EVERYBODY REACTS POSITIVE ON THE ELISA TEST FOR HIV

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For the last 6 years I have been working at the laboratory of clinical immunology in one of the most prestigious University Hospitals in the City of New York. Here I have had the opportunity to personally run and get to know in detail the current tests used for the diagnosis of HIV status, namely, the ELISA, Western Blott and Viral Load tests.

1. Diluting the serum for the ELISA test.

The ELISA test is a test for antibodies against what is supposed to be the Human Immunodeficiency Virus or HIV. To run this test, an individual's serum has to be diluted to a ratio of 1:400 with a special specimen diluent. According to the test kit manufacturer this diluent contains

> *0,1% triton X-100, Bovine and Goat Sera (minimum concentration of 5%) and Human T-Lymphocyte Lysate (minimum titer 1:7500). Preservative: 0.1% Sodium Azide* (1).

This extraordinary high dilution of the person's serum [400 times] took me by surprise. Most serologic tests that look for the presence of antibodies against germs uses neat serum [undiluted]. For example, the tests that look for antibodies to hepatitis A and B viruses, rubella virus, syphillis, hystoplasma and cryptococus, to mention a few of them, use straight serum [undiluted]. However, to try to prevent false positive reactions some serologic tests use diluted serum; for example this is the case with tests that look for antibodies to measles, varicelia and mumps viruses which use a dilution of 1:16, to cytomegalovirus [CMV] 1:20 and to Epstein-Barr Virus [EBV] 1:10.

The obvious questions are: What makes HIV so unique that the test serum nedds to be diluted 400 times?. And what would happen if the individual's serum is not diluted?.

2. Testing the ELISA test without diluting the serum.

To answer these questions I ran an experiment in a medical laboratory in Yorktown Heights, New York. I ran it using the same test kit reagents that are usually used to run the ELISA test in most clinical laboratories worldwide (1).

I first took samples of blood that, at 1:400 dilution, tested negative for antibodies to HIV. I then ran the exact same serum samples through the test again, but this time without diluting them. Tested straight, they all came positive.

Since that time I have run about 100 specimens and have always gotten the same result. I even ran my own blood which, at 1:400 reacts negative. At 1:1 [undiluted] it reacted positive. I should mention that with the exception of my own blood, the patient samples all came from doctors who

requested HIV tests. It is therefore likely that most of the blood samples that I tested belonged to individuals at risk for AIDS.

According to Abbott Laboratories, the absorbance value [yellow color intensity]

> *develops in proportion to the amount of antibodies to HIV-1 which is bound to the bead* (1).

What I noticied is that the absorbance values of the specimens that tested negative when diluted [1:400], but positive when undiluted [1:1], had lower values than the samples that, diluted, react positive on both the ELISA and Western Blott tests. This would probably mean that the blood that is negative when diluted but positive when undiluted has a lower level of antibodies than the diluted blood that is doubly positive and, therefore, may probably test negative on the Western Blott test. However, I have not had the opportunity to check this hypothesis.

The graphic below ilustrates how blood that reacts negative for HIV at 1:400 ratio always turn positive when run at 1:1 [undiluted].

Run of ELISA test for HIV with two different concentrations of the person's serum.	
(a) Results at 1:400	(b) Results at 1:1
9112324b G5 0.076 ---	9112324b G5 0.262 reactive
9112325b H1 0.081 ---	9112325b H1 0.259 reactive
9112326b H2 0.071 ---	9112326b H2 0.329 reactive
9112327b H3 0.060 ---	9112327b H3 0.401 reactive
9112328b H4 0.073 ---	9112328b H4 0.345 reactive
9112329b H5 0.062 ---	9112329b H5 0.343 reactive
9112330b J1 0.060 ---	9112330b J1 0.234 reactive
9112331b J2 0.077 ---	9112331b J2 0.306 reactive
9112332b J3 0.067 ---	9112332b J3 0.248 reactive
9112333b J4 0.086 ---	9112333b J4 0.222 reactive

Column (a) shows 10 specimens reacting negative at 1:400 dilution.
Column (b) shows the same specimens reacting positive at 1:1 dilution.

It is important to note that the Western Blott antibody test for «HIV» also needs serum to diluted. Although it too has an usually high dilution, here the individual serum is only diluted at the ratio of 1:50 (2). I have not yet had the opportunity to run this test with undiluted [1:1] specimens.

3. Discussion.

The following are three possible explanations for why undiluted specimens of blood always react positive at the ELISA test:

3.1. Everybody has HIV antibodies.

It is accepted worldwide that the ELISA test for HIV detects antibodies against what is known as the Human Immunodeficiency Virus (3-4-5-6).

And the pharmaceutical company that commercialises the ELISA kits states that

> *Abbott HIVAB HIV-1 EIA is an vitro qualitative Enzyme Immunoassay for the Detection of Antibody to Human Immunodeficiency Virus Type 1 (HIV-1) in Human Serum and Plasma* (1).

Since all undiluted blood specimens react positive on the ELISA test, a test that supposedly tests for antibodies to HIV, the results presented here suggest that every single human being has HIV antibodies. And this suggests that everybody has been exposed to HIV antigens.

This would mean that all of us have been exposed to the virus that is believed to be the cause of AIDS. The people that react positive even at a dilution of 1:400, would be the ones that have had the highest level of exposure to HIV antigens. The rest of the people -the ones that only react positive with undiluted serum [1:1]- would have had a lower level of exposure to HIV.

3.2. Everybody has different levels of HIV infection.

It is also believed worldwide that a person that reacts positive for antibodies against HIV has not only been exposed to but is infected with a deadly virus that causes immunodeficiency (3-4-5-6). Therefore, the positive reactions of all undiluted sera would mean that everybody, or at least all the blood samples that I have tested, including my own, infected with this «deadly» virus. The ones that react positive at a ratio of 1:400 would simply have a higher level of «deadly» infection than the «deadly» infection has by the ones that reacts positive only with undiluted serum.

3.3. The test is not specific for HIV.

The results presented here could also mean that the tests used for detecting antibodies to HIV are not specific for HIV, as has been explained previously (7-8-9-10-11-12-13-14). In this case, there would be reasons other than HIV infection, past or present, to explain why a person reacts positive to it. The test also reacts positive in the absence of HIV (7-8-9-10-11-12-13-14).

The scientific literature has documented more than 70 different reasons for getting a positive reaction other than past or present infection with HIV (7,10,11,14,15). All these conditions have in common a history of polyantigenic stimulations (15,16).

Even Abbott Laboratories is well aware of the specificity problems with the ELISA tesst. This is why they state:

> *EIA testing alone cannot be used to diagnose AIDS, even if the recommended investigation of the reactive specimens suggests a high probability that the antibody to HIV-1 is present*

and

> *Although for all clinical and public health applications of the EIA both the degree of risk for HIV-1 infection of the person studied and the degree of reactivity of the serum may be of value in interpreting the test, these correlations are impefect. Therefore, in most settings it is appropiate to investigate repeatably reactive specimens by additional more specific or supplemental tests* (1).

Interestingly, there are countries like Great Britain where the diagnosis of HIV status is based on the ELISA test alone. No Western Blott or any other test is needed there.

The only proper way for establishing the sensitivity and specificity of a given test is with a gold standard. However, since HIV has never been isolated as an independent purified viral entity (17-18-19), there cannot be a gold standard for HIV. The sensitivity and specificity of the antibody tests for HIV have instead been defined based on the assumption that HIV is the cause of AIDS. In this way,

> *The Abbot studies show that: Sensitivity based on an assumed 100% prevalence of HIV-1 antibody in AIDS patients is estimated to be 100% (144 patients tested).*

and

> *Specificity based on the assumed zero prevalence of HIV-1 in random donors is estimated to be 99.9/o (4.777 random donors tested* (1)*).*
>
> *At present there is no recognized standard for establishing the presence and absence of HIV-1 antibody in human blood. Therefore sensitivity was computed based on the clinical diagnosis of AIDS and specificity based on random donors* (1).
>
> [Emphasis is mine].

Since there is no scientific evidence that the ELISA test is specific for HIV antibodies, a reactive ELISA test at any concentration of the serum would mean presence of nonspecific or polyspecific antibodies (20). These antibodies could be present in all blood samples. They are most likely a result of the stress response, having no relation to any retrovirus, let alone HIV (21,22). In this case, a reactive test could be a measure of the degree of one's exposure to stressor or oxidizing agents (15,16).

The inevitable conclusion is that all positive reactions for antibodies to HIV are simply false positives. If nobody is positive for HIV, then people who react positive on the ELISA test do so due to something other than HIV.

4. Proposal to find out the real meaning of the «HIV antibody» tests.

To uncover the meaning of these tests I propose a simple experiment: Take blood from three groups of a people and run the tests highly diluted, undiluted and at a wide spectrum of dilutions in between. The first group would be a group of healthy people of many age groups; the second group would be a group of people from the conventional AIDS «risk groups»; the third group would be a group of people with clinical conditions both related and unrelated to AIDS. All groups would be subjected to both the ELISA and Western Blott tests.

Additionally, all blood samples could be subjected to «the viral load test for HIV».

The results of such an experiment could determine whether these test measurements bear any relationship to an individual's level of exposure to stressor or oxidizing agents. If so, the tests could be salvaged as a measure of an individual's level of intoxication.

Let us find the economic support necessary to run this experiment. In the mean time, since people are reacting positive on tests that are not specific for HIV, let's please stop labeling them as «HIV positive».

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