BACK

The Perth Group

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Montagnier's proof for the existence of the BRU p25, p45 and p80 proteins

Montagnier's isolation experiments from 1983

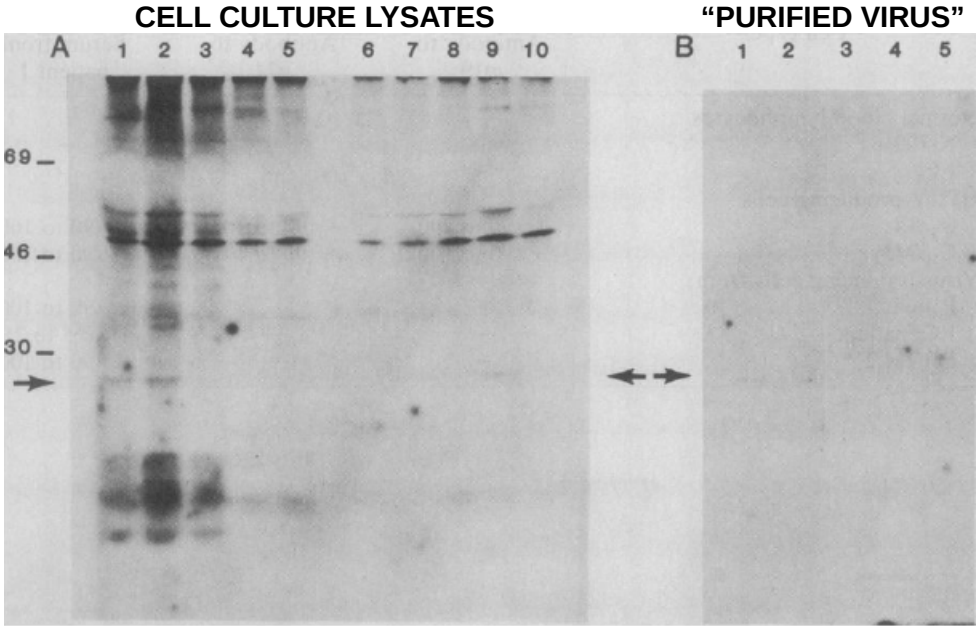
1983 Luc Montagnier and Francoise Barré-Sinoussi and their colleagues at the Pasteur Institute published a paper in *Science* entitled "Isolation of a T-lymphotropic retrovirus from a patient at risk for acquired immune deficiency syndrome (AIDS)".[1] In this paper they reported cell culture experiments performed on lymph node tissue excised from a male homosexual code named BRU. Montagnier claimed to have isolated a new human retrovirus from BRU which he named lymphadenopathy-associated virus (LAV). The name was derived from the generalised lymph node enlargement commonly seen in at risk patients who may later develop AIDS (as did BRU). LAV is now called the human immunodeficiency virus and for its discovery Montagnier and Barré-Sinoussi shared the 2008 Nobel prize in Physiology or Medicine.[2] In the *Science* paper Montagnier claimed to have purified the BRU virus but did not publish affirmatory electron microscopy evidence.

To prove the BRU virus was a new agent Montagnier had to analyse its proteins and show they were not those of two human retroviruses Gallo claimed he had earlier discovered – HTLV-I and HTLV-II. The obvious way to do this was to purify the viral particles, extract the proteins and then compare their amino-acid sequences with those of HTLV-I and HTLV-II proteins. This was not Montagnier's approach. Instead he conducted a laboratory technique known as immunoprecipitation, a procedure claimed to identify and characterise proteins by means of reactions between proteins and antibodies. This raises a number of problems.

Precipitation in "immunoprecipitation" has the same meaning as in chemistry. For example, both silver nitrate and sodium chloride are soluble in water and when a solution of one is added to a solution of the other a dense white precipitate of silver chloride forms. Precipitate is proof of reaction. "Immuno" signifies that one of the reactants is an antibody. The other is usually a protein and the reaction can be written **Ab + protein → Ab-protein complex**. Immunoprecipitation "Involves using an antibody that is specific for a known protein to isolate that particular protein out of a solution containing many different proteins. This process can be used to isolate and concentrate a particular protein from a sample containing many thousands of different proteins" (emphasis added). "Specific for a known protein" means the antibody reacts with the named protein to the exclusion of all others. (The procedure would not work if this were not the case). Hence if a precipitate forms when the specific antibody is added to a mixture of different proteins, the designated protein must be present. In the perennially anthropomorphised language of immunology the antibody is said to "recognise" the protein.[3]

In his fourth experiment Montagnier incubated "infected" umbilical cord lymphocytes for 20 hours with [^{35}S] methionine. (The amino-acid methionine is incorporated into the proteins produced in the culture. Radioactivity enables their detection following exposure to a photographic plate). From a sample of the cell free supernatant "The virus was purified by banding on a sucrose [density] gradient". Then Montagnier added BRU's serum containing the thousands of BRU antibodies to the "purified [radioactively]labeled virus", (the 1.16 g/ml density gradient material) and allowed the mixture several hours to react. Then the antibodies in BRU's serum that had not reacted (those that failed to "recognise" anything) were washed away leaving behind the Ab-protein complexes whose protein moieties had been "recognised". Then the antibody components were dissociated (removed) leaving behind the "recognised" proteins. Since the proteins were still in mixture a sample was

placed on a gel and the proteins separated from one another by application of an electric field (electrophoresis[4]). Finally a photographic plate was exposed to the gel whereupon the radioactive ^{35}S methionine revealed the locations of the proteins. The result is Lane 1, part B of Figure 3 on page 870 of Montagnier's paper.[1]



Part A is not relevant to this discussion but is included to demonstrate the typical findings of this technique. The black bands are the sites where ^{35}S reveals the proteins that have reacted with various antibodies.

The salient photograph is Part B which shows the results of adding BRU's and other sera (antibodies) to the proteins present the "purified-labeled virus". The five lanes in B are:
Lane 1 = serum from BRU
Lane 2 = serum from another patient
Lanes 3&4 = sera from healthy donors
Lane 5 = goat antiserum to HTLV-I p24 (provided by Gallo).

Montagnier reported that lanes 2-5 did not show any proteins. (The dots are artefacts). Montagnier asserts that Lane 1 contains three proteins, (p25, p45 and p80) that were "recognized" by BRU's antibodies. However we and several others could not see any proteins of any molecular weight in Lane 1. When we asked a local HIV expert what he saw (blinded to all but the figure), his reply was "nothing".

Given that Montagnier had no retroviral particles in his "purified" virus and this gel is Montagnier's sole evidence for the existence of p24, it is impossible to conclude p24 is an HIV protein and that p24 proves the existence of HIV. In 2012 the Perth Group wrote to the editor of *Science* pointing out the problematic nature of Montagnier's Figure 3B. There was no reply.

Nonetheless, since 1983 detection of p24 in plasma or cell cultures using an anti-p24 antibody has been used in countless studies to detect and quantify plasma viraemia and prove the "isolation" of HIV. These include studies which monitor the response to antiretroviral drugs, including that of children in the seminal ACTG 076 study[5] claimed to prove AZT prevents mother-to-child transmission of HIV.[6]

One should note that p24 is the only protein Montagnier reported in the BRU “virus” and is not a reverse transcriptase.

ENDNOTES

1. Barré-Sinoussi F, Chermann JC, Rey F, Nugeyre MT, Chamaret S, Gruest J, Dauguet C, Axler-Blin C, Vezinet-Brun F, Rouzioux C, Rozenbaum W, Montagnier L. Isolation of a T-lymphotropic retrovirus from a patient at risk for acquired immune deficiency syndrome (AIDS) Science 1983 220:868-871.
2. Lever AM, Berkhout B. 2008 Nobel prize in Medicine for discoverers of HIV Retrovirology 2008 5:91 http://www.biomedcentral.com/content/pdf/1742-4690-5-91.pdf
3. Antibodies “recognising" HIV proteins (and presumably not "recognising" all others) is an example of anthropomorphisms* that immunologists use to "explain" the immune system. Recognise means "to perceive something or someone as already known". Antibodies do not have memories. Antibodies are molecules which may or may not react with other molecules. Sodium chloride and silver nitrate react to produce silver chloride and sodium nitrate. Chemists do not claim this reaction is the result of molecular recognitions. From what Marcholanis and Kramer and many other scientists have reported, antibodies are unreliable witnesses: one and the same antibody may "recognise" many different molecules. Hence the identity of certain proteins as "HIV", and their use as antigens in the "HIV" antibody tests, is highly questionable.
*Further examples at
https://www.sciencedaily.com/releases/2014/11/141110124346.htm
www.abc.net.au/science/articles/2011/03/31/3177528.htm.

4. Electrophoresis is a procedure that separates a mixture of proteins in solution. An aliquot of the mixture is placed at one end of a gel across which a DC voltage is connected. The gel can be visualised as a molecular sieve which separates proteins according to their molecular weights and charge. Under the influence of the electric field proteins move through the gel – lighter proteins moving faster than the heavier. After several hours the voltage is removed and the gel stained with a protein-specific dye. This reveals the positions of the proteins as a series of dark, horizontal lines/bands. Bands are thicker lines and the darker the lines/bands the greater the concentration of protein at that position in the gel. The molecular weight of each protein is approximated by comparing its position with the positions of proteins of known molecular weights (markers) electrophoresed at the same time in a parallel gel. One should note that molecular weights determined by electrophoresis are not precise. For example, a protein of molecular weight 24K might be measured as 25K, especially if the electrophoresis is performed on material obtained from different experiments in a different or even the same laboratory.

5. Connor EM, Sperling RS, Gelber R, Kiselev P, Scott G, O'Sullivan MJ, VanDyke R, Bey M, Shearer W, Jacobson RL, et al. Reduction of maternal-infant transmission of human immunodeficiency virus type 1 with zidovudine treatment. Pediatric AIDS Clinical Trials Group Protocol 076 Study Group N Engl J Med 1994 331:1173-1180.
6. Papadopulos-Eleopulos E, Turner VF, Papadimitriou JM, Alfonso H, Page BAP, Causer D, Mhlongo S, Fiala C, Miller T, Brink A, Hodgkinson N. Mother to Child Transmission of HIV and its Prevention with ATZ and Nevirapine. Perth, The Perth Group, 2001, pp. 204.