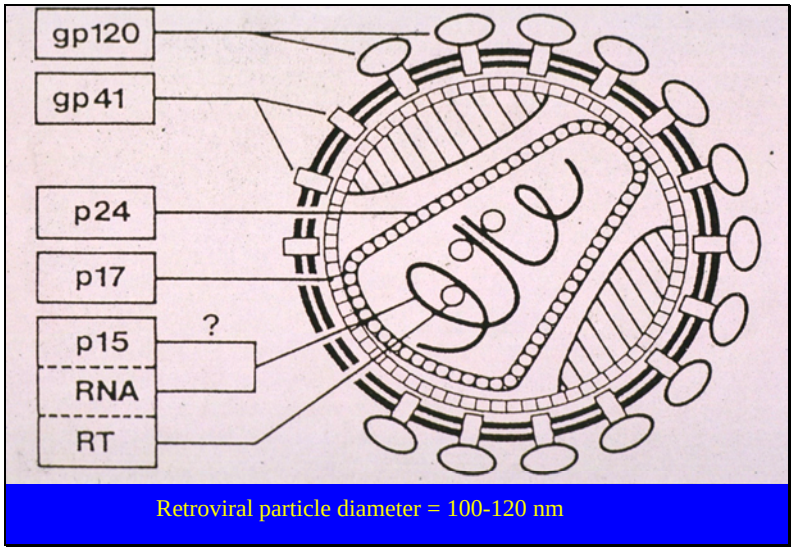
EXTRA
In 2000 the taxonomy of retroviruses changed. Here we deal with the classification that existed in 1983/84.

The newer classification (see link below) if anything makes matters more problematic in regard to particle taxonomy. In this taxonomy "Virions are spherical, enveloped and 80–100 nm in diameter".

Virus taxonomy Online
http://www.virustaxonomyonline.com/virtax/lpext.dll?f=templates&fn=main-h.htm

Retroviral taxonomy
http://www.virustaxonomyonline.com/virtax/lpext.dll/vtax/agp-0013/rtr03/rtr03-sec1-0001?f=templates&fn=document-frame.htm&2.0#rtr03-sec1-0001

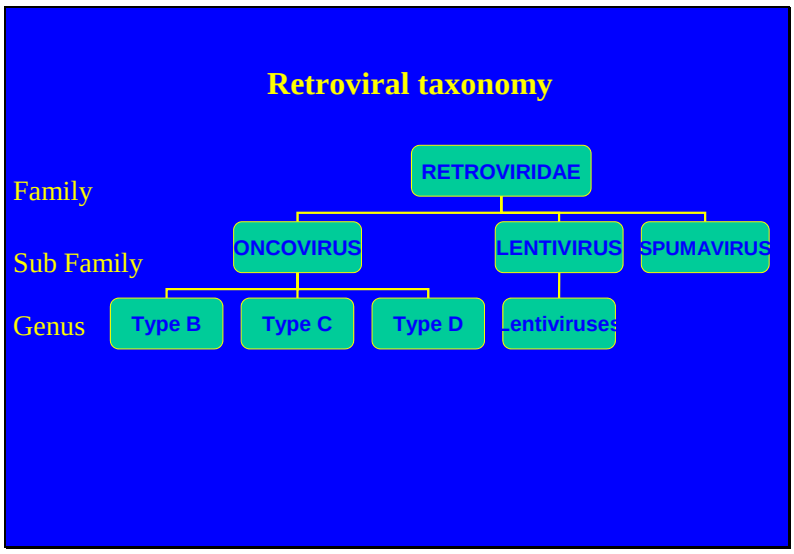
Slide 31



This is a diagram of the HIV particle published by Gelderblom. The main thing to see here is the diameter 100 to 120 nm, the conical core, the two lateral bodies and the knobs on the surface.

EXTRA
The boxes annotated gp120, gp41, p24, p17, p15, RNA and RT refer to the particle's constituent glycoproteins, proteins and RNA. (Recall that RT is a protein). A glycoprotein is a protein complexed wth sugar molecules.

Slide 32

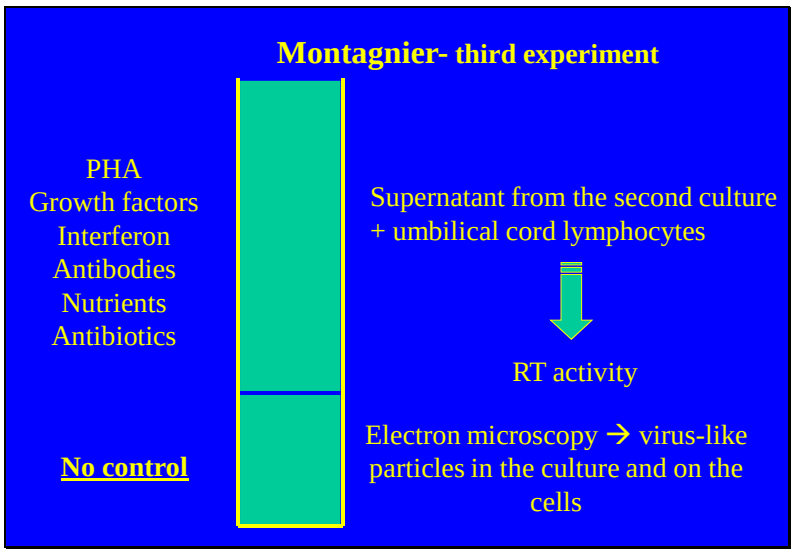


The *Retroviridae* Family is divided into sub-families known as Oncovirus, Lentivirus and Spumavirus.

Oncovirus is divided into three genera called Oncovirus type B, type C and type D.
Lentivirus has a single genus known simply as Lentiviruses.
Spumavirus is not relevant to this discussion.

We are now in a position to examine Montagnier's third experiment.

Slide 33



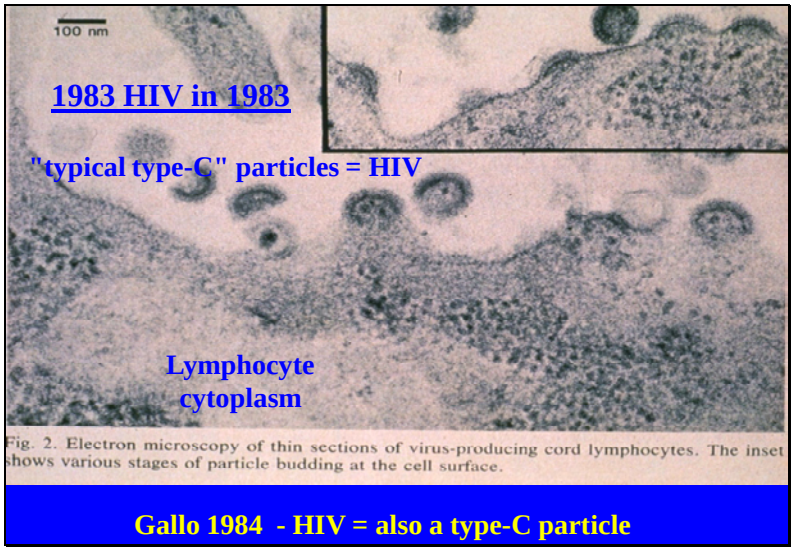
In this experiment Montagnier added supernatant from his second experiment to umbilical cord lymphocytes and PHA, growth factors, interferon antibodies, nutrients and antibiotics. Note that umbilical cord lymphocytes are cells obtained from the umbilical vein of fresh placentas and are the newborn's cells. They are not maternal cells.

After several days Montagnier again detected reverse transcriptase activity but on this occasion, and for the first time, Montagnier reported the presence in the cultures of particles which had some of the characteristics of retroviruses. These particles were both "budding" from and lying free outside the cell membrane.

Again, and it must be repeated, there were no controls. That is, umbilical cord lymphocytes cultured in another test tube to which were added supernatants originating from cultures of lymphocytes of a patient with clinical and biochemical abnormalities similar to BRU but not at risk of AIDS, as well as all the other chemical agents Montagnier employed. Hence again it is impossible to conclude that the particles originated from BRU's cells or that they were a virus.

EXTRA
In fact the particles could not have originated from BRU because the umbilical cord lymphocytes were cultured with cell-free supernatant from the co-culture. Even if the supernatant contained HIV particles the particles could not be infectious because the cell-free particles do not have knobs. According to the HIV experts, such knobs are critical to infectivity (see below).

Slide 34



This is the electron micrograph published by Montagnier.

In this slide the broad uninterrupted structure in the bottom half, is a portion of an umbilical cord lymphocyte. On its surface are some blebs (buds) and nearby are a few cell free particles. These appearances were reported as "virus-producing cord lymphocytes [showing] various stages of particle budding at the cell surface".

Montagnier classified these particles as genus type C particles of the subfamily Oncovirus. In fact in the abstract of the paper these are called "a typical type-C tumor virus" (=Oncovirus). In 1984, when Gallo reported his own experiments, he also classified the "new retrovirus" within the same genus, type-C particles.

EXTRA:
This is the slide shown by EPE in her evidence in chief and is the electron micrograph printed on page 869 of Montagnier's 1983 paper. Yet in the DPP's written submission it is stated:

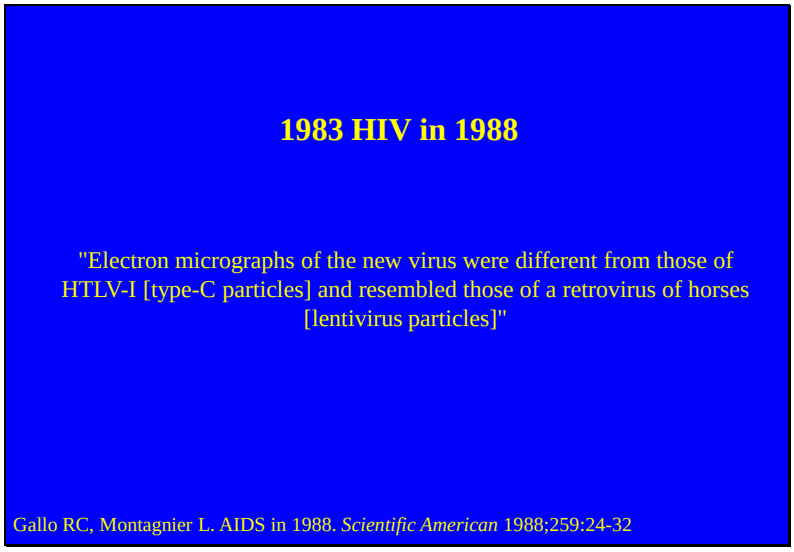
"Ms Papadopulos-Eleopulos claims that Montagnier he did not publish any picture of the virus in the culture. This is clearly incorrect as can be seen from page 869 of Montagnier's 1983 paper".

In fact it was one of the DPP's expert witnesses who was apparently unaware that Montagnier or Gallo had published electron micrographs of the virus. In his written report Professor French claimed that Montagnier' 1983 work was "groundbreaking". When he was asked if there were any deficiencies in the first paper claiming isolation he replied:

A. The initial isolation of the virus was by showing viral activity, showing it was a retrovirus, they hadn't actually seen a virus is, it wasn't until John Armstrong's publication that I have referred to and several other publications from Sweden and then subsequently the National Institutes of Health in America that we actually saw viral particles, viral structures.

Further on Professor French testified:
A. Yes, the initial isolation of the virus was from blood samples and lymph nodes of patients but the virus was not actually seen, it was detected indirectly. The first published report of the virus actually being seen in human tissues was by John Armstrong and his colleague Mr Horn and that was published in the Lancet in 1984 and has been confirmed by others since. (T790)

Slide 35



However, by the time of their joint 1988 *Scientific American* paper on the history of the discovery of HIV, Montagnier and Gallo no longer refer to HIV as an Oncovirus, type C particle but as a Lentivirus.

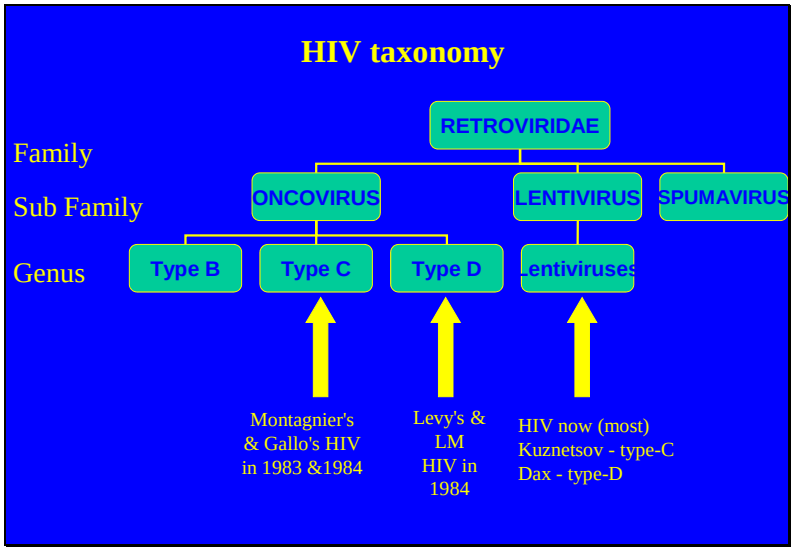
They wrote "Electron micrographs of the new virus were different from those of HTLV-I and resembled those of a retrovirus of horses". HTLV-I is a type C particle and the horse retrovirus is a Lentivirus. So in 1988, with no evidence and despite what Montagnier reported in 1983, Montagnier and Gallo said that what Montagnier saw in 1983 was not a type C particle but a Lentivirus.

This means that if HIV does exist and is truly a Lentivirus then either Montagnier's and Gallo's electron microscopers were unable to correctly identify retroviral particles in 1983/84 or what Montagnier and Gallo discovered in 1983/84 is not HIV.

This is just one example of the changing face of the new unique human retrovirus HIV.

EXTRA
See "The Perth Group revisits the existence of HIV".
http://theperthgroup.com/LATEST/PGRevisitHIVExistence.pdf

Slide 36



Even today there is no agreement as to what subfamily or genus HIV belongs.

1. In 1983 Montagnier and in 1984 Gallo said HIV is a type-C particle.
2. In 1984 Montagnier and Jay Levy, another US HIV expert from the Department of Medicine, Division of Hematology and Oncology, University of California, San Francisco, said HIV is a type-D particle.
3. Nowadays Montagnier and Gallo and many others say that HIV is a Lentivirus.
4. In 2003 Kuznetsov, a scientist at the Department of Molecular Biology and Biochemistry, University of California, reported HIV as a type-C particle [1].
5. In Constantine *et al's* textbook [2], published in 2005, co-authored by Professor Elizabeth Dax, HIV is classified as a type-D particle.

In order to get a feel for these taxonomic differences we reascend the evolutionary scale all the way back to Hominids.

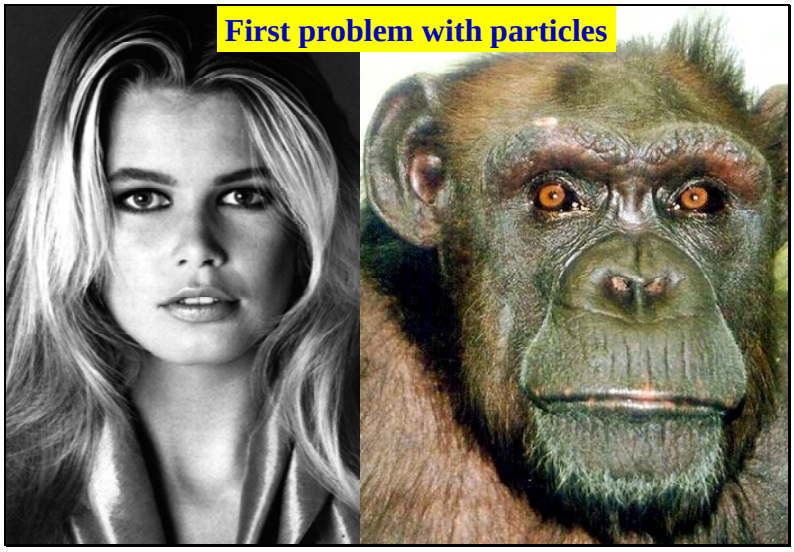
EXTRA

Kuznetsov wrote "The particles recovered from the medium and displayed on the glass substrate appear almost identical to those visualized on the surfaces of HIV-infected cells and are virtually indistinguishable from virions of MuLV". MuLV = murine leukaemia virus, is a type C Oncovirus particle.

1. Kuznetsov YG, Victoria JG, Robinson WE, Jr., McPherson A: Atomic force microscopy investigation of human immunodeficiency virus (HIV) and HIV-infected lymphocytes. J Virol 2003, 77:11896-11909

2. Constantine NT, Saville R, Dax E. Retroviral testing and quality assurance. Essentials for laboratory diagnosis. Halifax: MedMira Laboratories, 2005:701.

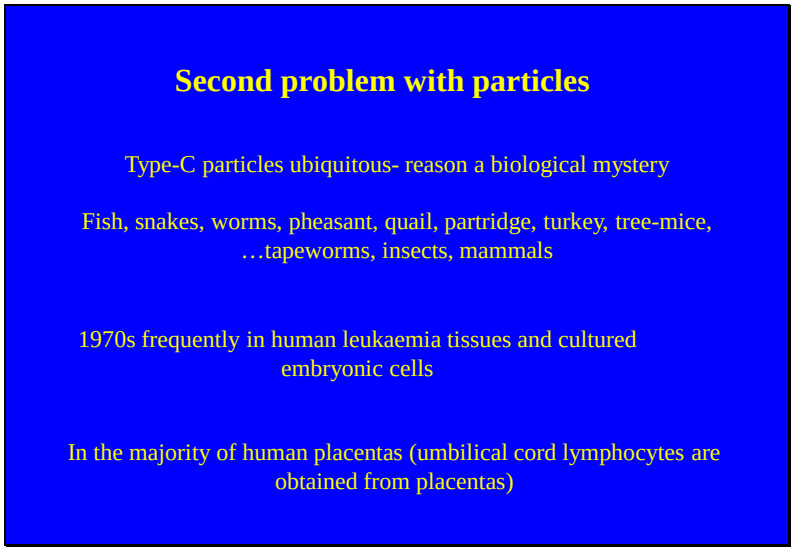
Slide 37



To say that HIV is both a type C particle and a type D particle is no different from saying an animal is both a human and a chimpanzee. To say that HIV is a type C particle and a Lentivirus is no different from saying an animal is both a human and a gorilla.

This taxonomical problem is what we refer to as the first problem with HIV particles. In our evidence this is the first of seven such problems.

Slide 38



The second particle problem is that type-C particles are ubiquitous. They range far and wide and their existence is a mystery. In the 1970s there were many reports of type C particles in human leukaemia patients, in embryonic cells and in the majority of human placentas. (In regard to the latter, the lymphocytes in Montagnier's third experiment were sourced from human placentas).

EXTRA
In the cross-examination on Professor Gallo stated: "Where they'd been seen [endogenous – retrovirus – like particles] occasionally is in normal human placenta" (T1276).
(In fact they are seen in the majority of human placentas [1]).

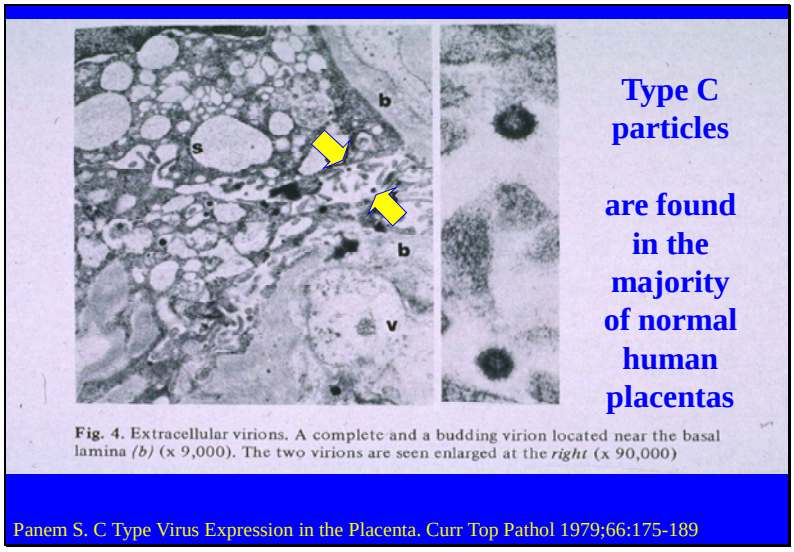
Also, writing in *Nature* in 1986 Gallo claimed that in 1984 his four *Science* papers had produced "clearcut evidence that the aetiology of AIDS and ARC was the new lymphotropic retrovirus HTLV-III [HIV]" [2]. However, in 1984 Gallo reported HIV as the same species as Montagnier's HIV, that is, as type C particle. Hence no matter how "clearcut" Gallo's evidence may have been, he was dealing with the wrong virus. (In fact the evidence was

not "clearcut" because there is no proof in Gallo's four *Science* papers of the HIV theory of AIDS).

1. Panem S. C Type Virus Expression in the Placenta. *Current Topics in Pathology* 1979;66:175-189.

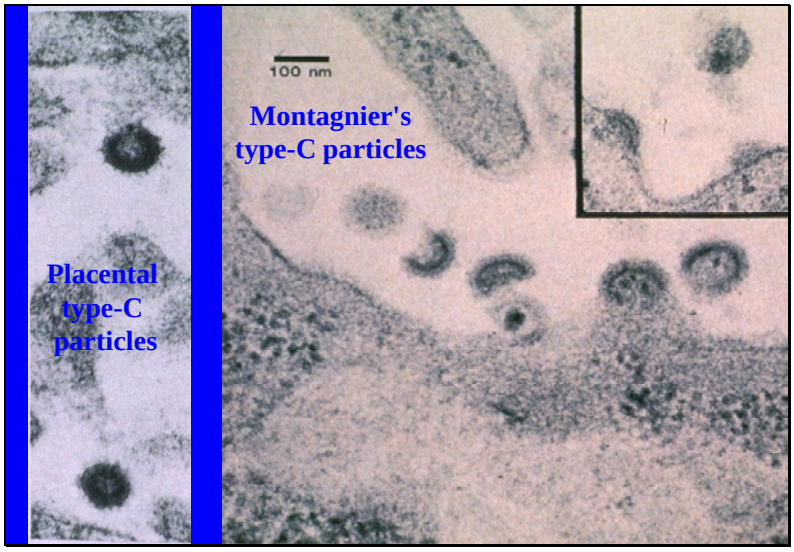
2. Gallo RC, Sarin PS, Kramarsky B, Salahuddin Z, Markham P, Popovic M. First isolation of HTLV-III. *Nature* 1986; 321:119.

Slide 39



Here it is a retroviral particle, a type-C particle, which is found in the majority of placentas. On the left is the low power magnification and on the right the high powered magnification.

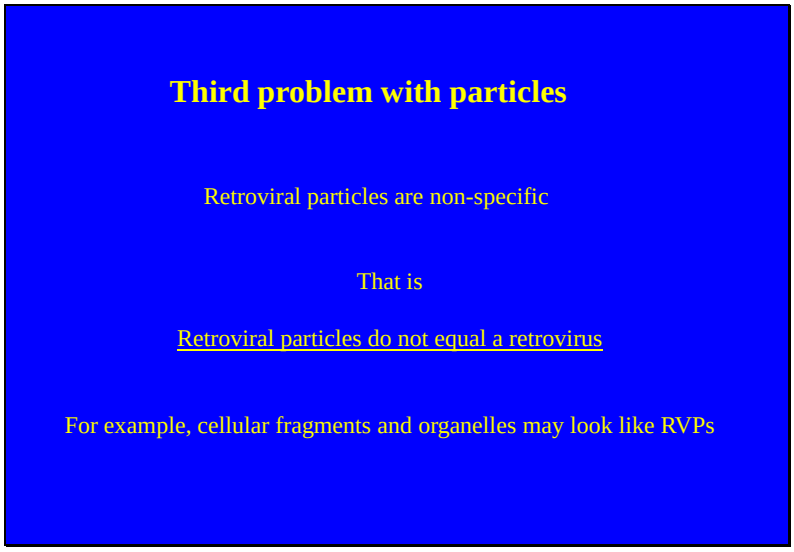
Slide 40



Here is an image of a placental type C particle alongside the Montagnier 1983 HIV particles. Those reported in 1983 as "typical type-C" particles.

Who can say that Montagnier's cultures would not have contained or produced such particles even if they had been made without supernatant from the co-culture containing lymphocytes from BRU? Or made with cell culture supernatants from sick, non-AIDS individuals? Without controls it is impossible to know and without controls these experiments should not have been submitted or accepted for publication.

Slide 41



To recapitulate, the first problem with the HIV particles is the lack of agreed taxonomy.

The second problem is that Montagnier and Gallo classified their 1983 and 1984 "HIV" particles as type-C particles of the subfamily Oncovirus yet these particles are found in many situations far removed from AIDS patients and there are no control experiments to obviate this fact.

The third problem is that the morphological appearances of retrovirus particles is nonspecific. By that we mean that non-retroviral particles may masquerade as a retrovirus. In the 1970s eminent retrovirologists* showed that in cell culture experiments designed to search for retroviruses, virus-like particles could appear which were nothing more than cellular fragments, microsomes or membrane bound vesicles. The occurrence of this cellular material was especially likely when lysis (breaking up) of cells occured (cells have a tendency to disintegrate in culture despite the best efforts to maintain their viability), or lysis was deliberately induced as part of the experiment, which is often the case in HIV research. It was also acknowledged that some of these cellular structures could also contain RNA or reverse transcriptase. (This may not be surprising since these

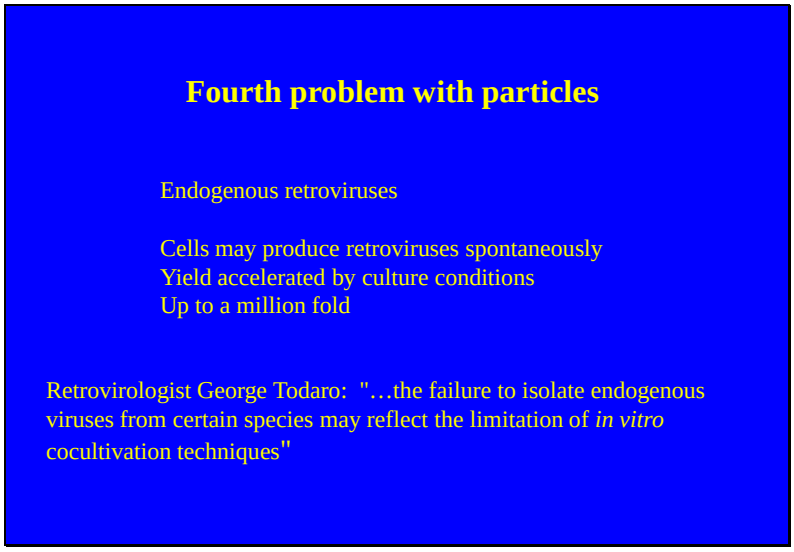
occur inside cells). Hence these non-retroviral particles could both structurally and biochemically resemble retroviruses.

EXTRA This was acknowledged by Gallo in 1976 "Release of virus-like particles morphologically and biochemically [with RT activity] resembling type-C virus but apparently lacking the ability to replicate have been frequently observed from leukaemic tissue" [1].

*Including Howard Temin who discovered reverse transcriptase and for this was awarded the 1975 Nobel Prize.

1. Gallo RC, Wong-Staal F, Reitz M, Gallagher RE, Miller N, Gillespie DH. Some evidence for infectious type-C virus in humans. In: Balimore D, Huang AS, Fox CF, eds. Animal Virology. New York: Academic Press Inc., 1976:385-405.

Slide 42



We repeat, Montagnier interpreted RT activity in his first two experiments as proof of virus production, isolation and transmission. However, viruses are particles and not an enzyme activity. And since Montagnier did not have proof that particles of any kind existed in the first two experiments, there is no proof that particles of any kind were the source of a reverse transcribing enzyme. The same applies in Montagnier's third experiment, where he demonstrated type C retroviral particles. There is no proof these particles were the source of such an enzyme.

How then do we account for the particles Montagnier demonstrated in his third culture? Do these particles prove they must have been present in the first or second culture and hence be proof of transmission of a retrovirus, as Montagnier claimed? No.

All retrovirologists including Temin, Todaro, Duesberg, Weiss and Gallo have pointed out that cultured cells in general, and in particular, chemically stimulated cell cultures or cells co-cultured with other cells (the types of cultures practically ubiquitous in AIDS research), release retrovirus-like particles even when not infected with a retrovirus.

One explanation for this apparently strange and unexpected phenomenon is the presence in cells of what is known as endogenous retroviruses. (Endogenous means "from within", the opposite of "exogenous, "from without"). Unlike all other viruses, whose presence means acquisition from without, retrovirus-like particles can arise *de novo*. This is because animals including humans are said to be born with retroviral DNA which they inherit from their parents. It is estimated that about 10% of the human genome contains such endogenous retroviral genetic sequences.

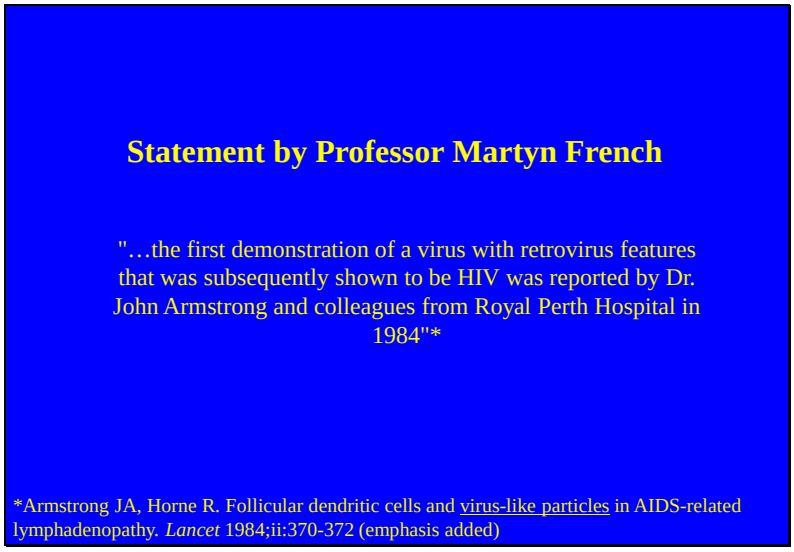
In 1976 George Todaro, the eminent retrovirologist, stated "the failure to isolate endogenous viruses from certain species may reflect the limitation of in vitro cocultivation techniques". In everyday language this means that if a scientist knew how to create the right culture conditions, he could induce the production of retrovirus particles in any cell culture.

Hence virologists draw a distinction between endogenous and exogenous retroviruses and we repeat, before the AIDS era it was known that endogenous retrovirus phenomena, such as RT activity and particles, can even arise spontaneously in cell cultures. Before the AIDS era scientists also had shown that the rate of appearance of endogenous viruses could be accelerated up to a million times by exposing the cultures to radiation or chemical stimulants or co-culturing them. The use of such agents is ubiquitous in HIV research. This means that both the RT activity and the particles Montagnier reported could have arisen for reasons unconnected to an exogenously acquired retrovirus.

EXTRA
Endogenous retroviruses pose a significant problem in biological research. So much so that in 1978 Professor Robin Weiss, a British retrovirologist, published a scientific paper in which he warned, "Retrovirus genomes exist as endogenous genetic elements in the cells of many species used in biomedical research. Many cell lines spontaneously release virus, and other cells are induced to do so by procedures commonly used in research laboratories…The expression of endogenous retroviruses can affect the results of seemingly unrelated experiments. Some retroviruses endogenous to animals grow avidly in human cells the activation of endogenous viruses may affect the significance of results obtained in a variety of experiments". There are "situations where endogenous virus …could affect the results or interpretation of biomedical research" [1].

1. Weiss RA: Why cell biologists should be aware of genetically transmitted viruses. National Cancer Institute monograph 1978:183-189

Slide 43



Thus far we have been concentrating on particles that occur in cell cultures. That is, outside the body in test tubes. What scientists refer to as *in vitro.* What about particles that occur inside the body? Which scientists refer to as *in vivo*.

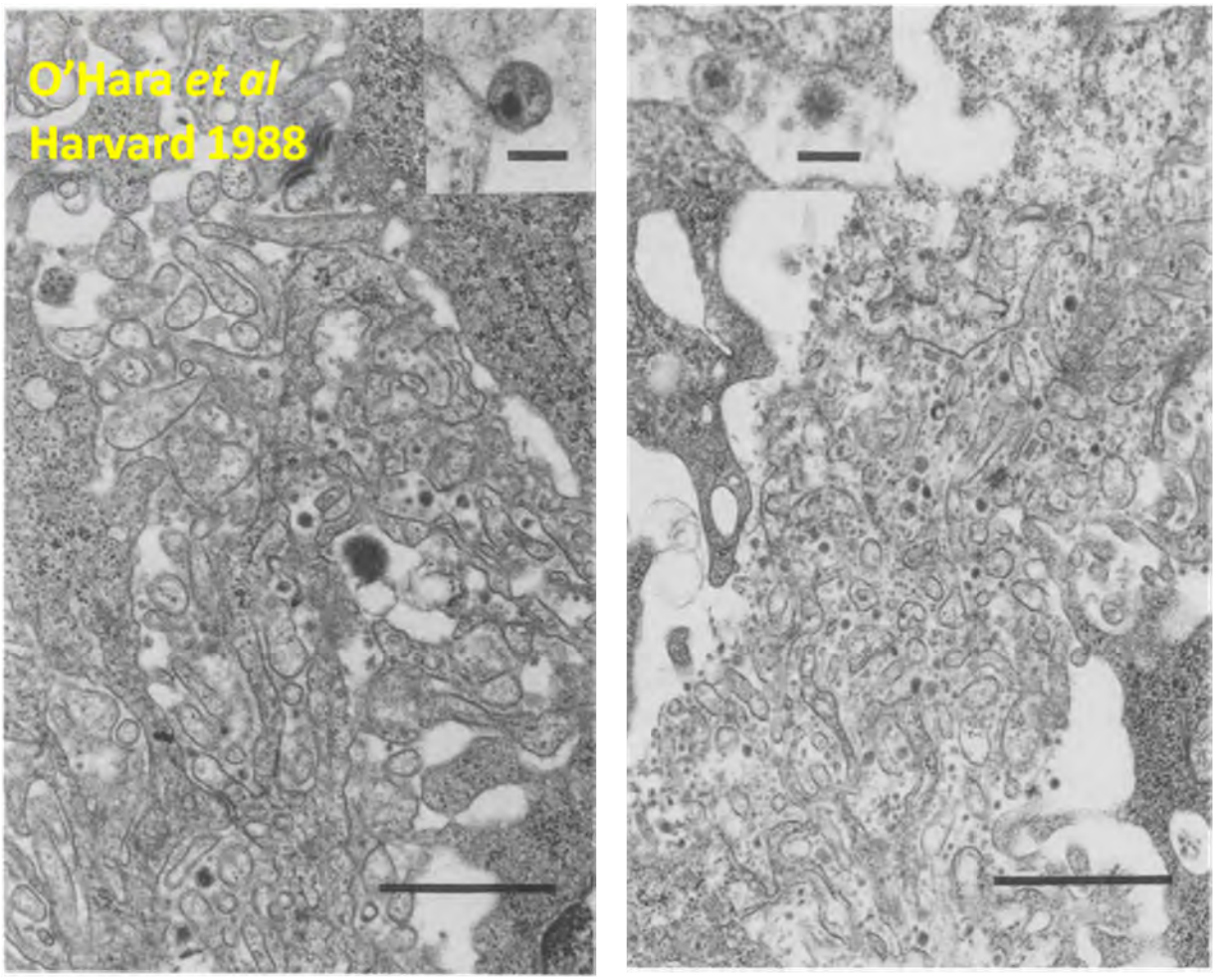
Instead of submitting affidavits, as both EPE and VFT were instructed to do, the prosecution witnesses presented statements to the court. The quote on the slide is from Professor French's first statement. As the title of the Armstrong paper says, in 1984 Dr Armstrong referred to the particles as "virus-like", not a retrovirus. This is an entirely appropriate description because the appearance of particles that look like a virus is not proof such particles are a virus. To obtain such proof requires data on replication.

EXTRA
During his cross-examination by Mr. Kevin Borick Professor French said the following:

Q. Do you know what type of particle that he looked at, what was the type.

A. They were whole viruses and I will provide the court with pictures...[viruses are always "whole", see slide 3].
Q. Obviously we won't be able to do that right now, but could you tell us what type the HIV particles, what are they said to be.
A. What Dr Armstrong saw in the lymph nodes was a structure, multiple structures that had the morphology of a virus. They were not particles, they were not part of the virus, they were whole viruses.
Q. What is HIV said to be, what sort of a virus.
A. It is a retrovirus.
Q. Isn't it regarded as a lentivirus.
A. A lentivirus, yes, it belongs to the lentivirus group, and lenti- meaning slow...
Q. Is it true that the particles which are the same as those which are said to be HIV have been found in the same proportion of AIDS–related and non-AIDS–related disease lymph nodes and the proportion I put to you is 90%.
A. No. These viral structures have only been found in the lymph nodes of patients with HIV infection… (T819)

Slide 44



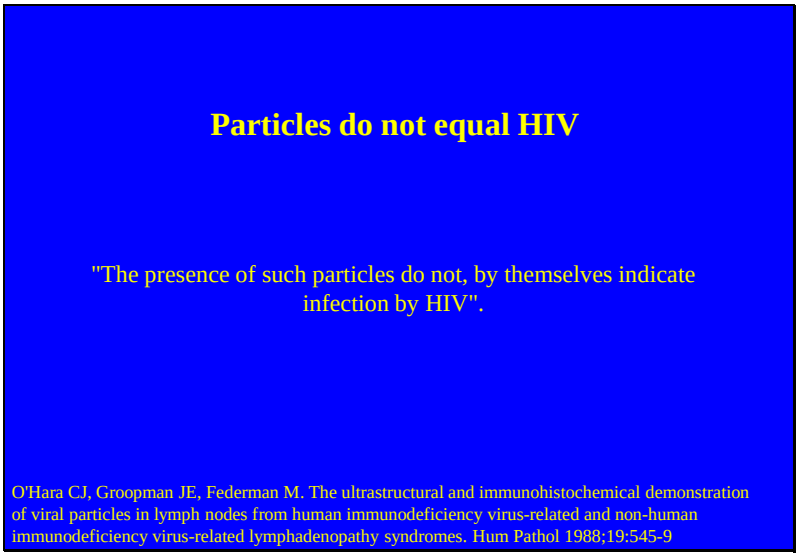
18/20 AIDS related 13/15 non-AIDS related

The minimum absolutely necessary but not sufficient condition to exclude the possibility that the particles seen by Armstrong were not cellular phenomena, unrelated to viruses, is to have controls. Professor Armstrong did not have controls.

However, in a paper published in 1988 researchers from Harvard University did report control experiments. In fact, to the best of our knowledge, this is the only study in all AIDS research which included proper controls and was conducted blindly.

The research showed that the "HIV particles" were present in 18/20 (90%) of patients with enlarged lymph nodes attributed to AIDS. However, identical particles were found in 13/15 (87%) of patients with enlarged lymph nodes not attributed to AIDS and at no risk for developing AIDS.

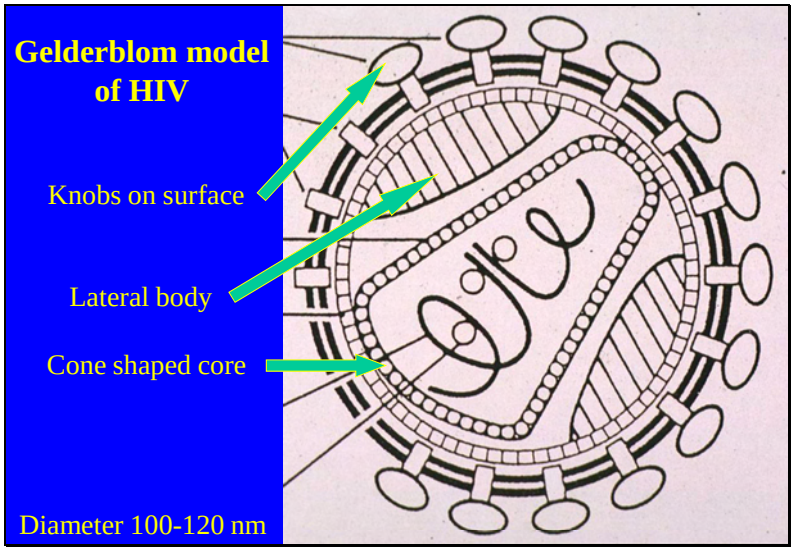
Slide 45



On this basis they concluded "The presence of such particles do not, by themselves, indicate infection with HIV" [1].

1. O'Hara CJ, Groopman JE, Federman M. The ultrastructural and immunohistochemical demonstration of viral particles in lymph nodes from human immunodeficiency virus-related and non-human immunodeficiency virus-related lymphadenopathy syndromes. Human Pathology 1988; 19:545-9.

Slide 46

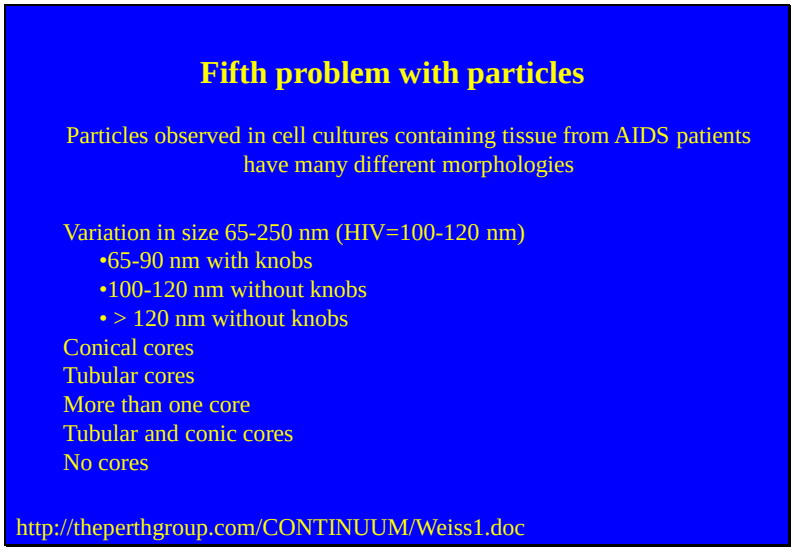


This slide is a reminder of the principal morphological characteristics of the putative HIV particle, as published by Hans Gelderblom.

1. A diameter between 100 and 120 nM.
2. A cone shaped core.
3. The presence of lateral bodies
4. Knobs on the surface. According to all HIV experts the knobs are critical for the particle to be infectious, that is, enter into the cell in which the particle replicates.

Bearing these features in mind let us now consider a fifth problem with particles.

Slide 47



In electron micrographs of cultures containing tissues from AIDS patients there is a plethora of particles.

Particle diameters vary between 65-250 nm.

Particles with a diameter of 65-90 nm have knobs while those with the "correct" diameter, 100-120 nm, do not have knobs.

Particles with a diameter greater than 120 nm devoid of knobs.

Some particles have conical cores while others have tubular cores.

Some particles have more than one core

Some particles contain a tubular and a conical core.

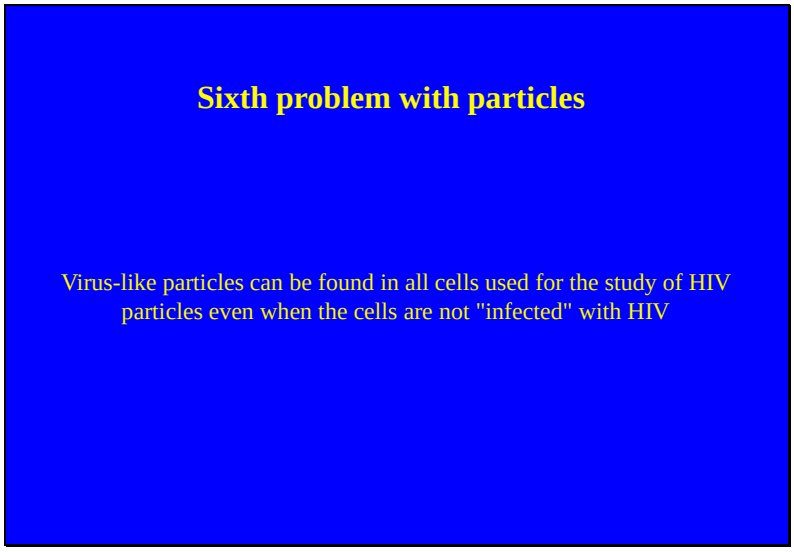
Some particles have no cores.

If we accept for argument's sake that the 100-120 nm particles are HIV and originated from AIDS patients who are infected with HIV, several question arise:

1. What are all the other particles?
2. Are these virtually forgotten, never discussed particles viruses?
3. Where did they come from?
4. With so many types of particles how is it possible to know which particle or particles are viruses?
5. Do any of the particles causes AIDS and which one(s)?
6. Do the particles cause AIDS or does AIDS or the culture conditions cause (produce) the particles?

For references see
http://www.theperthgroup.com/CONTINUUM/VirusChallenge.pdf
and search for "HIV particles look different" from "naturally existing viruses".

Slide 48

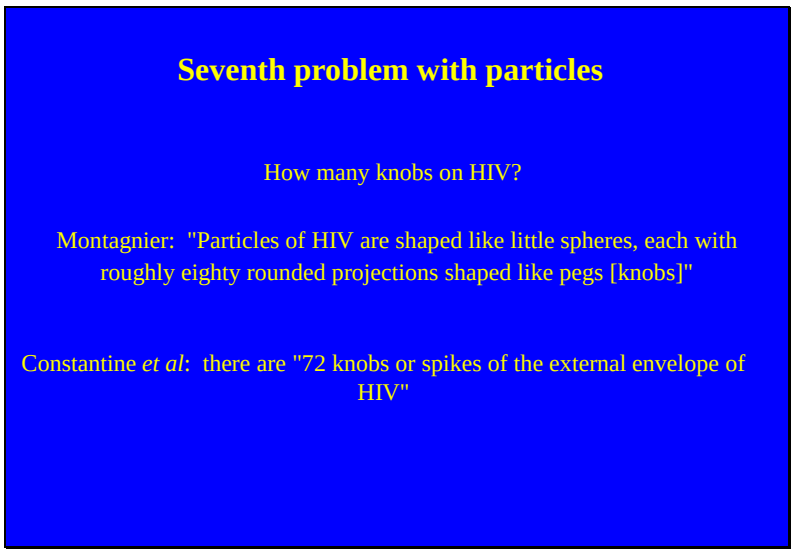


Here is the sixth problem with particles.

In the vast majority of cases HIV experts conduct experiments where they take cells from AIDS patients, whose lymphocytes they claim are infected with HIV, and for unknown reason(s) culture these with other cells (co-cultures). The other cells may be normal lymphocytes (as Montagnier used) but most often they are immortal cell lines. That is, they are cancerous lymphocytes which do not die in cultures as most cells do. In fact it is from such co-cultures that HIV experts and biotechnology companies obtain "HIV" for various purposes. Including producing vast amounts of "HIV" proteins and RNA for diagnostic tests, as well as conducting research into pathogenesis and the development of vaccines.

Significantly, cell lines used in co-culture experiments have been found to release retrovirus-like particles even when not cultured with cells from AIDS patients. In fact, Montagnier, in his 1997 interview with Djamel Tahi Montagnier stated that in such cultures it was a "real soup" of retroviruses.

Slide 49



The seventh problem with HIV particles concerns the number of knobs on the surface of the particle.

We may ask, how many knobs are there? According to Montagnier, in his book *Virus* [1], published in 2000, "Particles of HIV are shaped like little spheres, each with roughly 80 rounded projections shaped like pegs", or spikes, as they are also known.

In the 2005 textbook co-authored by Professor Dax, there are "72 knobs or spikes of the external envelope of HIV".

It may be argued that a discrepancy of eight knobs per particle is not significant and this may be true.

1. Montagnier L. *Virus.* New York: WW Norton & Company Inc, 2000.

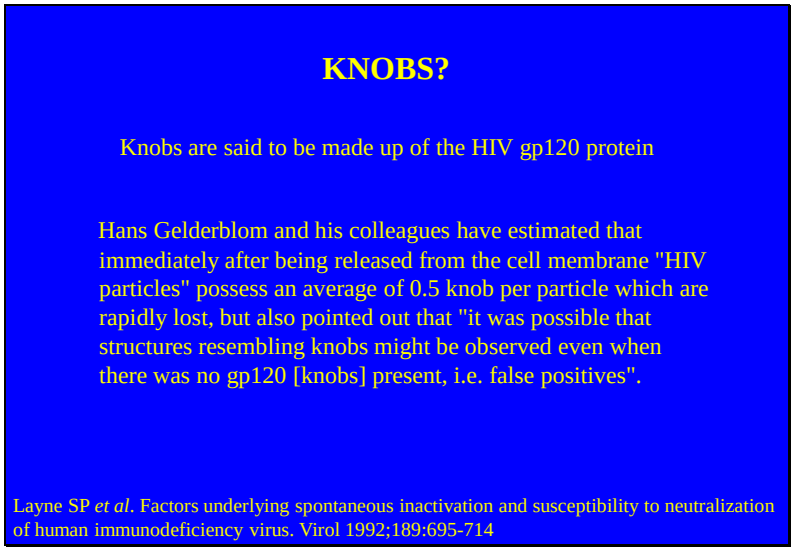
Slide 50

Before we further discuss the number of knobs we should remind ourselves of their function. What are the knobs for?

All the experts agree that it is the knobs that provide the viral particle with the ability to latch on to and thus get inside cells as the preliminary step in their replication. If the particle does not have knobs it cannot infect the cell. No knobs, no infection. Without knobs the particle cannot be an infectious particle. Knobs are critical. There is no exception. No knobs, no infection, no virus.

HIV experts claim the knobs are made of a protein, which is said to be an HIV protein, and is called gp120. The "p" stands for protein and the "g" stands for "glyco", from the Greek word "glycos" meaning "sweet", to indicate the protein contains sugar molecules. The "120" indicates the glycoprotein's molecular weight in thousands. The experts further say that it is the gp120 in the knobs that undertakes the task of fusing the particle to the cell surface thus allowing the particle to enter the cell.

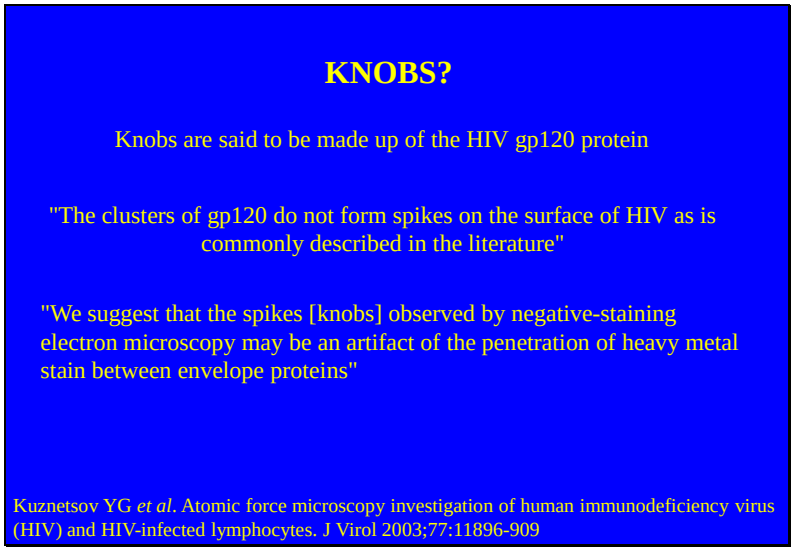
Slide 51



In a paper published in 1992 by Hans Gelderblom, who is the best known expert on the electron microscopy of HIV particles, he and his colleagues estimated that immediately after being released from the cell membrane HIV particles possess on average 0.5 knobs, that is, half a knob, per particle, and that the knobs are rapidly lost.

Equally important is the fact that Gelderblom and his colleagues, which include John Moore, did not exclude the possibility that the structures they reported as knobs may not have been knobs but artifacts, "false positives". In other words, the particles may have zero knobs.

Slide 52



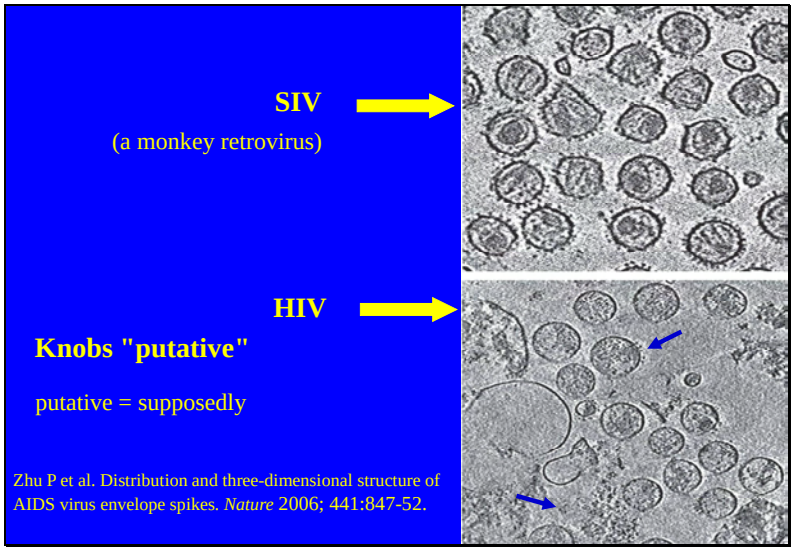
In a paper published in 2003 by researchers using one of the most modern methods to study virus particles*, Kuznetsov and his colleagues contradicted what virtually all HIV experts claim. They reported that "The clusters of gp120 do not form spikes on the surface of the HIV as is commonly described in the literature. The clusters are hardly protrusions at all. We suggest that spikes, knobs, observed by negative-staining electron microscopy may be an artifact of the penetration of heavy metal stain between envelope proteins. Indeed, the term "spike" appears to have assumed a rather imprecise, possibly misleading definition, and might best be used with caution" [1]. In other words, this posits zero knobs on the so called "HIV" particle. Such particles cannot be infectious and thus cannot be a virus.

Hence the literature is contradictory in regard to the number of knobs. The knob count for the HIV particle has been reported as 80, 72, 0.5 (on average), possibly zero and actually zero.

EXTRA
*Atomic absorption spectrometry
http://en.wikipedia.org/wiki/Atomic_absorption_spectroscopy

1. Kuznetsov YG, Victoria JG, Robinson WE, Jr., McPherson A. Atomic force microscopy investigation of human immunodeficiency virus (HIV) and HIV-infected lymphocytes. Journal of Virology 2003; 77:11896-909.

Slide 53



This slide, which also concerns knobs, is taken from a paper published in 2006 by Zhu and his colleagues from the USA*. (In this reproduction we have removed the authors' highlighting and introduced our own). Note: there were no controls, the study was not blind, and we would expect that like most authors publishing papers, these would be their best images.

The top right hand side of this slide is an electron micrograph of a retrovirus known as simian immunodeficiency virus, SIV. It is not difficult to see knobs on their surfaces.

Below this there is an electron micrograph of the HIV particles. We are unable to see any knobs on these particles apart what appear to be five knobs where the upper arrow is pointing. (This is the only particle on which the authors indicated there were knobs).

Furthermore, there are similar appearances where the lower arrow is pointing, in a part of the micrograph where there are no particles which means the "knobs" may just be artifacts.

We should also note that the authors refer to the knobs on the HIV particles as "putative", which means "supposedly".** Hence the authors themselves are unconvinced they are dealing with knobs.

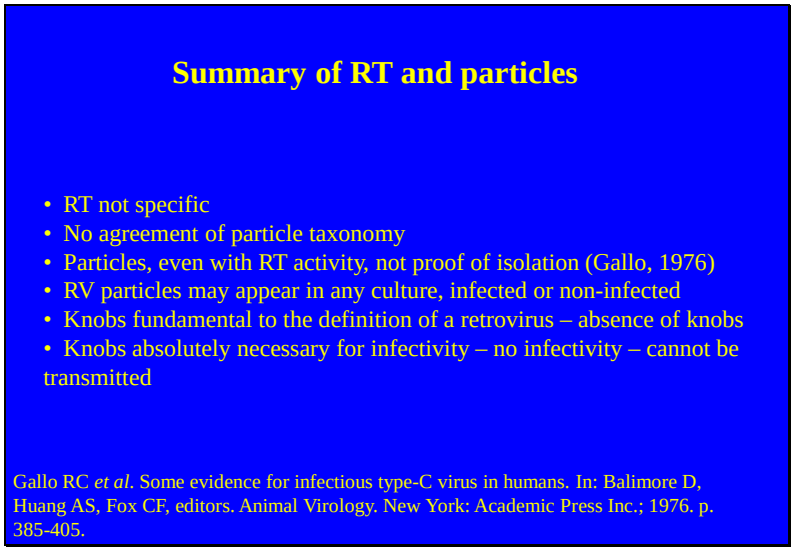
EXTRA
The Perth Group wrote a small commentary on this paper which we submitted to the journal *Nature* which the editor declined to publish.***

Zhu and co-authors affiliations:
Department of Biological Science and Institute of Molecular Biophysics, Florida State University, Tallahassee, Florida 32306, USA.
AIDS Vaccine Program, SAIC Frederick Inc, National Cancer Institute at Frederick, Frederick, Maryland 21702, USA.
Vaccine Research Center, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, Maryland 20892, USA.

**The caption to their figure 1 reads (in part):
"b right, HIV-1 Z-stack. Examples of putative Env [envelope] spikes on selected virions are indicated by arrowheads on the left".

*** http://theperthgroup.com/LATEST/ZhuNatureRejected.doc

Slide 54



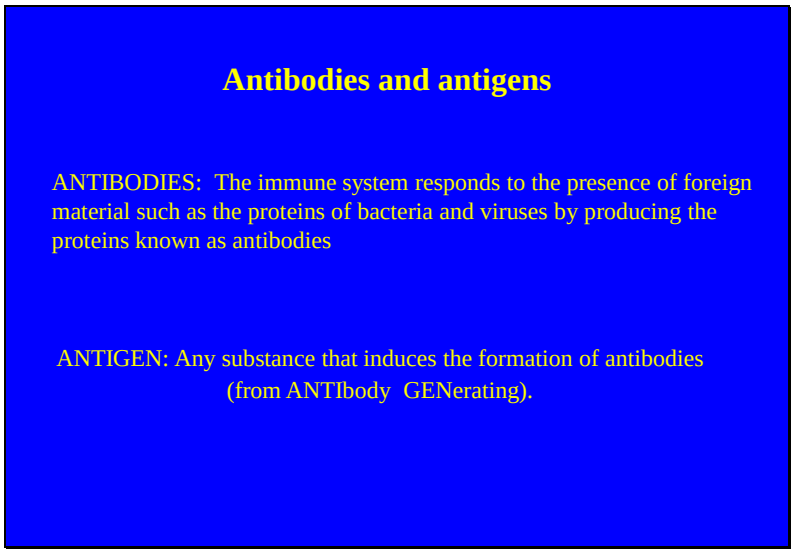
To summarise the evidence so far for the existence of HIV.

1. Reverse transcription is detection of the presence of an enzyme activity which is not specific to retroviruses.
2. Enzyme activity is not isolation of anything. Including a virus.
3. More than twenty year after its discovery there is no agreement in regard to the taxonomy of the particle. Or, to put it another way, taxonomically the HIV particle belongs to several mutually exclusive classifications.
4. Scientists accept the existence of retrovirus-like particles, even with RT activity or RNA, which are not infectious. Hence they cannot be viruses.
5. Retroviral-like particles are ubiquitous.
6. Retroviral-like particles appear in cell cultures which are not infected with "HIV".
7. Knobs are fundamental to the definition of retrovirus but so far nobody has proven they exist. If the particles said to be a unique retrovirus HIV do not have knobs they cannot be the "HIV" particles and they cannot be transmitted. Such particles cannot be a virus.

Montagnier's interpretation of his first three experiments was proof that BRU was infected with a retrovirus. He then wanted to prove that his virus was not one of the two human retroviruses Gallo claimed to have discovered earlier. These were known as HTLV-I and HTLV-II (HTLV=human T cell lymphotropic virus).

In order to prove his virus was new, and to characterise its proteins, that is, determine what proteins belonged to the virus, Montagnier used antibodies. So we need another digression to define antibodies, antigens and antibody tests.

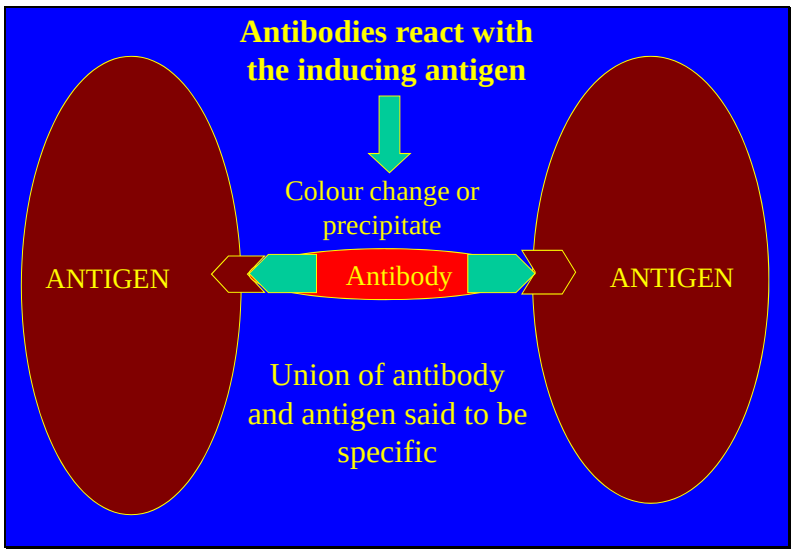
Slide 55



Antibodies are proteins produced by cells of the immune system known as B lymphocytes. One stimulus to the production of antibodies is the introduction something foreign into the body such as a bacterium or virus. Any substance that incites the production of antibodies is given the generic title antigen (ANTIbody GENerating) and may also be referred to as the "immunising" antigen. The antibodies that develop in response to a particular antigen are said to be "directed against" that antigen. Sometimes this is shortened to "antibodies to" that antigen.

One of the main properties of antibodies is that they react with the inducing or immunising antigen. Hence the theory is that if a person is infected by a virus for example, because the particular virus and its constituent proteins are foreign, the body's immune system will detect their presence and generate antibodies that react with the proteins of the virus. This theory is in fact the basis of antibody tests for a virus.

Slide 56

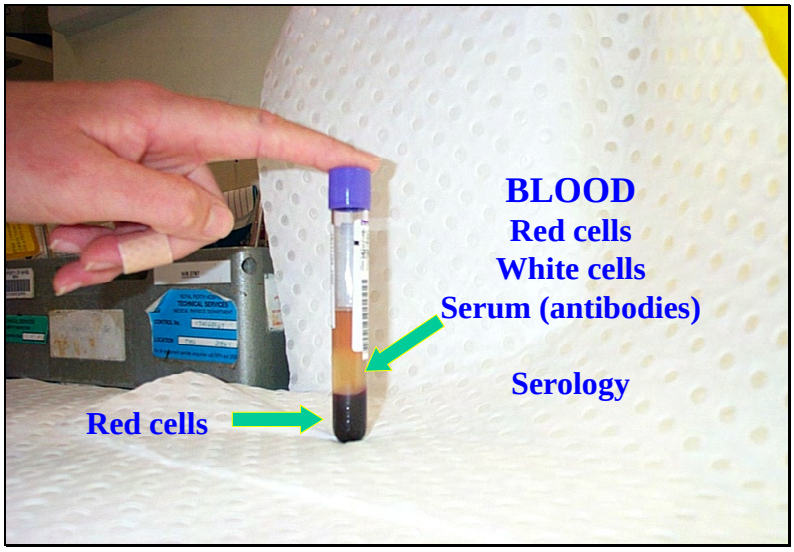


Immunologists have long taught that the union between antibody and antigen is specific. By this they mean the antibody reacts with the immunising antigen and no other antigen. In other words, the relationship between antibodies and antigens is monogamous. On this basis the antigen can be used in a test to identify the antibody. Because the union between antibody and antigen is specific, if antigen X reacts with an unknown antibody then the antibody must be directed against X. And for that antibody to exist the patient must have become infected with X. If X is a viral protein then a reaction is an indirect test for the presence of a virus infection.

How does the scientist know there has been a reaction? Simply by the fact that as the reaction takes place it produces some physical alteration in the appearance of the reaction mixture or medium. For example, fluid in a test may change colour or a precipitate may form.

Hence in order to perform an antibody test one has to obtain the virus proteins and the patient's antibodies. The antibodies are dissolved in the patient's serum.

Slide 57



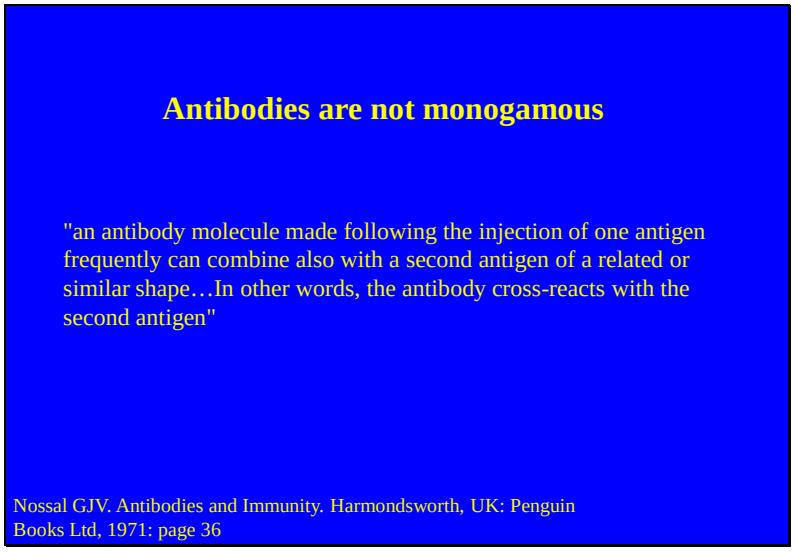
The main components of blood are plasma and red blood cells. Plasma is the yellowish fluid which makes up about half of a given volume of blood and in which antibodies are dissolved. The other half is made up predominantly of red blood cells, which transport oxygen around the body. The small remainder consists of white blood cells which include lymphocytes, one of which, the T4 lymphocyte, is depleted in AIDS.

When blood is removed from the body it clots and during this process clotting factors disappear from plasma leaving behind a liquid known as serum in which the antibodies remain. Because antibodies are in serum the practice of using antibodies as a test is sometimes called serology or serological diagnosis.

To do an antibody test serum, usually diluted, is added to a test tube containing the viral proteins.

Note: For an antibody test to "work" one must first know the identity of the proteins. In other words, in a serological test for a virus one must know the proteins are viral and not from some other source.

Slide 58

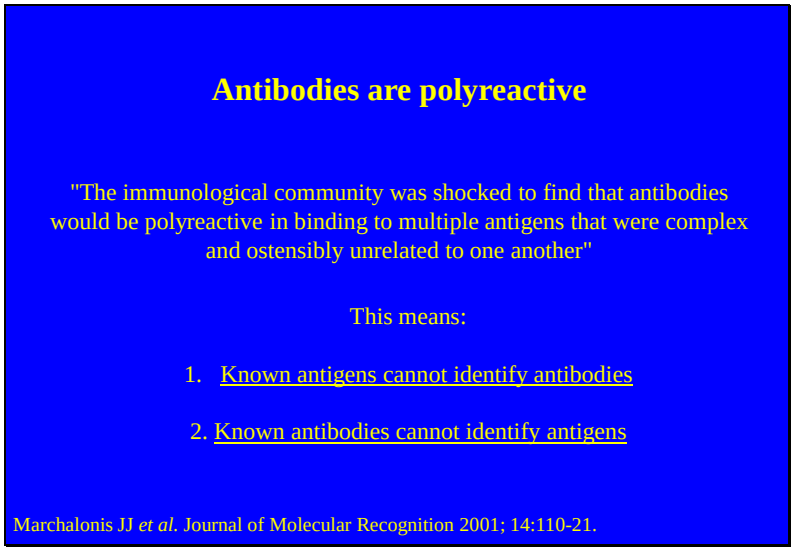


Unfortunately, the notion that the union between antibody and antigen is monogamous is not true*.

An antibody directed against one antigen can frequently combine with a second antigen. Immunologists refer to this as cross-reactivity or polyreactivity.

This is not unexpected. The whole of chemistry is based on the fact that molecules of one kind or another react with other molecules of one kind or another. There is no reason why proteins should be exempt from this general principle regardless of what they are or what function they perform. In fact one of the most rapidly growing areas of biomedical research is the study of protein-protein chemical reactions. There are now several journals devoted to this area of study.

Slide 59



Antibody polyreactivity is a chronically neglected inconvenient truth and a fact that has been known since antibodies were discovered in the late nineteenth century.

Yet only in 2001 did Marchalonis write how the immunological community was "shocked" to discover that an antibody induced by one antigen could also react with a second antigen or multiple antigens. And the antigens do not have to bear a chemically identifiable relationship to one another. The antibody can react even when the scientist cannot identify any similarity between the antigens.

This means it is impossible to identify an unknown antibody because it reacts with a known antigen, or *vice versa*, that is, that a known antibody can identify an unknown antigen. Yet the existence of "HIV" is built upon such a false notion.

EXTRA
When Marchalonis used the word "shocked" in regard to antibody cross-reactivity he cited papers from 1990, 1984 and 1965.

Marchalonis used the descriptor "promiscuous" and in his paper this word occurs twenty times.
In the immunological literature use of this term is now commonplace.

Notwithstanding, the immunological community appear unable or unwilling to revise its stance on how antibody/antigen reactions may be interpreted or indeed identify the circumstances where they can be interpreted at all.

For example,

In his 1997 interview Montagnier stated "antibodies are very specific. They know how to distinguish one molecule in one million. There is a very great affinity. When antibodies have sufficient affinity, you fish out something really very specific. With monoclonal antibodies you fish out really ONE protein. All of that is used for diagnostic antigen detection".

In his written report for the court Sir Gustav Nossal wrote "...high affinity monoclonal antibodies are extensively used in research as razor-sharp and highly specific identifiers of various structures".

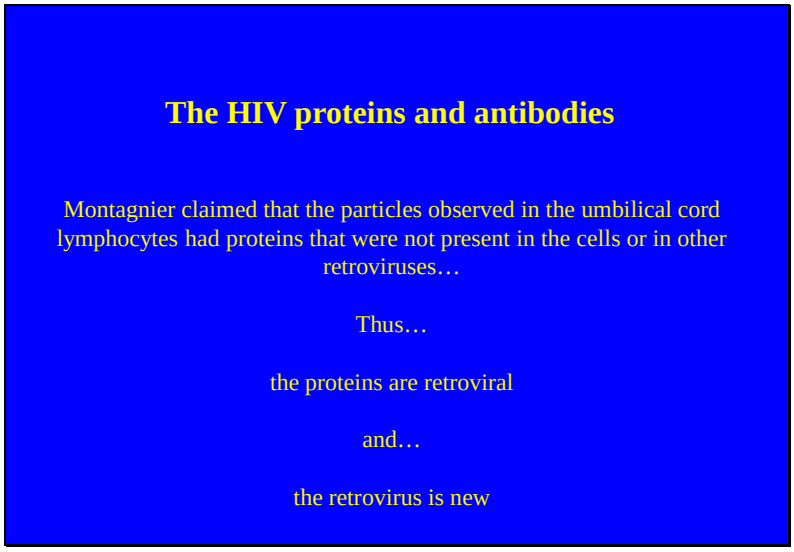
One should also note that according to Marchalonis, "As pointed out by Van Regenmortel (1998), there is no necessary correlation between affinity and specificity because low affinity antibodies can show better discrimination among antigens than the high affinity binders" [1]. And, in 1997 Kramer *et al* [2] wrote "high-affinity antibodies that have undergone antigen-driven somatic mutations are usually thought to be monospecific. Nevertheless, antibody cross-reactivity and polyspecificity have been observed since the earliest immunological studies" and "even high-affinity binding monoclonal antibodies are able to recognize [react with] more than one peptide epitope" [peptide epitope = antigen].

1. Van Regenmortel MH. From absolute to exquisite specificity. Reflections on the fuzzy nature of species, specificity and antigenic sites. *Journal of Immunological Methods* 1998; 216:37-48.

2. Kramer A, Keitel T, Winkler K, Stocklein W, Hohne W, Schneider-Mergener J: Molecular basis for the binding promiscuity of an anti-p24 (HIV-1) monoclonal antibody. *Cell* 1997, 91:799-809.

For further discussion see slide 91.

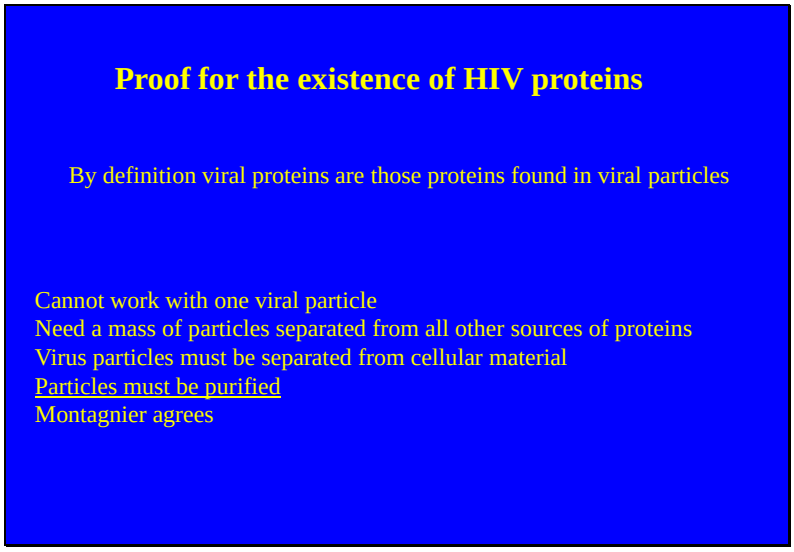
Slide 60



With that brief background to antibodies and antigens we continue by stating that in his 1983 paper Montagnier reported further experiments which led him to conclude that he had discovered a new retrovirus in BRU. He called this retrovirus lymphadenopathy associated virus, LAV, which nowadays is known as HIV.

Before considering Montagnier's additional experiments let us consider for ourselves the matter of characterising the proteins of a new virus. How should this be done?

Slide 61



No one can debate that ownership of viral proteins, like the ownership of body parts, is defined by whose body the parts come from. In the case of viral proteins the "body" can only be the viral particle.

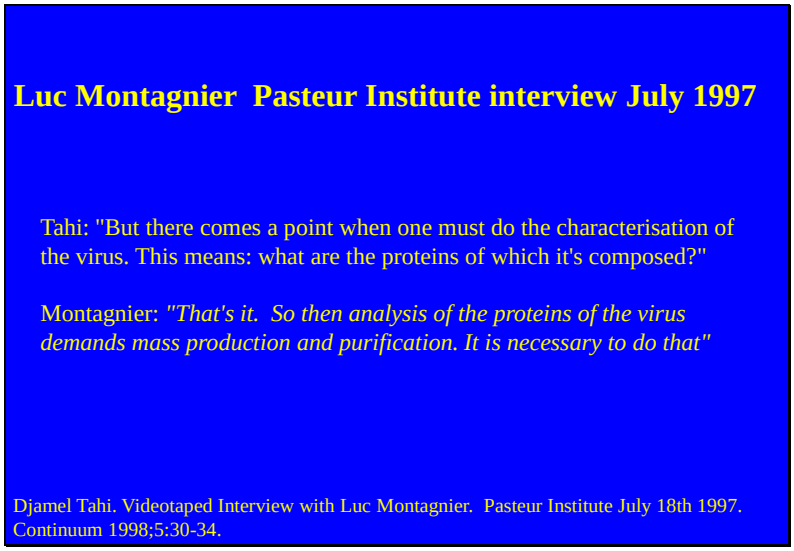
If a scientist wants to analyse the proteins of apples he goes to an orchard and picks objects which are or can be proven to be apples. He does not pick oranges or pears.
If the orchard has already been picked and sorted he obtains a box of apples. Not a box of oranges or pears.
If the orchard has already been picked but not sorted first he has to sort it. He walks around the packing shed with a box selecting apples. He does not select oranges or pears.
In all cases the scientist ends up with apples separate from everything else that is not apples. No oranges and no pears.

Unlike apples where it is feasible to obtain just one apple to perform a protein analysis, retrovirus particles are too small to obtain just one particle. Even if it were possible one particle does not contain a measurable amount of protein. So a scientist needs to obtain a mass of particles, analogous to a box of apples, to work with.

If a scientist wishes to analyse proteins of a retrovirus his first task is to obtain a mass of virus particles separated/isolated from everything else that is not virus particles. In other words, first the scientist must purify the virus particles or, in terms of the definition given earlier, he must isolate them. This means separating the particles from everything else that contains proteins including the cellular material.

The need for purification is a view shared by Montagnier.

Slide 62



In his 1997 Pasteur Institute interview Montagnier was asked:

"But there comes a point when one must do the characterisation of the virus. This means: what are the proteins of which it's composed?"

Montagnier replied "…analysis of the proteins of the virus demands mass production and purification. It is necessary to do that".

Both Montagnier in 1983 and Gallo in 1984 claimed to have purified "HIV" by using the well known method of retroviral purification called banding in density gradients. However, neither scientist published evidence to prove what they called "purified virus" contained any particles of any description, viral-like, non-viral-like or of any other description. Their notion of "purified virus" was built on the presumption that virus particles were present because in the material they called "purified" virus they detected reverse transcriptase activity.

In the Pasteur Institute interview Montagnier was also asked if he purified HIV. Despite the claim of "purified" virus in his 1983 paper he replied "I repeat we did not purify". When asked if Gallo purified he answered "I don't know if he really purified. I don't believe so".

Since by his own admission Montagnier did not purify the particles he claimed were HIV Montagnier had no grounds for claiming he had identified certain proteins as HIV and thus a new virus.

Despite the misgivings expressed in Montagnier's interview, in 1984 Gallo and his colleagues did claim to have purified HIV but, like Montagnier, had no electron micrographic proof for the presence or purity of particles in the "purified" specimen.

In September 2003 we emailed Dr. Gallo and asked him if he was aware of Professor Montagnier's Pasteur Institute interview. We particularly wanted to know what Dr. Gallo thought about Montagnier's lack of retroviral-like particles and purification and how it is possible to distinguish between retroviral and cellular proteins and nucleic acids without purification.

Gallo's response was one sentence: "Montagnier subsequently published many EM pictures of purified HIV particles, as, of course, we did in our first papers*. You have no need of worry. The evidence is obvious and overwhelming".

*Gallo's "first papers" and none since have electron micrographs of "purified" virus.

EXTRA
Like Montagnier, the Prosecution expert witnesses accepted the only way to obtain the viral proteins is to purify the viral particles.

Professor David Cooper:
"Once the virus is purified, it's then genetically sequenced…." (T673)
“Once that virus is purified, it's then genetically sequenced and those sequences are unique, just like every organism on the planet has unique sequences and markers". (T673)

Professor David Gordon:
"I'm not sure he did or didn't [if Montagnier purified]. I mean it's highly likely that he attempted to separate out the virus to purify the virus because purification of virus is then very useful for further studies for the nature of the virus and the nature of the immune response against the virus". (T1032)

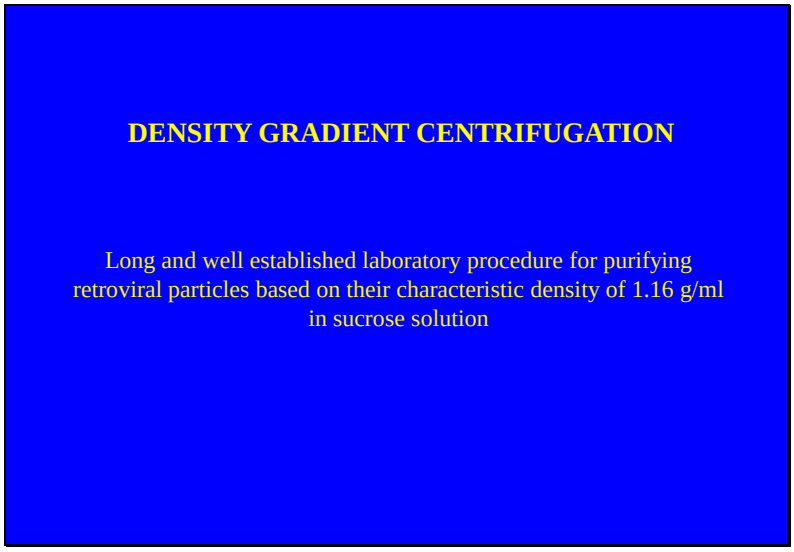
"It's a natural step from obtaining the virus in cell culture to then obtain purified virus". (T1034)

Professor Dominic Dwyer:
"If they want to go on and do further studies with the virus, yes like everybody else they [Montagnier and his colleagues] would be purifying large amounts of virus and extracting protein and genetic material, doing the analyses and so on…The purification, as far as one can go, is important in analysis of any virus or bacteria, for that matter as well". (T1199)…So when a new virus emerges, like SARS, you can't necessarily use, reliably, nucleic acid testing until you get the sequence of that new virus for the first time. So then in fact you are in a first identifier, you are required to use these more traditional methods of virus culture and microscopy and so on", that is, purification. (T963)

Robert Gallo:
"You have to purify". (T1257)

See http://www.theperthgroup.com/LATEST/PGRevisitHIVExistence.pdf

Slide 63

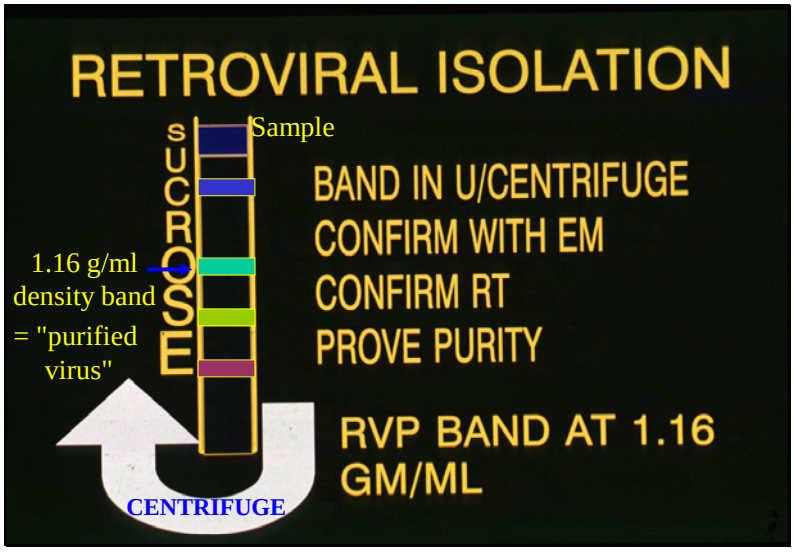


How then are retrovirus particles purified?

They are purified using a laboratory procedure which has been used for over 40 years. This is called density gradient ultracentrifugation, often shortened to density gradient centrifugation, and it is based on the fact that retroviral particles have unique density, 1.16 g/ml.

This procedure is applied to the culture supernatant fluid because, if the culture contains retrovirus infected cells, the viral particles will be released from the cells into the culture fluids.

Slide 64



This is how the actual purification/isolation procedure is performed:

Sucrose, that is, table sugar, is dissolved in water. This produces a solution more dense than water. That is, it has more mass than the same volume of water to which sugar has not been added. When sugar is dissolved in water in one's kitchen the density is uniform throughout the solution. But there is a way of filling a test tube with a sugar solution such that the density of the solution gradually increases from top to bottom, thus creating a "density gradient". In the slide this is indicated by the increasing size of the letters of "sucrose" as one progresses down the tube. Somewhere in the solution the density will be 1.16 g/ml, that is, the density of retroviral particles.

To do the purification a sample of the cell culture supernatant is placed at the top of the sugar solution. The tube is then spun at extraordinarily high speeds in a specially designed centrifuge. The spinning goes on for many hours and the force that is generated by the spinning gradually forces the particulate matter further and further towards the bottom of the test tube. When material arrives at a place in the solution where its density is the same as the solution it will travel no further. This means that material of

similar density aggregates or is concentrated in the form of a "band" of material at a particular distance from where the sample was originally placed. In the case of retrovirus particles this is where the density of the sugar solution is 1.16 g/ml. Other particulate matter of a different density will band at a different place in the gradient, that is, at another distance from the top of the solution. This is illustrated in the slide by the three bands at densities that are not 1.16 g/ml. At the end of the procedure the centrifuge is stopped, the tube is removed and a hole pierced in the bottom. Then the fluid is released in several, separate, aliquots where each density band can be analysed.

Note this technique separates material only according to differing densities. All cell culture supernatants contain cellular material and if this includes particulate matter whose density is 1.16g/ml, density gradient centrifugation cannot separate this matter from retroviral particles.

Analysis of the density gradient bands must include electron microscopy to prove (a) retroviral particles are present and (b) they are pure. Seeing is believing. If electron microscopy is not undertaken the scientist is flying blind. Only be using electron microscopy can the scientist know what kind of particles, cellular, viral-like, pure or impure are present. Or indeed that any particles of any kind are present.

EXTRA
In the 1960s density gradient centrifugation was introduced to separate and isolate sub‑cellular particles including viruses. Because some cellular constituents were found to have the same buoyant density as viruses, when viruses were isolated from cell cultures, the best results could be obtained with supernatant fluids which had high viral concentration and low cellular contaminants. This was best satisfied by non‑cytopathic (cell killing resulting in lysis) viruses and by culture conditions which maintained maximum cellular viability. All retroviruses isolated prior to HIV satisfy the above conditions. Taking advantage of the above retroviral properties, by repeated suspensions and sedimentation in sucrose density gradients, one could obtain, at a density of 1.16 g/ml, a relatively pure concentration of retroviral particles, that is, obtain retroviral particles separate from everything else, and thus isolate them [3]. Nonetheless, as many eminent retrovirologists pointed out, contamination of the viral preparation with particles which contain RT, but could be nothing more than "cellular

fragments", microsomes from disrupted cells, "membraneous vesicles which may enclose other cellular constituents including nucleic acids", especially when "inadvertent lysis of cells" was induced, could not be avoided [1,2,3,4]. Because of this, to prove that the material which banded at 1.16 g/ml contained nothing else but particles with "No apparent differences in physical appearances", and that the particles were indeed retroviruses, every retrovirus preparation was further analysed using the following assays:

(a) physical--EM for virus count, morphology and purity;

(b) biochemical--RT activity, viral and cellular RNA, total protein, gel analyses of viral and host proteins and nucleic acids;

(c) biological--infectivity in vivo and in vitro [3,5].

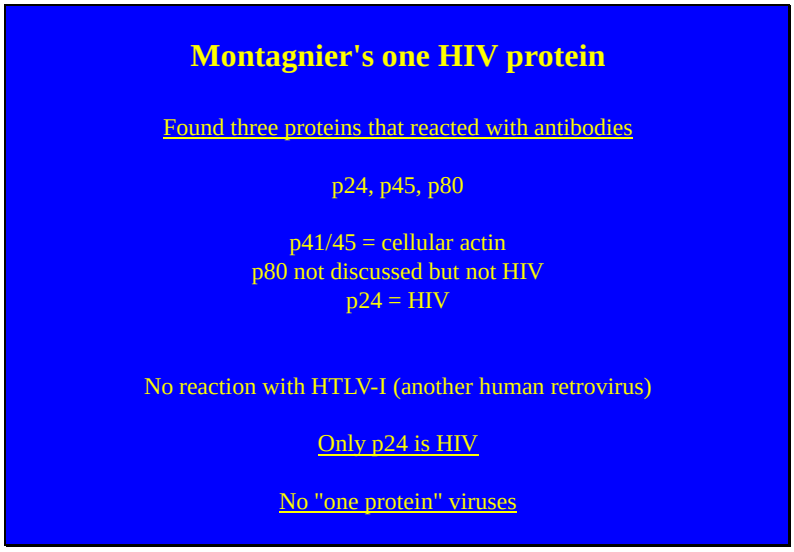
In other words, the first step in the effort of isolation of a retrovirus is the demonstration that:

1. The particles seen in the cultures band at 1.16 g/ml;
2. In the 1.16 g/ml band there is little present but the particles;
3. "No apparent differences in physical appearances" between particles are seen.

1. Weiss R,Teich N, Varmus H, Coffin J. RNA Tumor Viruses. Cold Spring Harbor Laboratory. Cold Spring Harbor, New York;1982.
2. Temin HM, Baltimore D. RNA-Directed DNA Synthesis and RNA Tumor Viruses. Adv Vir Res 1972; 17:129-186.
3. Toplin I. Tumor Virus Purification using Zonal Rotors. Spectra 1973;No. 4:225-235.
4. Bader JP. Reproduction of RNA Tumor Viruses. In: Fraenkel-Conrat H, Wagner RR, eds. Comprehensive Virology Vol.4. New York: Plenum Press, 1975:253-331.
5. Sinoussi F, Mendiola L, Chermann, JC, et al. Purification and partial differentiation of the particles of murine sarcoma virus (M. MSV) according to their sedimentation rates in sucrose density gradients. Spectra 1973;No. 4:237-243.

An mpg demonstrating the principles of density gradient centrifugation will be posted at the Perth Group website.

Slide 65



Montagnier took the culture supernatant fluid from the umbilical cord lymphocyte culture and banded it in a sucrose density gradient. He claimed that the 1.16g/ml band was the "purified virus" which originated from BRU. However, he did not publish any electron micrographs to show that the 1.16 g/ml band contained any retroviral-like particles or indeed particles of any kind, purified or unpurified.

Then Montagnier took the "purified virus" and separated the proteins that were present in that material using an electric field. To the separated proteins he added a sample of BRU's serum and another serum obtained from a second patient who had enlarged lymph nodes, and who "had been in close contact with an AIDS case" and was assumed infected with the new retrovirus. Three proteins, p24, p45, p80, in the "purified BRU virus", were found to react with BRU's serum.

Of these three proteins, which reacted with antibodies present in the BRU serum, Montagnier claimed the p45 protein was the ubiquitous cellular protein actin. "The 45K protein may be due to contamination of the virus by cellular actin which was present in immunoprecipitates of all the cell extracts". (Nowadays, without any further evidence, this p45 protein is

known as p41 and is claimed to be an HIV protein. The method used to determine the molecular weight is not very precise).

The p80 protein was not further mentioned but was not claimed to be an HIV protein. However, in the 27th July issue of *Science* 1984 Montagnier wrote "The 43-kD [p41/45] band and the 84-kD band are cellular contaminants that are immunoprecipitated in all the tested sera" [1].

The third protein which banded at the 1.16 g/ml (the "purified virus") and reacted with antibodies present in the BRU serum was a p25 protein (now known as p24). Unlike p80 and p45, the p24 protein was claimed to be an HIV protein and the antibodies in the BRU serum which reacted with it antibodies induced by HIV infection.

Since:
1. Montagnier and did not publish any of electron micrographs to prove the "purified virus" contained only virus–like particles, pure or impure;
2. Cellular fragments (debris, microvesicles) contain proteins, including at least one of molecular weight 24,000 (p24), one of the light chains of the ubiquitous cellular protein myosin;
3. AIDS patients and those at risk contain auto-antibodies (antibodies that react with their own constituents including proteins including actin and myosin [2,3]

it is scientifically impossible for Montagnier to interpret his data in the manner he did. From a reaction between proteins present in the 1.16 g/ml sucrose density gradient and antibodies in patient sera, it is not possible to identify the origin of one reactant even when the other is known. Yet from such reactions, Montagnier and subsequently others including Gallo, claim to have proven that the proteins in the 1.16 g/ml band are "HIV" and the antibodies in the patient sera are antibodies directed against "HIV" which has infected the patients. We repeat, such conclusions are a scientific impossibility.

Montagnier also added serum containing antibodies to the p24 protein of HTLV-I and, since he found no reaction, concluded the virus he had discovered was not another retrovirus but a new virus.

Hence Montagnier concluded BRU was infected with a new retrovirus and this retrovirus contains one protein, a p24 protein.

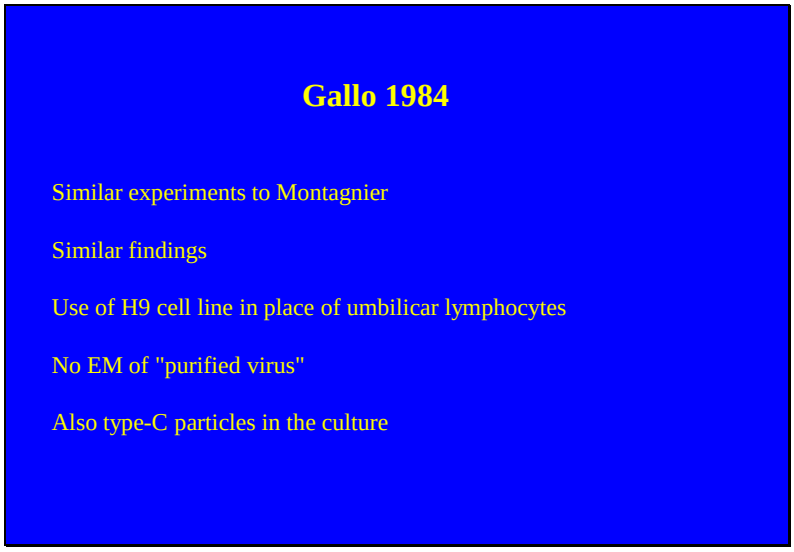
One should also comment that:

1. Since two of Montagnier's proteins were not "HIV" then his "purified virus" was not purified.
2. If p45 (p41) is actin and p80 was not viral then why is p24 also non-viral?
3. HIV is said to have about a dozen proteins. Why did Montagnier not identify all the proteins of the new virus?
4. The p24 protein is said to be a major core protein of HIV (see slide 31). Since viral reverse transcriptase is a protein, and it is not p24, then the new retrovirus could not have been the source of reverse transcription identified in Montagnier's "purified" virus.
5. There is no precedent for a "one protein" retrovirus. Retroviruses need several proteins in order to make a retroviral particle, replicate and produce their biological effects.

Yet today p24 is considered to be the most specific HIV protein. Biotechnology companies manufacture antibodies directed against this protein and, when they react with antigens in a cell culture, they are said to prove HIV isolation.

1. Brun-Vezinet F, Rouzioux C, Montagnier L, et al. Prevalence of antibodies to lymphadenopathy-associated retrovirus in African patients with AIDS. *Science* 1984; 226:453-456.
2. Calabrese LH. Autoimmune manifestations of human immunodeficiency virus (HIV) infection. *Clin Lab Med* 1988;8:269-279.
3. Matsiota P, Chamaret S, Montagnier L. Detection of Natural Autoantibodies in the serum of Anti-HIV Positive-Individuals. *Ann Inst Pasteur Immunol* 1987;138:223-233.

Slide 66



In 1984 Gallo performed similar experiments to Montagnier but with two main differences.

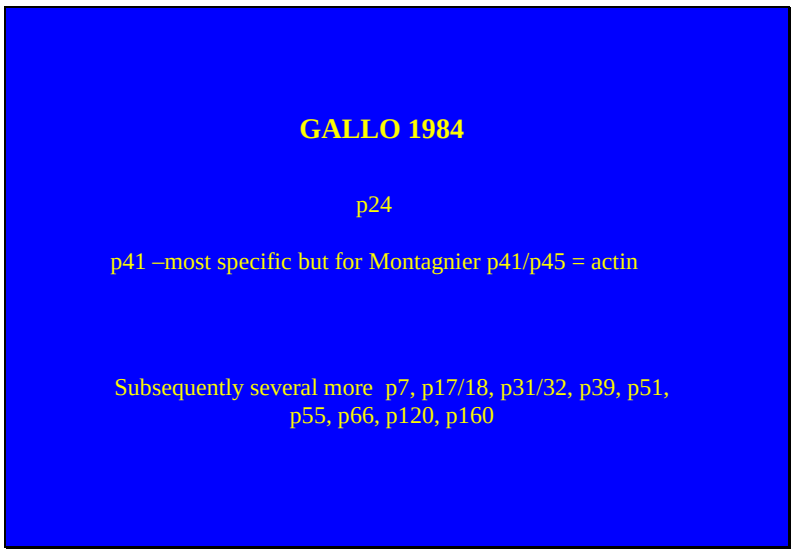
The first was that instead of using umbilical cord lymphocytes to obtain "purified virus", Gallo used a leukaemic cell line known as H9. Using a leukaemic cell line creates many problems, as Gallo himself knew. This is because leukaemic cell lines, even when not infected with retroviruses, produce virus-like particles. And later on, after the investigation into Gallo's scientific practices, it was found that the H9 leukaemic cell line originated from a patient who had a type of leukaemia Gallo claimed is caused by HTLV-I. In fact, in 1983 he published a paper in which he reported that the parental cell line of H9 contained "HTLV[I] proviral [DNA] sequences" [1]. This means this cell line is infected with a retrovirus even if not cultured with tissue from AIDS patients.

Like Montagnier, Gallo did not publish an electron micrograph of what he termed "purified virus", that is, the material which in sucrose density gradients banded at 1.16 g/ml.

We also note that the particles he reported in the unpurified culture material were type-C particles, the same taxonomic group that Montagnier reported and which are not Lentiviruses.

1. Wong-Staal F, Hahn B, Manzuri V, et al. A survey of human leukemias for sequences of a human retrovirus. Nature 1983; 302:626-628.

Slide 67



The second major difference is that unlike Montagnier, Gallo tested more than one AIDS patient. He reported that some of the sera reacted only with p24, others only with p41 and yet others with both. In regard to the p24 protein, Gallo said it is not specific to HIV because this protein also reacts, that is, cross-reacts, with antibodies to HTLV-I.

Thus, what Montagnier considered to be the most specific (and only) HIV protein, Gallo considered not to be HIV specific. And the protein that Montagnier considered cellular and not HIV, p41/45, Gallo considered to be the most specific for HIV.

These are diametrically opposite opinions impossible to reconcile.

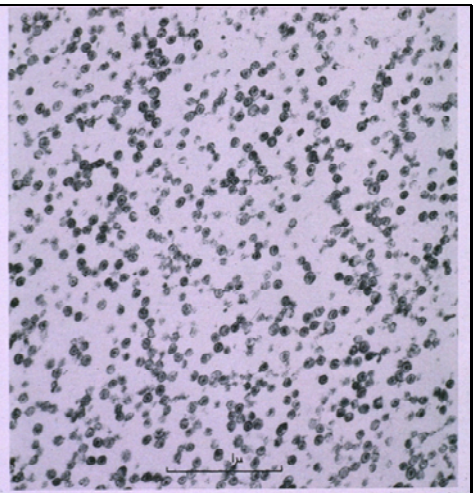
Gallo also reported that proteins apart from p24 and p41/45 also reacted with some of the AIDS patients' sera. Because of this he claimed a number of other proteins, p7, p17/18, p31/32, p39, p51, p55, p66, p120 and p160, were also HIV proteins.

Hence these proteins became HIV proteins despite their being no evidence that the "purified virus" contained particles of any kind, retroviral or non-retroviral, pure or impure. Exactly as it was with Montagnier's experiments.

Nowadays, on the same basis, that is, reactivity with antibodies in AIDS patient sera, HIV is said to have approximately a dozen proteins.

Slide 68

Electron micrograph (EM) of a density gradient purified retrovirus



Crawford LV *et al*. The properties of Rous sarcoma virus purified by density gradient centrifugation. Virology 1961; 13:227-232

FIG. 1. Electron micrograph of preparation of Rous sarcoma virus. Fixed in potassium permanganate, sectioned and the section stained in uranyl acetate. The electron micrograph was taken with a Siemens Elmiscop II at an instrumental magnification of 15,000 times. Actual magnification of figure: × 30,000.

We now wish to address several matters in regard to purification of the "HIV" particles.

This slide was published in 1961 and confirms it is possible to take an electron micrograph of the 1.16 g/ml band and show purified, retroviral particles. These are particles of the Rous sarcoma virus, an animal retrovirus discovered by Rous in 1911. Notice that unlike many images of HIV, this EM has a size bar measuring 1000 nanometres.

For over a decade we asked for similar evidence in regard to HIV. That is, evidence that what Montagnier and Gallo called "purified" virus, consists of retroviral particles and nothing but retroviral particles. Nobody responded.

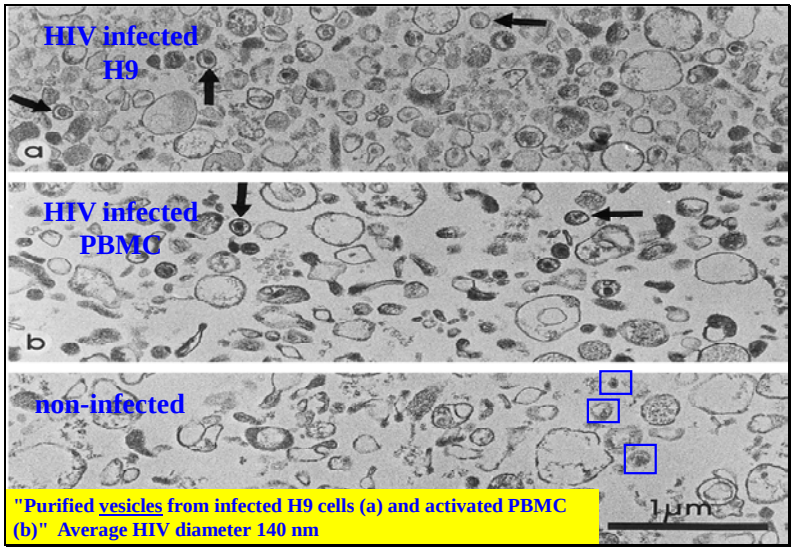
However, in 1997, fourteen years after the alleged discovery of HIV, two groups of researchers, one a Franco/German collaboration and another from the US National Cancer Institute, published papers in the March issue of the journal *Virology.*

Slide 69

These researchers accepted that up till 1997, the "HIV" "used for biochemical and serological analyses or as an immunogen [antigen] is frequently prepared by centrifugation through sucrose gradients", and that in none of the studies "has the purity of the virus preparation been verified".

In other words, for fourteen years the community of HIV experts claimed to have obtained purified HIV, and then used this material to obtain proteins and RNA as if it were unique to a retrovirus HIV, and employed it time and time again for research and producing and patenting various diagnostic tests. All without a shred of proof it contained even one particle of any description let alone a retroviral-like particle.

Slide 70



Here is the first, published electron micrograph, from the Franco/German group*, to show the actual composition of this material, what all HIV experts claim is "purified HIV".

The upper part labeled (a) is the 1.16 g/ml band obtained from cultures of "infected cells", that is, H9 cells "infected" with HIV. This middle part, labeled (b), is again the 1.16 g/ml band, again from "infected" cells, but this time the cells "infected" with HIV are peripheral blood mononuclear cells [PBMC=white blood cells], obtained from normal individuals.

The bottom part represents the 1.16 g/ml band obtained from non-infected cell cultures, that is, cells to which no "HIV" has been added.

Any man, woman and child devoid of any scientific training whatsoever can see that whatever the material in these pictures represent, it is not pure. In something that is purified every object looks the same as every other object. Just think of a box of apples. In fact the authors of this paper labeled this EM as:

"FIG. 2. Purified HIV-1 preparations are contaminated by cellular vesicles. Purified vesicles from infected H9 cells (a) and activated PBMC (b) supernatants (fraction 6 from Fig. 1a and fraction 5 from Fig. 1b, respectively) or from noninfected H9 cells (c) (fraction 6 from Fig. 1e) were treated for electron microscopic analysis as indicated under Materials and Methods. The cellular vesicles appear to be a heterogeneous population of both electron-lucent and electron-dense membrane delineated vesicles ranging in size from about 50 to 500 nm. (Original magnification 136,000). Virions are indicated by arrows".

The statement "Purified HIV-1 preparations are contaminated by cellular vesicles" is an oxymoron. Notwithstanding, in the next sentence the authors then opt to describe their EM not as "purified HIV" but as "Purified vesicles" obtained either from (a) H9 "infected" cell cultures or (b) PBMC "infected" cell cultures.

In other words, after fourteen years HIV experts themselves confirm there is no such thing as purified HIV. From the "infected" cultures they were able to obtain "Purified vesicles", that is cellular material but not HIV and such cellular material contains many proteins and RNA.

If we examine the structures which make up the pictures in more detail what do we see? Judging by the number of arrows, which point to particulate matter the authors claim is "HIV", nearly all the material consists of cellular fragments. However, in the bottom section, which contains no "HIV" particles, has at least three particles, which we have outlined in blue, which look similar to those the authors consider as "HIV" in the "infected" culture material.

Looking at the sections (a) and (b) it is difficult to decide exactly what criteria the authors used to classify particles as HIV. And whatever these criteria may have been, the particles do not have all the morphological characteristics of retroviral particles.

One of the first and unambiguous observations one can make about the arrowed particles is their diameter. We have measured all these particles with a micrometer and their average diameter is 136 nM and no particle has a diameter less than 120 nM. Hence these particles are too large to fit the definition of a retroviral particle. Recall that the retroviral diameter is 100-120 nanometres**.

Another matter is that the chemical agents used in the cultures depicted in sections (a) and (b). The fact is that these chemicals, which include the mitogenic agent PHA, were not used in preparation of the cultures in the bottom section, that is, the "controls". This means that these cultures were all not treated in an identical manner apart from the addition of "HIV". Hence the bottom section is an invalid control. Given that retroviral particles can appear even spontaneously, or be induced by culture conditions, because the authors did not add these chemicals, including PHA, the possibility cannot be excluded that the appearances in the bottom section may have otherwise turned out identical to sections (a) and (b).

We must also keep in mind that these studies were not blind and authors always publish their best evidence, including electron micrographs, to support their claims.

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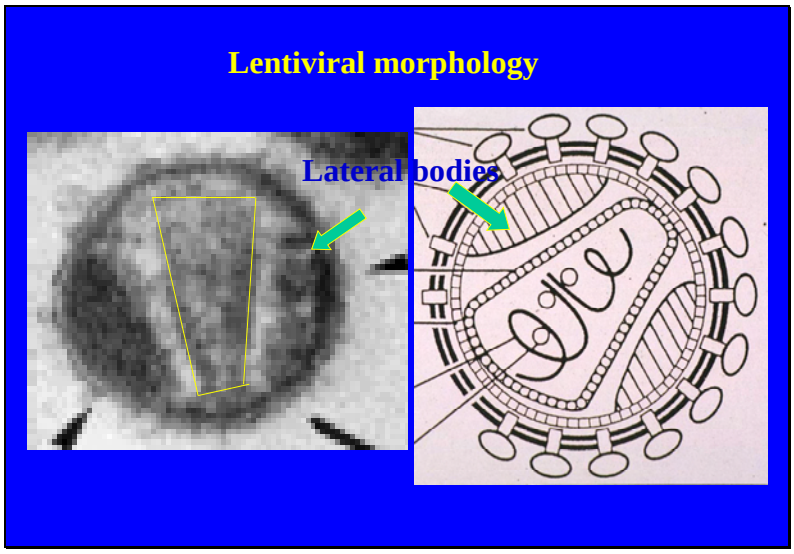
Centre d'Immunologie de Marseille-Luminy, Case 906, 13288 Marseille, France; and †Robert Koch Institute, Nordufer 20 D13353, Berlin, Germany

** In the revised 2000 taxonomy of retroviruses, under the heading "Morphology", "Virions are spherical, enveloped and 80–100 nm in diameter". Hence the diameter of the Franco/German "HIV" particles is even further removed from the defining diameter.

See http://www.virustaxonomyonline.com/virtax/lpext.dll/vtax/agp-0013/rtr03/rtr03-sec1-0001?f=templates&fn=document-frame.htm&2.0#rtr03-sec1-0001

Slide 71



According to the HIV experts, HIV is classified as a Lentivirus and some of the main characteristics of a Lentivirus, as we can see in the picture on the left, is the cone shaped core, knobs on the surface (allegedly present where the black arrowheads are pointing) and the "lateral bodies". On the right is Gelderblom's drawing of HIV.

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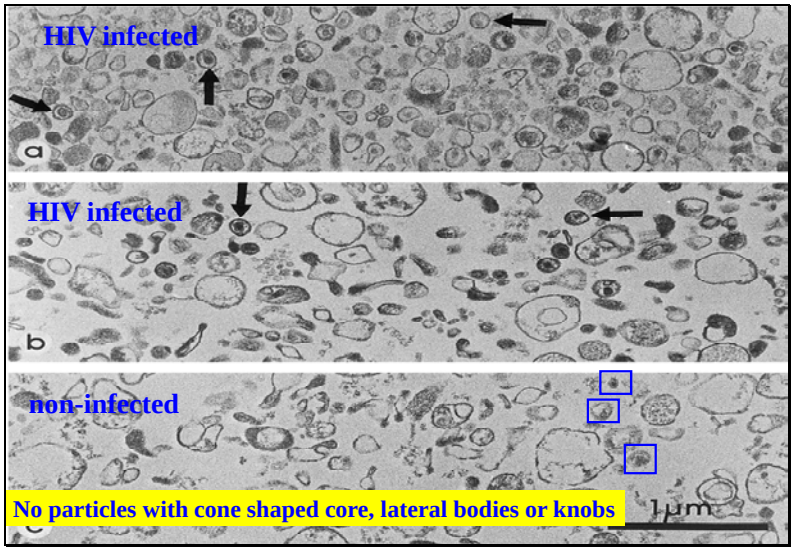

Yet none of the particles illustrated in the Franco-German study have knobs, or a cone-shaped core, or the lateral bodies. Which means the Franco/German particle cannot be HIV.

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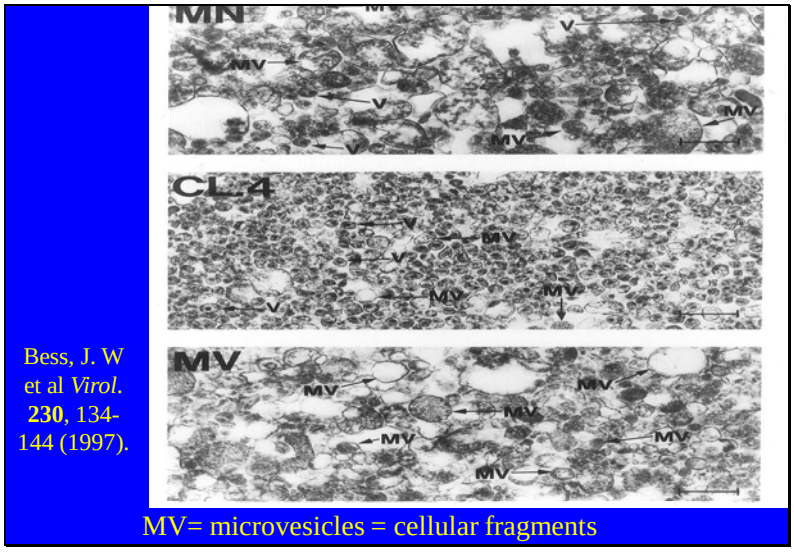

Next we look at the second paper published in the March 1997 issue of *Virology* by Julian Bess and his colleagues from America.*

This electron micrograph, also in three sections, represents their efforts to purify HIV.

The top section labeled MN is density gradient material obtained from "infected" H9 cells. The middle section labeled CL4 is an "infected" clone of H9 cells called CL4. This culture originated from another culture which was drastically manipulated, including being co-cultured with cells which had been heavily irradiated. The bottom section labeled MV again represents a density gradient band from a non-infected culture. This is the “control” but it was not irradiated and this is no small matter because radiation can also cause the appearance of retroviral-like particles [1]. (For an unknown reason Bess and his colleagues called the "control" culture "mock virus", (MV), although it is not infected and is not a “virus”).

Again, as anyone even with poor eyesight can see, nothing is purified. In fact it is difficult to discern any differences between the three sections,

although CL4 appears to have more particles bearing some but not all the morphological characteristics of retroviruses.

What is labeled MV are microvesicles, that is, cellular structures. As can be seen most of the material is made up of microvesicles and other cellular material.

EXTRA

*Affiliations of the American authors
AIDS Vaccine Program, SAIC, National Cancer Institute-Frederick Cancer Research and Development Center, Frederick, Maryland 21702-1201

1. Weiss RA. Why cell biologists should be aware of genetically transmitted viruses. National Cancer Institute monograph 1978:183-9.

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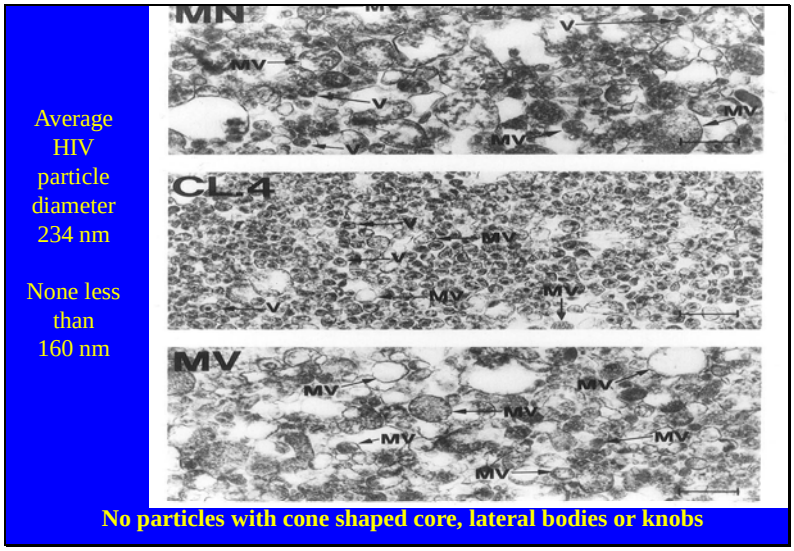

There are a few objects labeled V (for virus) which the authors claim are HIV. In this study their average diameter is 234 nm with no particle having a diameter less than 160 nm. So again it is impossible for these to be a retrovirus.

On this point we corresponded with Dr. Bess via email. He agreed the particles are of this size but he could not provide an explanation as to why. He said he would consult with his electron microscopers but never got back to us on this point.

Again, there is no evidence that these particles have cone shaped cores, or lateral bodies, or knobs. Three more reasons why they cannot be a Lentivirus or a retrovirus or “HIV”.

If the particles labelled "HIV" are indeed "HIV", then the density gradient band containing them should have extra proteins not present in the band obtained from the non-infected cells. The answer to this was provided by Julian Bess and his colleagues.

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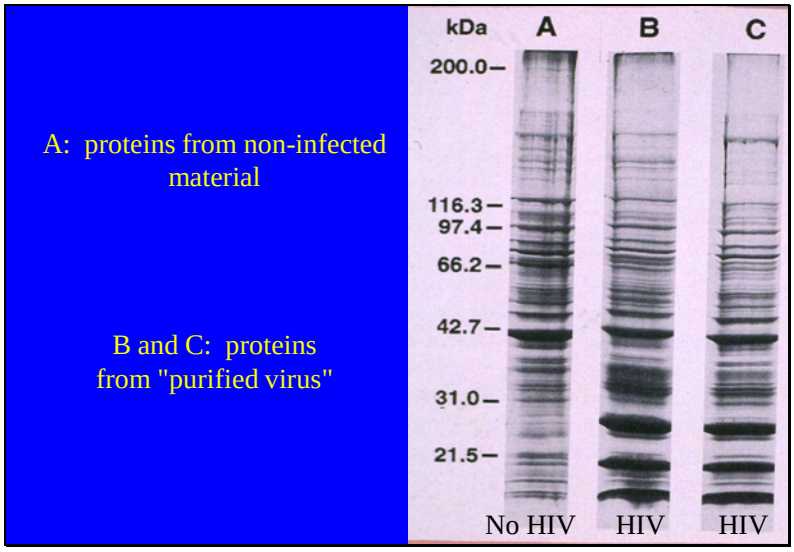

Note: Labels "No HIV" and "HIV" – added by us.

Bess and his colleagues did an experiment not previously reported. They took the mixture of proteins present in each of the three sucrose density bands obtained from the "purified virus", that is, MN, CL4, as well as MV, the uninfected "mock virus". Each of these protein mixtures was put into a polyacrylamide gel. The gels were then subjected to an electric field in order to separate the proteins from one another. This is a standard laboratory technique known as electrophoresis. In this procedure about a 100 volts positive is applied at one end of the gel and because proteins carry a negative charge they begin to move through the gel towards the positive charge. The higher molecular weight proteins move slowly and least while the lightest proteins move faster and farthest. After several hours the proteins become separated according to their molecular weights and charge. Then the gels are stained with a protein specific stain which shows the individual proteins as a series of black, horizontal lines. Note that the thickness or darkness of a band is determined by how much protein is present in that band. The proteins are identified by marker proteins of known molecular weights which are run in a parallel gel (and which can be seen on the left side of the slide). (Take care not to confuse

electrophoretic bands, which represent individual proteins in a gel, with density gradient bands*. The same word is used for two completely different things).

Lanes A, B and C are the proteins obtained from the uninfected, the "infected" H9 and the "infected" CL4 cell culture density gradient bands respectively.

Our interpretation of this electrophoresis experiment is that there is no difference between the three Lanes except that, in some parts, the darkness of the bands varies. In other words, in our view, the same proteins with the same molecular weights are present in all three density gradient banded specimens. "Infected" and non-infected. The only difference between all these bands is quantitative, not qualitative.

EXTRA

*During her cross-examination Professor Elizabeth Dax, Head of the Australian National Reference Laboratory, confused electrophoretic bands with density gradient bands.

Q. I want to put to you a passage from the evidence of Ms Papadopulos-Eleopulos of the HIV virus and just ask for your comment. p272, line 20, I have been asking her about the Gelderblom article we looked at this morning, which had pictures in there. 'Q. During your [EPE's] evidence you have told us many times that HIV has never been photographed. A. No, no, no, no. I never said that HIV has not been photographed...There are numerous photographs of what is called – what is meant to represent HIV particles from the cultures...electronmicrographs - what is meant to represent, apart from Bess and Gluschankof 1997 papers, there are no photographs of the banded material to show that what they are saying is pure HIV actually is pure HIV'. Do you have any idea what Ms Papadopulos-Eleopulos was talking about there.

A. It seems a little difficult to decipher because she's talking about – she would appear to be talking about photographing bands or the material that went to the bands, which is not really terribly sensible because, as I indicated this morning, once the virus is disrupted and run on the gels, there is no longer a virus to photograph. Perhaps she is referring to the material that is isolated and then put on the gels or on the test base, but I read that part earlier so I am familiar with the passage you are quoting but I don't -- it is nonsensical, I'm afraid" (T883).

In later testimony Mr. Borick sought clarification about the above answer from Professor Dax, in fact by reading her response back to her from the court transcript. Her second answer did not appear to resolve the confusion.

A. Yes. Afterwards I reflected on that. I expect what she [EPE] is talking about is gel bands, which are in the purification material, the ultracentrifugation of the gels. Is that correct?" (T1147).

Comment

Gels are not subjected to ultracentrifugation (centrifugation). In both instances Professor Dax was being asked about the lack of electron micrographic evidence of purification of particles in density gradient bands, as typified by the Glushankof and Bess papers. The question was not about electrophoresis of proteins in gels or antibody reactions with proteins in Western blot bands.

NOTE

Professor Dax is a recognised international expert in HIV testing and knows the protein antigens used in the Western blot test are obtained from the viral lysates, that is, the disrupted 1.16 g/ml density gradient material, the “purified” virus. In the book she co-authored on HIV testing [1] she wrote "the best antigen preparations to detect established HIV infection are viral lysates because these contain native antigens from virtually all structural components of the virus" (page 138). (Viral lysates = disrupted retroviral particles).

1. Constantine NT, Callahan JD, Watts DM. Retroviral testing: essentials for quality control and laboratory diagnosis. Boca Raton, Florida: CRC Press, 1992: 138.

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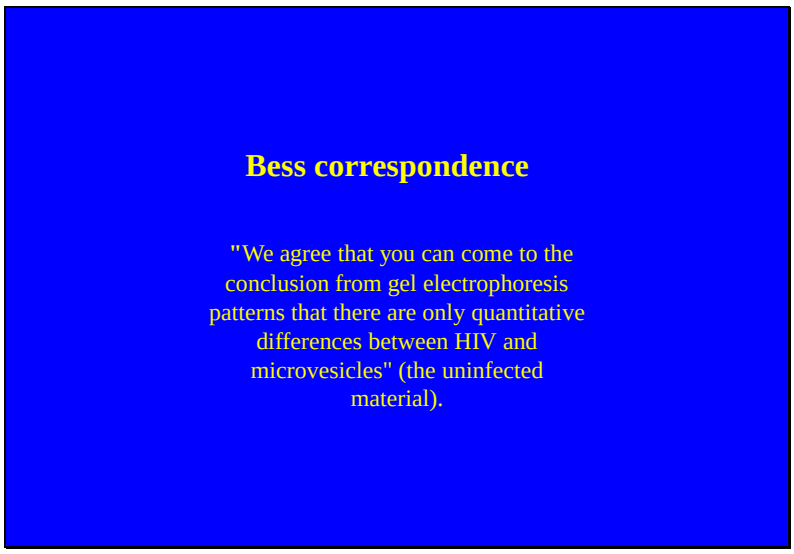

We emailed Dr. Bess our opinion that his electrophoretic patterns showed only quantitative differences and hence, as far as we are concerned, the same proteins were present in all the Lanes and hence in all the density gradient bands.

Dr. Bess replied "We agree that you can come to the conclusion from gel electrophoresis patterns that there are only quantitative differences between HIV and microvesicles".

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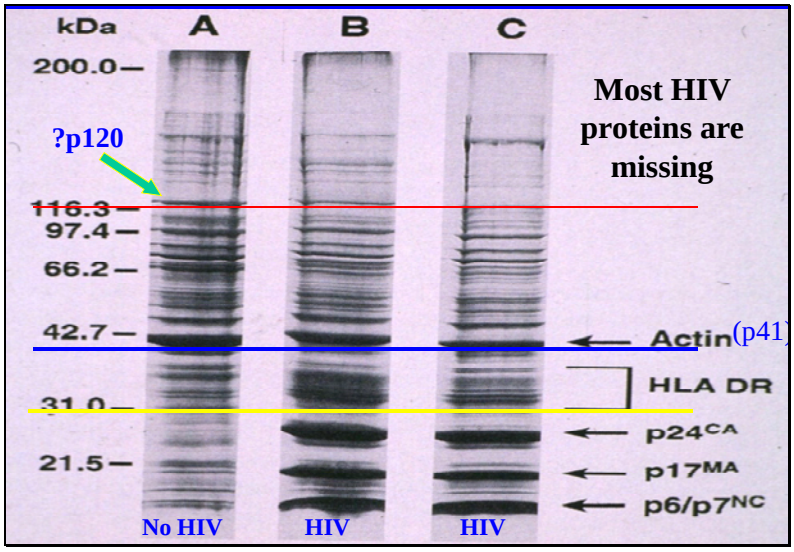

EXTRA
Note: Labels "No HIV", "HIV", "(p41)", "?p120", "Most HIV proteins are missing" and the three horizontal lines – added by us.

It is instructive to examine the Bess electrophoresis data in more detail.

1. Looking above the blue line the pattern is identical both qualitatively and quantitatively. These sections could be cut and pasted without anyone being able to tell the difference.
2. Between the blue and yellow lines Lane B has some quantitative differences.
3. Below the yellow line lanes B and C have thick, darker bands which are labelled by arrows p24CA, p17MA, or p6/p7NC. (These terms mean the same as p24, p17 and p6/p7). In Lane A these bands are present but are not as pronounced. Only if the corresponding area of Lane A was blank could one say these proteins are not present in Lane A.
4. If the same proteins are present in non-infected and "infected" material then the HIV proteins must be cellular proteins. The fact there are more of the same proteins in the "infected" specimens does

not prove they are HIV. The differences could easily be accounted by the different history of the cultured cells and the laboratory conditions under which they were cultured.

5. In the "HIV infected" material, Lanes B and C, there are no labelled "HIV" proteins above p24. Where are all the other "HIV" proteins? HIV is said to contain about 12 proteins which means 9 are missing.
6. In the region above the three labelled "HIV" proteins and below p42.7, one would expect to see the "HIV" proteins p32 and p41. These are absent.
7. In the region where one would expect to find p41 there is a protein, present in all Lanes, labelled actin, thus confirming Montagnier's claim that the "HIV" p41 protein is cellular actin.
8. Significantly missing from the "purified" virus is p120, the protein from which the knobs are made and which is crucial for infectivity. Which means that "purified" HIV is not infectious.
9. There is protein in the estimated 120 molecular weight region (116.3) just above the red line but this band is present in all Lanes. Which means that if this band is HIV p120 in Lanes B and C (but not labelled by the authors) then uninfected cells contain the HIV p120 protein.

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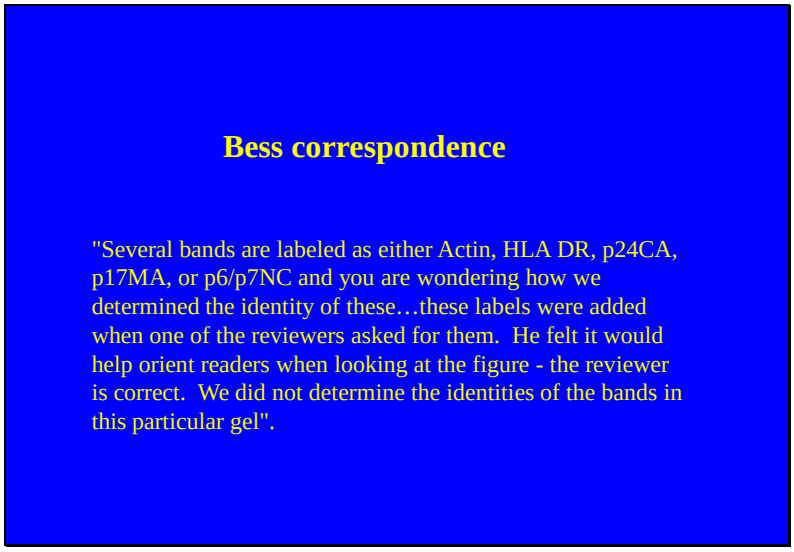

Since there are only three proteins labelled HIV one would expect proof these were HIV proteins. This is because a protein's molecular weight is not sufficient to identify a particular protein. Just as humans can share the same weight, so too can different proteins have the same molecular weight.

However, in correspondence Dr. Bess told us "Several bands are labeled as either Actin, HLA DR, p24CA, p17MA, or p6/p7NC and you are wondering how we determined the identity of these. First of all, these labels were added when one of the reviewers asked for them. He felt it would help orient readers when looking at the figure - the reviewer is correct. We did not determine the identities of the bands in this particular gel".

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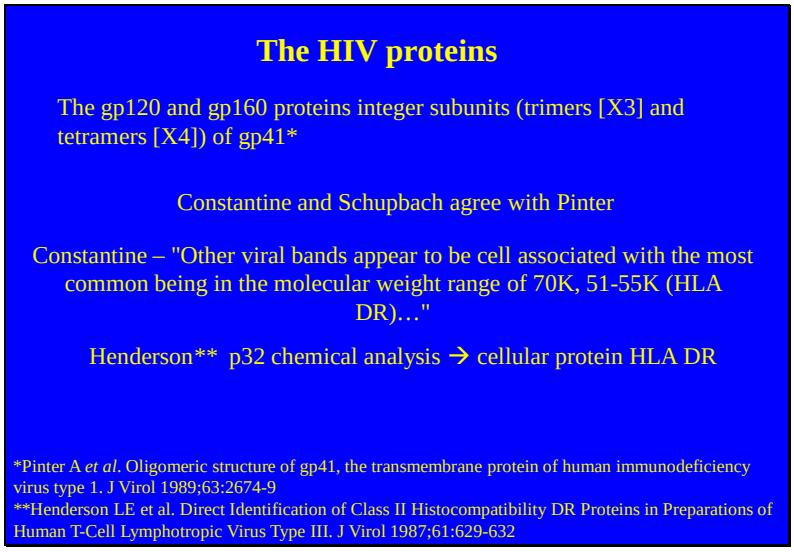

In 1989 Pinter *et al* published data that the p120 and p160 proteins are composed of three or four subunits of p41 respectively. And according to Montagnier, and Bess, p41 is the cellular protein actin. So p41, p120 and p160 are not three different proteins. They are the one protein packaged in three different ways. Constantine and his colleagues, and Schupbach, one of the main collaborators with Gallo from 1984, support this view. In other words, if p41 is actin, then p120 and p160 are also actin.

In 2000 Constantine and his colleagues also wrote "Other viral bands appear to be cell associated with the most common being in the molecular weight range of 70K, 51-55K".

As far back as 1987, Henderson, analysed the p32 HIV protein and proved it was a cellular protein known as HLA DR.

Hence several HIV experts accept that all the proteins with molecular weights higher than 24,000 are cellular proteins. They call them viral proteins but they are actually cellular proteins.

And it must also be mentioned that the lower molecular weight proteins, the p7 and p6 proteins, are fragments of proteins which have a molecular weight higher than 32,000, which again makes them cellular proteins.

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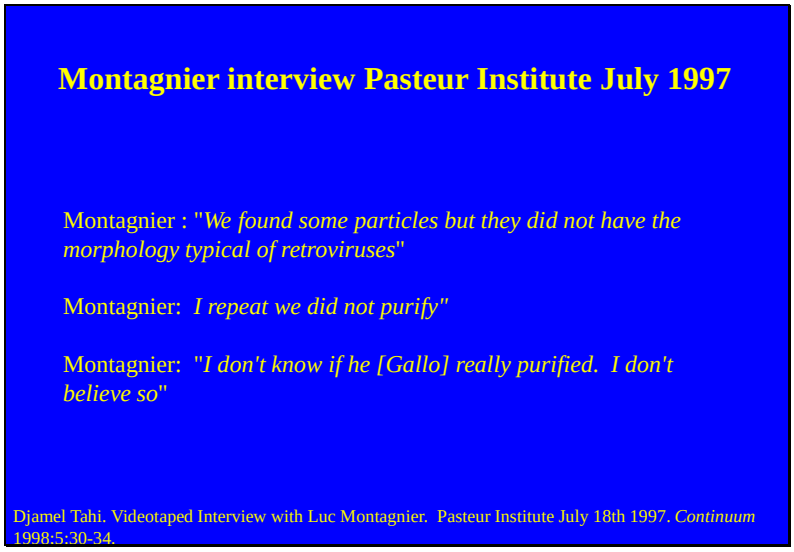

Since many HIV researchers argue that the proteins of molecular weight greater than 24,000 in the "purified" virus are cellular proteins, we are left with Montagnier's one, "HIV" protein, p24.

What evidence do we have that this is the protein of a unique, retrovirus? None.

In fact, we can dismiss Bess's data and see how Montagnier responded in the 1997 interview when asked about why he did not publish electron micrographs of his "purified" virus.

Montagnier's response was stunning. The reason, he said, was that despite "a Roman effort", they were unable to find any particles which looked like retroviruses. The quote from the interview transcript reads "We found some particles but they did not have the morphology typical of retroviruses". And he repeated "I repeat, we did not purify". And when he was asked if Gallo had managed to purify HIV he replied "I don't know if really purified. I don't believe so".

So we have now Montagnier finding a protein in material in which he did have even one retrovirus-like particle and merely because this protein reacted with antibodies whose identify was unknown he said the protein was HIV and the antibodies were HIV and BRU was infected with HIV.

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In his book *Virus* Montagnier wrote that researchers "...must have the mentality of a gambler or fisherman. As for me, I am only interested in big fish".

So let us compare Montagnier's finding with that of a fisherman casting his net. The difference is that a net catches fish according to size while Montagnier's net caught retroviruses according to their density.

Montagnier threw his net into his culture hoping to catch not only a fish but one particular type of fish, a never seen before fish. He pulled up his net and saw much material that occurs in the sea but no fish, not even a single fish. Unperturbed, he set about analysing the proteins in the non-fish material and claimed one of the proteins was from a fish. In fact a fish never seen before.

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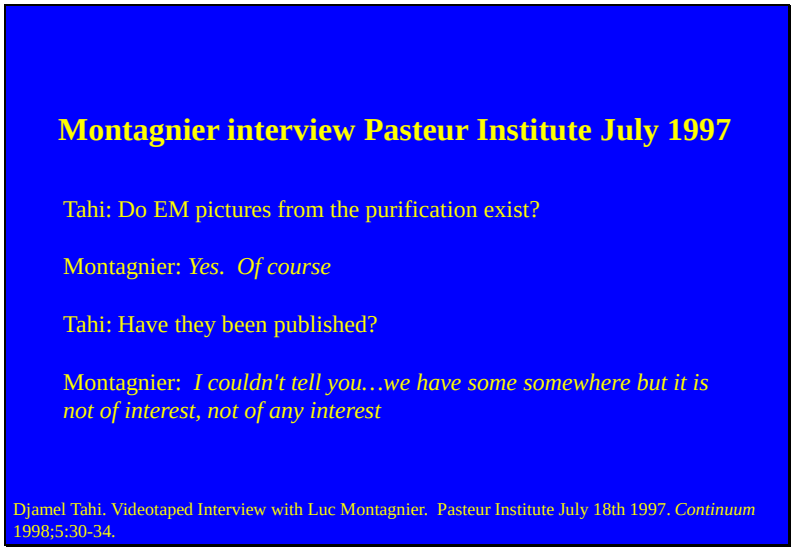

Continuing his interview, once Montagnier accepted that what he called "purified virus" did not contain one virus-like particle, Tahi asked

Tahi: Do EM pictures from the purification exist?

Montagnier: *Yes. Of course*

Tahi: Have they been published?

Montagnier: *I couldn't tell you…we have some somewhere but it is not of interest, not of any interest.*

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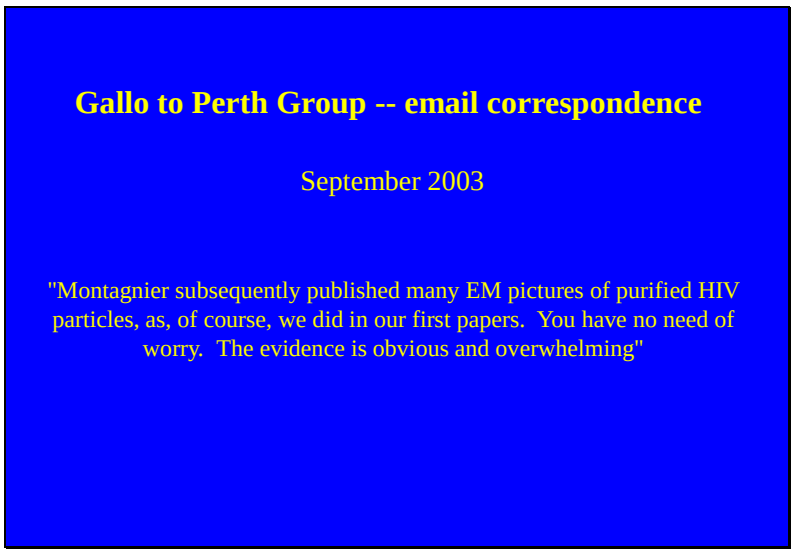

In September 2003 we emailed Gallo and asked if he was aware of the Tahi interview and Montagnier's response in regard to their being no EM of purified virus.

Gallo replied "Montagnier subsequently published pictures of purified HIV particles as, of course, we did in our first papers. You have no need of worry. The evidence is obvious overwhelming".

In fact, there was not one single picture of purified HIV published by Gallo in 1984 or at any time since. Neither did Montagnier publish any such pictures.

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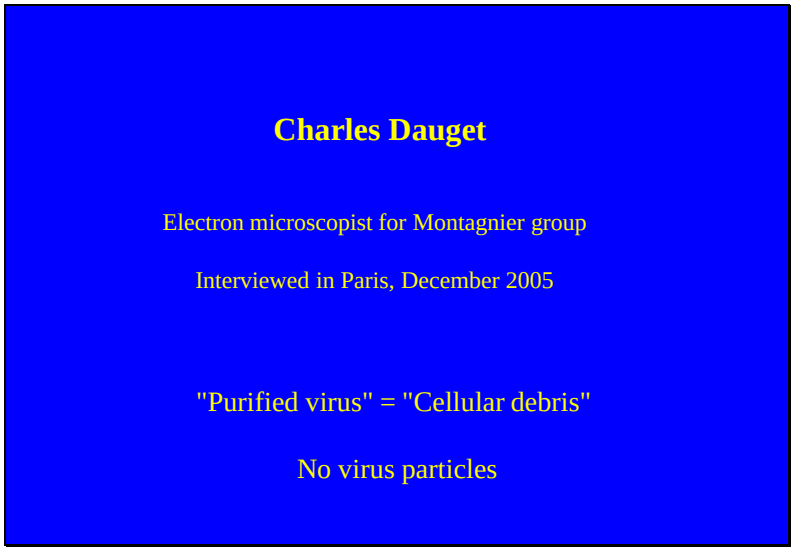

In December 2005, Djamel Tahi interviewed Charles Dauget. Dauget, now retired, was the Pasteur Institute electron microscopist and one of the co-authors of the 1983 Montagnier paper.

Like Montagnier Dauget was asked why no electron micrographs of purified HIV were published.

His response was "We have never seen virus particles in the purified virus. What we have seen all the time was cellular debris, no virus particles".

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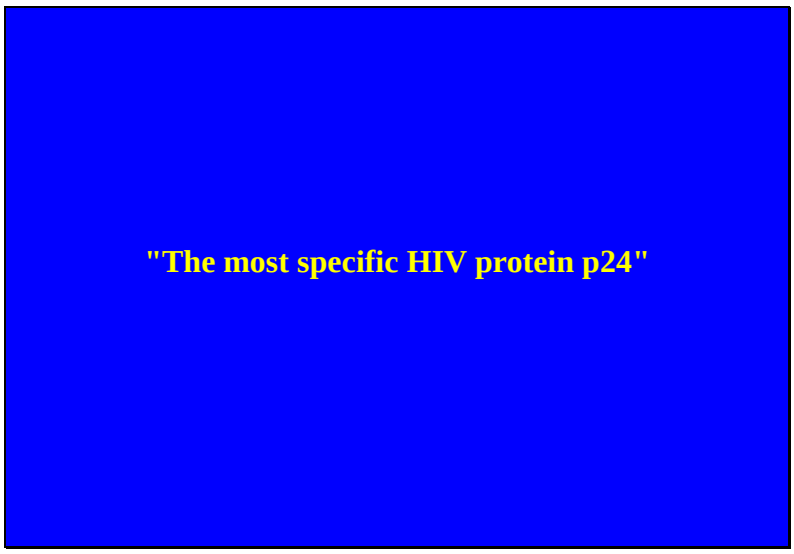

So we are left with the conclusion that the most specific HIV protein originated from material in which there were no retroviral particles. This is as good a scientific proof any scientist can have that this protein is nothing more than a cellular protein.

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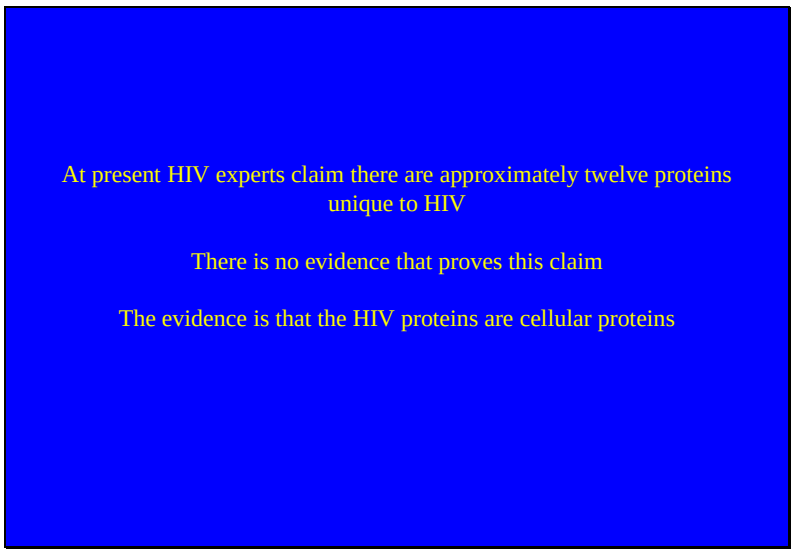

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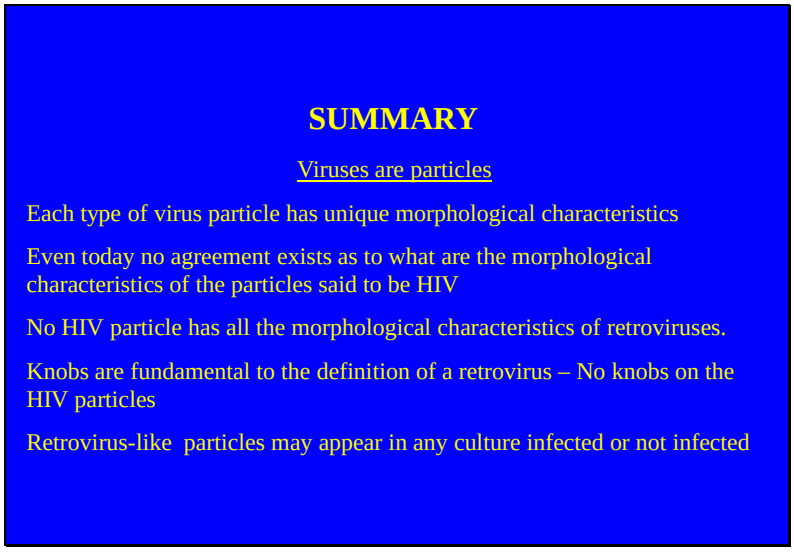

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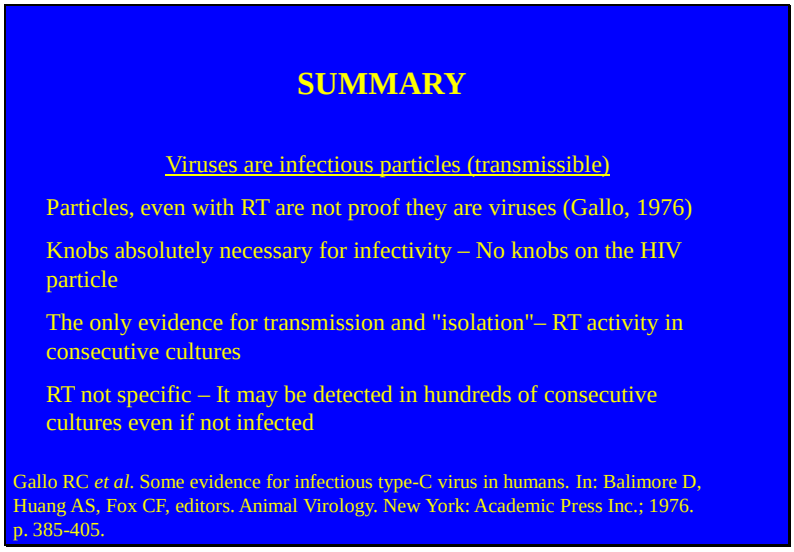

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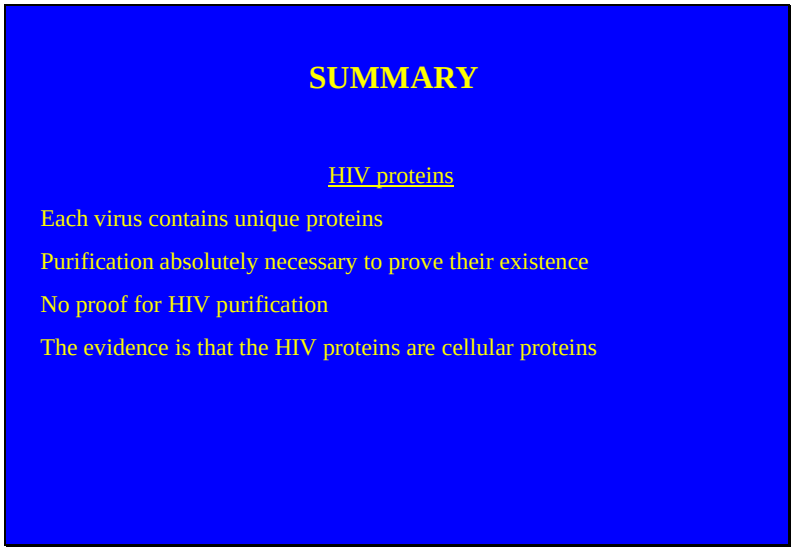

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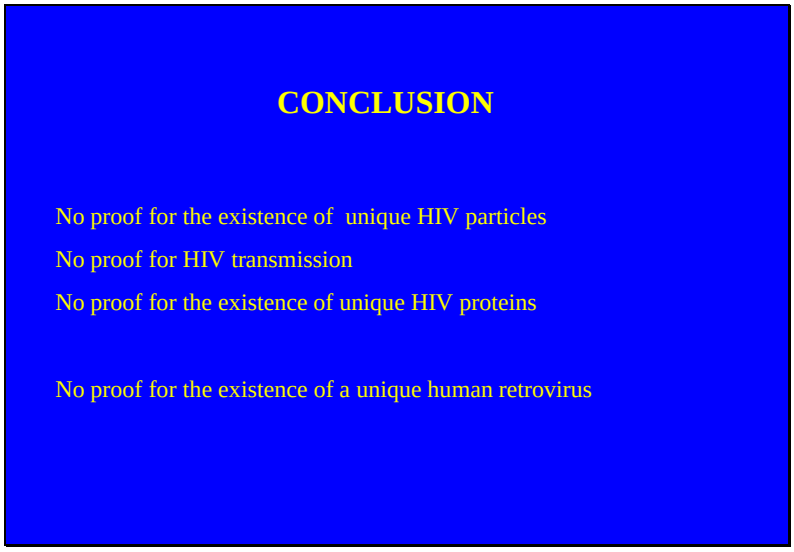

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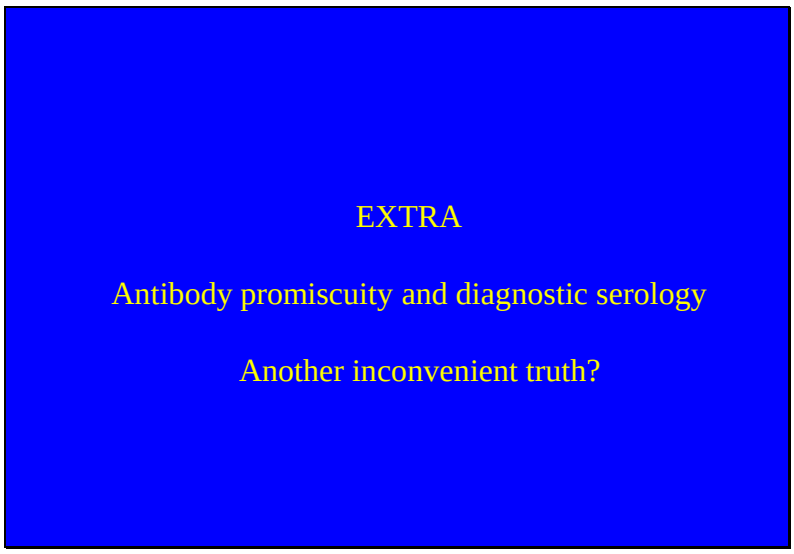

EXTRA

Antibody promiscuity and diagnostic serology. Another inconvenient truth?

In 1971 Sir Gustav Nossal wrote that antibody molecules possess "exquisite specificity...For each antigen there is a corresponding, different antibody. As with locks and keys only certain pairs fit". Notwithstanding, in the same book Nossal acknowledged that "an antibody molecule made following the injection of one antigen frequently can combine also with a second antigen of a related or similar shape…In other words, the antibody cross-reacts with the second antigen".[1]

Since then many authors have embraced the term "promiscuity" to express the fact that antibodies may react with more than one antigen.[2] Marchalonis states that "'epitope recognition promiscuity' is a property of antibodies of all vertebrate species…For many years, it was considered that a single antibody molecule bound only to the antigen to which it was raised, or at most to structurally homologous cross-reactive molecules. In fact the concept arose that monoclonal antibodies must be monospecific. The immunological community was shocked to find that B cells could be

polyreactive in binding to multiple antigens that were complex and ostensibly unrelated to one another".[3] It is also asserted that "Promiscuity is not a new concept…many antibodies elicited against a particular antigen have also been shown to bind other, structurally unrelated antigens.[2] According to Avrameas "…antibodies are polyspecific, that is, they are able to react with various dissimilar antigens such as: proteins, nucleic acids" and "they are able to react with more than to self or non-self antigens, often without any apparent antigenic similarities".[4] In 1997 Kramer *et al* noted that "high-affinity antibodies that have undergone antigen-driven somatic mutations are usually thought to be monospecific. Nevertheless, antibody cross-reactivity and polyspecificity have been observed since the earliest immunological studies" and "even high-affinity binding monoclonal antibodies are able to recognize more than one peptide epitope".[5]

Examples of the extent of antibody promiscuity are not difficult to find. In 1989 Baccala *et al* reported two monoclonal IgM natural autoantibodies (E7 and D23) that reacted with 11/12 unrelated antigens.[6] In their 1997 study entitled "Molecular basis for the binding promiscuity of an anti-p24 (HIV-1) monoclonal antibody [CB1-4]", Kramer *et al* reported reactivity against five unrelated peptides that competed with each other for binding to the paratope region of the antibody. The authors were able to construct binding supertopes derived from each peptide and "Data-base searches for proteins that match the supertopes resulted in the identification of more than 6000 heterologous proteins. A substantial number (>16%) [160] of those protein-derived peptides was able to bind CB4-1". Furthermore, the authors were able to obtain and test 11 heterologous proteins containing CB4-1 binding supertope sequences found amongst the "50 strongest CB4-1 binding peptides". These proteins included alcohol dehydrogenase (*E. coli*), UmuD (*E. coli*), candidapepsin (*Candida albicans*), myosin II heavy chain, non muscle (*A. castellani*) and X-Pro-dipeptidase (human)". "All of them were recognized by CB4-1 in denatured and/or native from using solid phase enzyme-linked immunosorbant assay".[5]

In 2005 Predki *et al* stated "In the research lab, antibodies are commonly used tools for affinity purification, co-immunoprecipitation, quantitation and localization of proteins within tissues or cells. In the clinical setting, antibodies are used to quantitate protein levels for diagnostic purposes, and their ability to either inhibit biological action or target specific cells for destruction forms the basis of their use as therapeutics. The success of each of these applications is largely due to, and contingent upon, the high

affinity and specificity of antibodies for their antigen targets. Even though specificity is a hallmark of antibodies, cross-reactivity is not infrequently observed. Unrecognized, such cross-reactivity can have adverse consequences. The ability to assess and identify antibody cross-reactivity is an important but often inadequately addressed requirement for both research and clinical applications". Predki *et al* also affirmed that "The literature is replete with examples of cross-reactive antibodies"; "Clearly, antibody cross-reactivity is very prevalent despite marketing efforts that suggest otherwise" and "The large number of cross-reactive antibodies is certainly cause for concern. However, perhaps more concerning are antibodies in current use with unrecognized cross-reactivity. Literature reports of cross-reactivity possibly represent the tip of a very large 'iceberg'".[7] These authors presented the case for using protein microarrays as a "new tool for profiling antibody cross-reactivity" and tested a monoclonal antibody directed against a phosphopeptide from the kinase MAPK-APK2 in a microarray consisting of approximately 2000 proteins. "Signals from the protein microarrays were normalized by the amount of protein estimated on the array. The top ranked protein has the highest signal:protein ratio. The protein towards which the antibody was directed was ranked 17". In other words, of the 40 proteins which reacted with this single antibody, binding by 16 was greater than that which occurred with cognate antigen. The authors also acknowledged that "the lack of a complete human proteome microarray prevents a comprehensive specificity analysis". One can likewise note that the universe of antigens is not confined to the full complement of human proteins and reactivity is also dependent on many other factors including culture and testing conditions. Hence, at present, the true extent of antibody specificity is not measurable. Since a polyclonal antibody response is a set of monoclonal responses, this problem of defining antibodies and their cognate antigens is even further compounded.

In light of such evidence the scientific community has recognised that antigens and antibodies do not react monogamously and moved on from the notion of "one antigen, one antibody". Obviously, as Predki *et al* stated, "Unrecognized, such cross-reactivity can have adverse consequences. The ability to assess and identify antibody cross-reactivity is an important but often inadequately addressed requirement for both research and clinical applications". Yet such "adverse consequences" appear to be under appreciated, including by clinicians. Indeed, in a straw poll undertaken by a colleague in a teaching hospital in 2006, all resident staff

asked were of the opinion that antibody reactivity with a microbial protein proves infection with that microorganism.

It takes only moments to appreciate that if antibodies are promiscuous so too are antigens. Of primary interest to clinicians is not the chemistry of antibody/antigen interactions but whether or not the reactions observed *in vitro* between antibodies in a patient serum and a given antigen are specific for exposure to or infection with a particular antigen or microorganism. Our view is that the only way to obviate the problem of antibody promiscuity and determine the specificity of a serological test, (not to be confused with antibody specificity), is to measure it against a gold standard which best represents whatever the test is claimed to prove. However, in doing so one must distinguish between testing to confirm a syndromic diagnosis and testing to prove infection with a particular microorganism. If the former, the gold standard is the syndrome (however defined). If the latter, the gold standard must be the organism itself (isolation). In the literature there is a serious dearth of data in regard to the use of microbial isolation as a gold standard for serology, especially in the case of viruses. In view of this and the new appreciation of the unknowable extent of antibody/antigen cross-reactivities, what confidence can clinicians place in serological diagnoses of infectious agents?

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Link to http://theperthgroup.com/LATEST/PGRevisitHIVExistence.pdf

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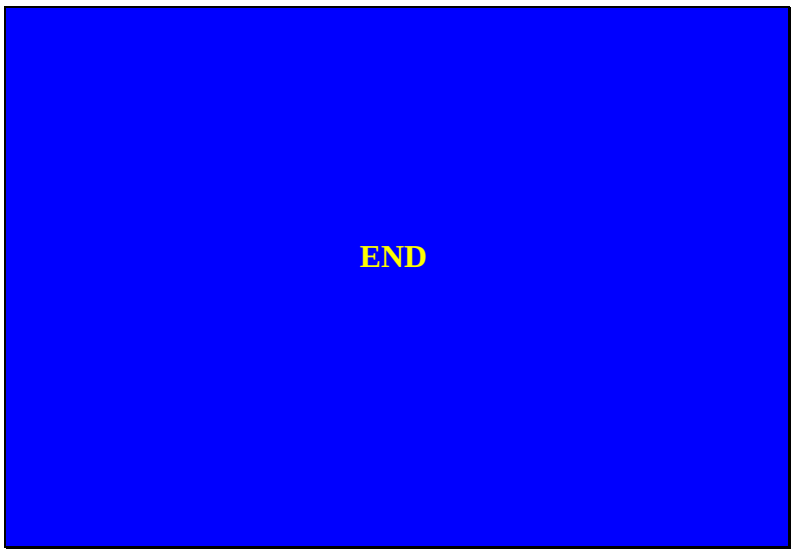

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