The Link - Update

THE NEWSLETTER OF HOUSTON CLINICAL RESEARCH NETWORK A DIVISION OF MONTROSE CLINIC

April, 1995

CHILD APPEARS TO HAVE CLEARED HIV INFECTION

A report in *The New England Journal of Medicine* published March 30 documents the case of a child identified shortly after birth who appeared to be infected with HIV but then became negative on all laboratory tests for HIV by three months after birth. The child is now five years of age and is free of any laboratory or clinical evidence of HIV infection.

Both the report and an accompanying commentary on it acknowledge there have been previous reports of both infants and adults who appear to have cleared HIV infection; however, because prior case reports have not been well controlled and well documented, they have usually been dismissed as laboratory errors. What is different about this case is the authors of the report very carefully documented all of their laboratory and clinical findings.

The child was born without laboratory or clinical signs of HIV infection. The mother of the child had become HIV-positive during pregnancy and has remained positive when tested using both the standard HIV antibody test as well as polymerase chain reaction (PCR) testing (a very sensitive test which measures virus in the blood). By 19 days after birth, the child was positive by HIV culture (growing virus in a laboratory out of blood drawn from the child) and by standard testing for antibodies to HIV.

By three months, PCR and viral cultures were negative, and the child was losing antibodies to HIV.

Through 51 days after birth, the child continued to test positive for both antibodies to HIV as well as measures of HIV in the blood. By three months, PCR and viral cultures were negative, and the child was losing antibodies to HIV. Since that time, all laboratory tests for HIV have been negative. The child remains healthy, with normal growth and development.

The scientists studying this child also performed several tests to match DNA of the virus and of the child's blood to ensure all laboratory results were from the same child with the same virus. The virus in the child was similar by DNA typing to that in the mother, lending further evidence that the child was infected during birth.

In the discussion section of the report as well as an in an accompanying commentary, several implications of this finding are reported. There may be a much greater incidence of persons being exposed to HIV without permanent infection being established than has been known. Several studies of HIV-negative gay men at high risk have suggested clearance of an exposure to HIV; however, these reports have been controversial and not well accepted. This new evidence may stimulate a renewed interest.

Certainly, if some individuals are capable of clearing an infection with HIV, the study of such cases may have profound implications for developing treatments and vaccines. Such study may also provide an opportunity for greater understanding of how HIV establishes infection and causes disease.

Continued on next page

Other Reading Related to Possible Clearance of HIV Infection

Baur, A. et al. Continuous clearance of HIV in a vertically infected child. Lancelot 1989;2:1045.

Borkowsky, W. et al. Human immunodeficiency virus type 1 antigenemia in children. J. Pediatr 1989; 114:940-5.

Clerci, M. et al. Cell-mediated immune response to human immunodeficiency virus (HIV) in seronegative homosexual men with recent sexual exposure to HIV-1. J Infect Dis 1992;165:1012-9.

Cheynier, R. et al. Cytotoxic T lymphocyte responses in the peripheral blood of children born to human immunodeficiency virus-1-infected mothers. Eur J Immunol 1992;22:2211-7

Pan, L.Z. et al. Lack of detection of human immunodeficiency virus type 1 infection in homosexual men who remain seronegative for prolonged periods. N Engl J Med 1989;320:1458-62.

Several important questions are raised by the prospect of some individuals apparently clearing HIV infection. How often and how does it happen? Is it the result of genetic variation in the virus which has created some less pathogenic strains, or is there an immunologic variation which allows some people to clear infection but not others?

If the mechanism is immunological, an immune-based approach to prevent mother to child transmission during pregnancy may be possible. Likewise, if immune responses that allow exposure but not permanent infection can be identified and understood in detail, new approaches to vaccine development as well as immune-based therapies for those already infected might be possible.

If, in fact, less pathogenic strains of the virus are the underlying reason clearance of HIV infection may occur, the study of such strains compared to strains which establish infection and cause disease may lead to greater understanding of HIV. This in turn might lead to different treatment approaches for pathogenic strains.

Finally, it is possible this clearance of HIV occurred at a cellular level -- fetal or neonatal cells may in some way permit transient infection by HIV that would be pathogenic in adults.

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HIV AND HEART MUSCLE DISEASE

A new threat surfacing for persons living with AIDS?

Dilated cardiomyopathy (dilated -- enlarged, cardio -- heart, myopathy -- muscle disease) may play a more significant role in HIV disease than had previously been thought according to a recent report in the *Journal of the American Medical Association.* Although heart disease in HIV has been studied previously, this was one of the largest studies (296 people participated) and showed the strongest correlation between HIV disease progression and development of dilated cardiomyopathy.

Persons with CD4 cell counts of less than 100 were much more likely to experience dilated cardiomyopathy. This indicates cardiomyopathy develops in relation to the HIV disease process and is not always an independent condition. The study also found a significant difference in survival related to whether or not dilated cardiomyopathy was present in persons with AIDS. Those with this myopathy survived an average of 101 days, while those with normal hearts and a CD4 cell count less than 20 survived an average of 472 days.

Those with this myopathy survived an average of 101 days, while those with normal hearts ... survived an average of 472 days.

This report raises several issues. The underlying etiology (causes) of these heart problems are not always known and may vary. Periodic screening for cardiac abnormalities may be indicated; however, with the possibility of unknown etiology, treatment for abnormalities may be difficult if possible at all. However, if the underlying cause can be identified, treatment may be possible.

Research has indicated cardiomyopathy and other forms of heart disease may develop in relation to opportunistic infections. Several infections have been found in the heart, including tuberculosis, cryptococcus, salmonella and mycobacterium-avium complex (MAC). In many cases, if the underlying pathogen could be identified, the heart conditions were successfully treated.

There is also evidence indicating cardiac problems in AIDS may occur as either a direct or indirect result of HIV infection. A few cases of HIV actually infecting the heart have been identified. Some researchers have also found evidence of heart problems developing due simply to the increased stress placed upon the cardiac and respiratory systems by HIV and associated infections. It is possible some of the therapies required to treat and prevent HIV-related opportunistic infections may also contribute to the development of cardiac problems in the later stages of HIV disease progression.

Finally, there is research indicating some of the heart abnormalities identified in late stage AIDS may be the result of a previous infection of the heart muscle which has since healed. This may be particularly related to prior pnuemocystis carinii infection.

Clearly, more research examining dilated cardiomyopathy and other heart problems in HIV disease is needed. In the meantime, most of the scientist studying HIV-related heart disease recommend periodic cardiac monitoring, especially in persons with a CD4 cell count less than 100.

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Kinney, E.L. et al. The treatment of infectious causes of AIDS associated heart disease. Int Conf AIDS. 1989 Jun 4-9; Abstract no. M.B.P.282.

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Nyamathi, A. AIDS-related heart disease: a review of the literature. J Cardiovasc Nurs 1989; 3(4):65-76.

MORE ON PROTEASE INHIBITORS

Protease inhibitors are a promising new class of anti-HIV drugs which work in a different manner than reverse transcriptase inhibitors such as AZT. A new clinical trial of a protease inhibitor will begin enrollment soon through HCRN. Anther protease inhibitor study may also begin before the end of the year.

The first study slated to begin enrollment is a clinical trial of the protease inhibitor produced by Merck, called MK-639. The study will compare MK-639 used alone to MK-639 combined with AZT and to AZT used alone. Enrollment is open to persons with a CD4 cell count between 50 and 500 who have not yet begun anti-HIV therapy.

If enrollment in this study is successful, Merck has indicated HCRN will then also be a site for another clinical trial of MK-639 which will study the drug in persons with a CD4 cell count less than 50 who have been taking AZT. It is hoped this study will begin this summer.

Several other companies have protease inhibitor products which may also become available in Houston in the future. Besides the Merck product, the protease inhibitors produced by Abbott and Roche have reached the most advanced stages of testing. The Roche product has had problems with not enough drug being absorbed into the bloodstream. Roche has a new formulation that may address this problem. HCRN is negotiating to conduct a study examing the newer version in combination with several other drugs.

Research Information / Treatment Action (RITA) has initiated a letter to all of the different pharmaceutical companies who produce protease inhibitors. The letter encourages these companies to make protease inhibitors more widely available and to include Houston in protease inhibitor studies.

Treatment activists across the country have joined in similar efforts to advocate for greater access to these promising new drugs by calling for the companies to provide expanded-access studies. For more information, contact RITA at 523-1519.

For more information on protease inhibitors and their current development status, consult:

- *Beta, March 1995 issue, pages 30-37.*
- *Research Initiative / Treatment Action (RITA), March 1995 issue, page 3.*
- *The Link - Update, May 1994 issue, page 3.*
- *The Link - Update, February 1995 issue, page 2.*

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THE NEWSLETTER OF HOUSTON CLINICAL RESEARCH NETWORK A DIVISION OF MONTROSE CLINIC

May, 1995

LOCAL DESIGN OF HYDROXYUREA STUDY TO BEGIN ENROLLMENT SOON

Houston Clinical Research Network (HCRN) will open enrollment for a study of hydroxyurea in middle or late June. The study, which was designed by Wayne Bockmon, M.D. and HCRN staff with consultation from the MARC group, will examine hydroxyurea in persons with a CD4 cell count between 0 and 500 and a baseline viral titer above 10,000 copies as measured by polyermase chain reaction (PCR). The study will be open to persons who have not yet begun anti-HIV therapy or who will go off their current therapy for six weeks before entering the trial.

In the test tube, hydroxyurea inhibits HIV replication used alone or in combination with AZT, ddI and ddC. It seems most effective when used with ddI. Thus far, there is no published data concerning the use of hydroxyurea in humans. Several studies of combining hydroxyurea with other therapies are being planned; however, HCRN is unaware of any study examining the drug used alone. (For more information on hydroxyurea, consult the April 1995, July 1994 and January 1995 issues of *The Link*).

The HCRN study will compare hydroxyurea taken alone at 500mg daily, 1,000mg daily and 2,000mg daily. Twenty people will be assigned by chance to each off the three dosage regimens; thus, 60 persons total will participate.

Primarily, the study is designed to determine if persons within any of the three dosage regimens have less virus in their blood after twelve weeks of taking hydroxyurea than they did before they began taking it.

Also, the study will determine if any of the three doses of hydroxyurea is more effective than the others at reducing the amount of virus in the blood.

The study will also examine the safety of the drug and whether any of the three doses appears to be better tolerated than the others.

This study of hydroxyurea is the first of several locally designed studies HCRN hopes to conduct. It is a good example of the potential inherent in community-based research.

Hydroxyurea is a drug approved by the Food and Drug Administration for other uses. It is being used in the clinical setting to treat HIV, but data to substantiate its potential is not being collected. Community-based research organizations are well-equipped to collect data on drugs being used in the clinic for new indications.

FOR MORE INFORMATION ON HYDROXYUREA, CONTACT CHRIS JIMMERSON OR JAMES HARRISON AT 520-2000

NEW RESOURCES AVAILABLE

☑ *Computerized drug information -- research drug side effects, interactions with other drugs, dosing information and much more.*

☑ *Literature Searches --work with an HCRN staff member or volunteer to retrieve the latest HIV research information via a modem or through compact disk software.*

ONGOING TRIALS FOR HIV-RELATED CONDITIONS

HCRN continues to enroll participants for several clinical trials of therapies intended to battle HIV-related infections and conditions. Three clinical trials address ano-genital herpes infections in persons who are also infected with HIV. One study compares **valaciclovir** to standard acyclovir for suppressing herpes outbreaks. Another study compares the two drugs for treating these outbreaks when they occur. Finally, another study examines a drug called **cidofovir** (HPMPC), a topical treatment, used when a herpes outbreak has occurred and has not responded to standard therapy.

Another study of cidofovir uses the drug for treating ano-genital warts in persons also infected with HIV. This study also uses a topical form of cidofovir. Both the herpes and warts cidofovir studies are open-label, meaning all participants will receive the drug and will know what dosage they are getting.

Neutropenia is a condition that can be caused by HIV disease, as well as by many of the drugs persons with HIV take. This condition is a low number of a type of white blood cell called a neutrophil. Nuetropenia can leave a person susceptible to bacterial infections. A study currently open to enrollment at HCRN examines **Filgrastim** (Neupogen) to treat persons with HIV and neutropenia.

For information on any of these studies, please call 520-2000.

STUDY OF COMBINATION THERAPY WITH d4T AND ddI TO BEGIN

A study examining combination treatment with ddI and d4T will begin enrollment soon. The study will be open to persons with a CD4 cell count between 200 and 500 who have not yet begun anti-HIV therapy. The study will compare five different dosage regimens of each of the drugs taken together.

It is hoped the two drugs used together will prove beneficial to a greater degree than when used alone; however, because they can have similar side effects, the study will also examine safety very closely. It is hoped an optimum dosage for combining the drugs safely can be established.

SELECTED LABORATORY VALUES NORMAL RANGES

How low is too low?

LABORATORY TEST	NORMAL RANGE
White Blood Cell Count (WBC)	5,000 to 10,000 cells/mm^3
Neutrophils	3,000 to 7,000 cells/mm^3
Lymphocytes	1,000 to 4,000 cells/mm^3
CD4 Cells	493 to 1,191 cells/mm^3, 34% to 67%
CD8 Cells	182 to 785 cells/mm^3, 10% to 42%
Platelet Count	150,000 to 350,000 cells/mm^3

ENROLLMENT CONTINUES FOR ANTI-HIV CLINICAL TRIALS

Enrollment continues for two clinical trials of a drug known as ***BuCast*** (MDL 28,574A). BuCast inhibits a cell enzyme known as alpha-glucosidase-1, which HIV uses to assemble its outer envelope. It is hoped this drug will reduce the ability of newly produced HIV to infect new cells.

The first study examines BuCast in persons with a CD4 cell count between 100 and 300 with less than or equal to six months cumulative prior therapy with other anti-HIV drugs. Persons who enter this study will be assigned by chance to take one of four different doses of BuCast alone for a certain time period and then to take this dose of BuCast with AZT. One group will take AZT alone.

The second study is open to persons with a CD4 cell count between 301 and 500. It works like the study above, except it is placebo-controlled.

HCRN is also enrolling a clinical trial of the **Merck protease inhibitor**, also known as MK-639. Protease inhibitors work differently than AZT, ddI and similar drugs, which interfere early in HIV's replication process. Protease inhibitors interfere with the protease enzyme the virus uses to begin reassembling its genetic material into new virus.

The clinic trial for which HCRN is enrolling currently is open to persons with a CD4 cell count between 50 and 500 who have not yet begun anti-HIV therapy. People who enter the study will be assigned by chance to take either real MK-639 plus real AZT, fake MK-639 plus real AZT or real MK-639 plus fake AZT.

At a certain point in the study, if MK-639 seems safe and effective, all participants will be allowed to go on real MK-639. For more information on protease inhibitors, consult the May 1994, February 1995 and April 1995 issues of *The Link* and the March 1995 issue of the RITA newsletter.

Enrollment continues for a clinical trial of an immune modulating drug called **thymopentin**, which demonstrated some benefit in a previous study when used with AZT. Thymopentin may stimulate maturation of T-cells; however, its exact mechanism of modulating the immune system is not entirely understood.

This clinical trial is open to persons with a CD4 cell count between 100 and 400 who are taking no antiviral therapy, AZT, ddI, ddC, d4T, 3TC or a combination of these drugs. Participants must have at least six months treatment with AZT at any time prior to enrolling. Once enrolled, participants will be assigned by chance to get either thymopentin or placebo.

There is a provision in this study for all participants to eventually receive thymopentin if it proves useful.

HCRN is also conducting a clinical trial of **nevirapine**, which is a drug that fights HIV at the same point in its replication process as AZT; however, it has a much different chemical structure and toxicity profile.

For this study, participants must have a CD4 cell count between 200 and 500. Persons who have never taken anti-HIV drugs may be assigned by chance to get either nevirapine or placebo. Persons with three to 24 months prior experience with AZT will be assigned to either AZT plus nevirapine or AZT plus nevirapine placebo.

Again, at a certain point, all participants will be offered real nevirapine.

HCRN continues to seek participants for a trial examining whether therapy with AZT taken immediately after someone is infected with HIV will affect long-term disease progression.

The study seeks persons who have had a potential exposure to HIV, test negative on antibody testing and positive on antigen testing or who can document sero-conversion within the previous 30 days.

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THE NEWSLETTER OF HOUSTON CLINICAL RESEARCH NETWORK A DIVISION OF MONTROSE CLINIC

July / August, 1995

SPECIAL ISSUE -- LABORATORY MEASURES OF HIV VIRAL LOAD

Recently, two technologies for measuring viral load have advanced significantly. Viral load is sometimes used to mean different things. Used here, viral load refers to the amount of free virus in the blood. A large viral load can indicate HIV is replicating, potentially doing damage to the immune system. A very low viral load can indicate HIV is not replicating as much and therefore not doing as much harm.

Viral load tests usually express the amount of HIV in the blood in terms of a the number of RNA strands per milliliter of blood. These RNA strands are part of the structure of HIV. Viral RNA contains a chemical message which instructs HIV to integrate into cells and form new virus. There are two strands of RNA in each free viral particle. The number expressed by viral load tests refers to a measurement of the number of these HIV RNA strands in the blood.

There are two kinds of tests now available which can measure HIV viral load. One is called Polymerase Chain Reaction (PCR). The other is referred to as Branch DNA Analysis (bDNA). The two tests, how each of them work and their potential advantages and disadvantages are explained in the sections below.

PCR -- How it Works

PCR is a several step process. Without getting into the rather complicated science of how it works exactly, it is important to know PCR is a process in which a man made genetic molecule is used to specifically target HIV RNA. Through a heating and cooling process, this molecule causes the amount of HIV RNA present in a blood sample to double. This heating and cooling process is called a PCR cycle. The resultant, doubled amount of HIV genetic material is called PCR product.

If this PCR product is then put through the PCR cycle again, the amount of RNA produced is again doubled. Through this PCR cycling, an amplification of more than a million fold can be achieved over the course of 25 cycles. This Amplified RNA can then be used to calculate how much HIV was in the blood to begin with[1].

The advantage to this process is that it is very sensitive -- that is, it can detect and measure even tiny amounts of HIV. In fact, the company which makes the test claims it can detect HIV even when there are as little as 200 RNA copies in a milliliter of blood. However, this sensitivity can also be the test's disadvantage. It is subject to a tiny amount of HIV RNA being transmitted from one sample to another by a laboratory technician. Also, if one of the RNA strands breaks during a PCR cycle, the resultant two RNA strands may then be amplified many times over in subsequent cycles, leading to a falsely high copy number.

bDNA -- How it Works

bDNA analysis involves a process which uses man made molecules, genetically engineered to "match up" and bind to a specific region of HIV's structure. Through a process more complicated than will be explained here, a chemical which produces light binds to these molecules.

The amount of light produced is then measured and used to quantify the amount of HIV material present. Since the chemical which produces the light is bound to HIV, the more light produced; the more HIV. The less light produced; the less HIV[2].

Because bDNA doezs not involve multiplying RNA over and over, it is not as subject to the potential for contamination. It is also not susceptible to RNA breakage and a falsely elevated copy number as is PCR[3]. There is some indication bDNA results may be less variable due to the technology itself, meaning relatively small changes in viral load may be meaningful by bDNA but not by PCR.

bDNA's potential disadvantage is it is not as sensitive as PCR. The company which produces the test reports its lower limit to be 10,000 equivalents per milliliter -- there must be 10,000 HIV RNA strands in a milliliter sample of blood for the test to measure it.

Some scientists, using a variation of the bDNA test, have reported being able to detect as little as 1,000 RNA equivalents using an even smaller blood sample[4]. However, the version of the test currently commercially available remains at the 10,000 RNA equivalents lower limit.

bDNA OR PCR -- WHICH SHOULD I USE?

On the previous page, the potential advantages and disadvantages of these tests are outlined. However, such factors as cost and availability may be the most important factors to consider. In fact, the two tests correlate very well. That is, measurements of viral load done by PCR and bDNA on the same blood samples are very similar.

Studies comparing PCR and bDNA indicate a strong, positive correlation ranging from .78 to .89.

Correlation is a statistical test used to determine if changes in one variable are associated with changes in another variable. If the variables are viral load as measured by PCR compared to viral load as measured by bDNA from the same patients and blood samples, a strong correlation would indicate the two tests are very similar in their measurement of viral RNA copies. A weak correlation would indicate they are very different.

The statistical number generated by a correlation can range from -1 to 1. A correlation close to 1 indicates a strong, positive correlation -- high viral burden by PCR will equate with high viral burden by bDNA. Likewise, low viral burden on one test is likely to be the same or similar on the other.

A correlation near -1 would indicate a strong, inverse correlation. If as viral burden by PCR got higher, it tended to get lower when measured by bDNA, the tests would be inversely correlated. That, of course, would not be a good thing.

Finally, a correlation near 0 would indicate there is no correlation. If viral burden by bDNA generated numbers that had no relation whatsoever to viral burden measured by PCR, it could indicate the two tests are not measuring the same thing.

Studies comparing PCR and bDNA indicate a strong, positive correlation ranging from .78 to .89[5,6]. Because these correlations are very close to 1, it indicates if one has their viral load assessed by PCR and bDNA at the same time, the results would be very similar.

Another study found that in 37 patients with HIV, viral load could be detected and quantified in 86% of these patients using PCR and in 83% using bDNA[7], indicating the difference in lower limits may not be a large factor.

Besides accuracy and sensitivity, other important factors concerning laboratory tests are the tests' precision, reliability and variability. If a test result changes over time in an individual patient for reasons having little to do with disease progression, such as time of day or what laboratory performs the test, it can limit the usefulness of the test. Similarly, if the same test is performed twice on the same blood sample by the same laboratory and two very different results are obtained, it can indicate something problematic in the test methodology or laboratory performance.

Research indicates PCR is relatively precise and reliable and is not subject to large variance[8]. Other research found that while CD4 cell counts are subject to diurnal variation -- they change throughout the day -- PCR viral load is not[9]. It is likely the same is true of bDNA.

Studies examining bDNA have found it also to be precise, and reliable[10,11]. Viral load by bDNA has likewise been found to have relatively low variation[12].

Another consideration is how well a laboratory test hoped to be a marker for disease progression correlates with other markers for that disease. In this instance, one important relationship is how viral load relates to CD4 cell count.

Research indicates viral load by both bDNA[13] and PCR[14] are inversely correlated with CD4 counts. In general, people with high CD4 counts tend to have a lower viral load. People with lower CD4 counts tend to have a higher viral load. This is not an exact correlation, and there are many exceptions; however, it seems to indicate viral load and CD4 count are related.

Thus, viral load determined by either PCR or bDNA appears to be accurate, precise, reliable and related to CD4 cell count.

therapy long enough to determine whether it extends life can raise ethical issues, particularly if potentially more effective treatments become available during a trial. Additionally, because many people with HIV live many healthy years before developing AIDS, it may not be possible from a practical standpoint to maintain as many patients for as long a time period needed in a clinical trial to test a treatment's effect on clinical outcome.

This is where surrogate markers are useful. If a laboratory result can reasonably be assumed to predict clinical outcome, it can be substituted as the endpoint in a clinical trial.

In other words, if viral load is found to be a reasonable surrogate marker, it could be used in clinical trials in place of things like developing AIDS. In the example used above, a certain amount of reduction in viral load after twelve weeks would not only demonstrate anti-HIV activity, but would be used to predict that the drug will delay progression to AIDS and/or extend survival.

Thus, the effectiveness of a drug could be tested in a clinical trial using less people for a shorter time period if viral load is demonstrated to be a surrogate marker.

◆ *To diagnose HIV infection in infants born to HIV-infected mothers* - One problem encountered when women with HIV give birth is that their infants will be born with maternal antibodies to HIV and carry those antibodies for up to 18 months. Thus, using conventional antibody testing to determine if such infants are infected is not feasible because the infants will test positive due to having their mother's antibodies -- even if they do not actually have HIV themselves.

Preliminary studies indicate PCR may be used to diagnose HIV infection in infants early after birth[15,16]. This may be one instance in which PCR is currently more useful than bDNA because of its lower limit for detection.

◆ *To predict transmission from mother to infant* - Another use of viral load may be its use to predict whether an HIV-infected, pregnant woman is likely to transmit HIV to her unborn infant. One study found that pregnant women with high HIV viral load were more likely to transmit HIV to their infants. The researchers suggested pregnant women with high viral load are especially strong candidates for anti-HIV therapy to reduce this viral load[17].

◆ *Other Uses* - Some researchers have speculated viral load in the cerebral spinal fluid may be useful to predict HIV-related neurological disorders; however, research has been contradictory[18]. These technologies can also be used to quantify hepatitis C[19] and CMV[20] in HIV-infected individuals. The use of PCR to quantify CMV in the blood may predict CMV retinitis and could be used to confirm a diagnosis of CMV retinitis made by examination by an ophthalmologist[20].

EVIDENCE SUPPORTING THE USE OF VIRAL LOAD AS A CLINICAL AND SURROGATE MARKER

At a recent American Foundation for AIDS Research Science of AIDS Seminar, Martin Markowitz, M.D. of the Aaron Diamond AIDS Research Institute outlined several criteria a laboratory test would have to meet to be an ideal clinical marker (see the June 1995 issue of *The Link - Update*)

Scientifically, the marker would have to be detectable in all patients. It would change rapidly in response to treatment. It would correlate with disease progression and clinical outcome. To be used as surrogate marker in clinical trials, the same criteria would have to be validated within the context of randomized studies.

While many would say the scientific evidence supporting the use of viral load measures as either a clinical or surrogate marker is incomplete, studies continue to accumulate data which support this use. For the purpose of brevity, the following information combines data supporting the use of viral load as a clinical or surrogate marker, regardless of whether PCR or bDNA was used.

A cross-sectional analysis involving 112 persons living with HIV found rapid and substantial changes in viral load as a result of initiating or withdrawing antiviral treatment[21]. This indicates viral load is a marker for drug activity.

Another study, in which early versus delayed treatment with AZT was evaluated, found the patient's baseline viral load predicted their clinical outcome. This indicates usefulness as a clinical marker for predicting an individual patient's prognosis. The study also found a decrease in viral load lasting for at least four months after initiating treatment predicted clinical outcome[22], which may indicate it could be used as a surrogate marker, replacing clinical outcome with a reduction in viral load.

The viral load of long-term non-progressors was "orders of magnitude" lower than would be normally expected.

In a large cohort study, several laboratory markers were studied to determine whether course of disease progression could be predicted. High viral burden, especially shortly after infection with HIV, was strongly predictive of HIV disease progression[23]. This could indicate that in the clinic, newly infected persons with high viral load may be especially strong candidates for early antiviral intervention.

A similar study in infants found high viral load in the first few weeks of life correlates with early onset of AIDS[24]. Another study of 62 men who had recently been exposed to and tested positive for HIV, found that the level of viral load during early infection strongly predicted progression to AIDS, independent of what occurred regarding CD4 cell count[25].

Other studies which could indicate viral load is correlated with disease progression and a potential clinical marker compared the typical profiles of persons with HIV who progressed to AIDS with long-term non-progressors (people who had been infected for at least 12 years but were healthy and had normal immune functioning.). The viral load of long-term non-progressors was "orders of magnitude" lower than would be normally expected[12, 26].

Similarly, a study of long-term AZT therapy[27] found low viral load predicts improved health status while high viral load predicts progression to AIDS.

Thus, the available evidence seems to support the use of viral load as clinical marker. Viral load would appear to be useful for predicting an individual patient's risk for progression. It would also appear to be useful in monitoring the long-term effectiveness of anti-HIV treatments by monitoring whether a treatment suppresses viral load over time. Increasing viral load during the use of any particular therapeutic regimen could indicate the need for a treatment change.

The evidence supporting substituting viral load for clinical endpoints in a clinical trial is less substantial. However, accumulating evidence indicates viral load predicts disease progression. The few trials in which reduction of viral load after initiation of treatment predicted clinical outcome offer intriguing signs that viral load may well prove to be a useful surrogate for clinical outcome.

If a few more, larger, randomized clinical trials indicate the effects of anti-HIV treatments on viral load predict clinical outcome, viral load may well become the new standard for evaluating HIV treatments in clinical studies.

For more information on viral load assays, see –

- *The upcoming Issue of the RITA! Newsletter.*
- *Treatment Issues, Volume 8, Number 10.*
- *AIDS Treatment News, Issue Number 211.*
- *The Link, July 1994 Issue.*

HYDROXYUREA AND ddI REDUCE HIV VIRAL LOAD

A study published in the September issue of the Journal of Acquired Immune Deficiency Syndromes and Human Retrovirology reports upon the results of a study conducted in France examining combination therapy with hydroxyurea and ddI[30].

The study was conducted in 12 patients with a CD4 cell count greater than 250 who had never taken antiretroviral therapy. The participants in the study were all asymptomatic. Their baseline viral load by PCR ranged from 3,521 copies to 128,973. Nine males and three females participated.

Patients were followed on therapy for 90 days. The combination of 200mg ddI twice daily and 500mg hydroxyurea twice daily appeared very safe. There were very few, relatively minor side-effects and no toxicity requiring drug withdrawal or dosage adjustment.

Six patients who had low viral load to begin with did not have detectable viral load after 90 days. The other six patients, who had high viral titers at the beginning, also had significant reduction of viral load after 90 days of treatment.

At the beginning of the study, the average CD4 cell count was 343. After 90 days, it was 541. CD4 cell changes from beginning to end of study ranged from an increase of 577 cells to a decrease of 69 cells. Only one patient had a decrease in CD4 cells.

The average increase in CD4 cells for the group as a whole was 120. For the subset of patients who did not have detectable viral load by the end of the study, the average CD4 cell increase was 244.

Preliminary results from two other studies printed in the abstract book for the upcoming *35th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC)* offer less complete information regarding ddI and hydroxyurea.

A yet to be completed study which examines 12 patients who are clinically stable, have not taken antiretroviral therapy in the previous three months and have a CD4 cell count of less than 400 will test 500mg of hydroxyurea twice daily for twelve weeks. After the twelve weeks, ddI taken at a dose of 200mg twice daily will be added.

Ten participants have been enrolled in this study thus far. Six have completed one month of hydroxyurea therapy. Adverse effects have been mild. CD4 cell counts in these six patients have fallen from an average of 181 to 143; however, CD4 percentage has remained unchanged. No viral load data is yet available[31].

Another small study, examining 200mg of ddI twice daily plus 500mg of hydroxyurea twice daily, was undertaken in six persons with an average CD4 cell count of 79[32]. Five participants had been on previous antiretroviral therapy, which was discontinued for 10 days prior to beginning the study. Five of the participants had never taken ddI.

After seven days on treatment, three of the six subjects had marked reductions in viral load by bDNA. All three were ddI naive. The other three subjects did not have a significant change in viral burden after seven days.

The data so far on hydroxyurea, especially from the French study, is encouraging and warrants further investigation of the drug. However, since these were not randomized, comparative trials, it is not possible to know if results are due to initiation of ddI, hydroxyurea or the combination of the two. Little is known regarding whether hydroxyurea has any anti-HIV activity used alone in humans. It is also not known whether 500mg twice daily is the optimal dose to use.

HCRN will shortly begin enrollment for a study examining hydroxyurea used alone at three different dosages (see the May, 1995 issue of *The Link*). The study will examine the safety of the three doses and compare their activity against HIV viral load (if any) as measured by bDNA assay.

Later in the year, HCRN will also participate in an AmFAR CBCTN study of varying doses of hydroxyurea and ddI combined in differing patient populations. Call 520-2000 for more information on these studies.

NEW REPORTS EXPAND SEEMING CONTRADICTIONS REGARDING AZT

Two reports in the latest issue of the *New England Journal of Medicine*, expand upon the data regarding AZT.

The first study reports upon the last part of ACTG study 019. This part of the study compared beginning AZT at a CD4 cell count above 500 to waiting until after the CD4 count fell below 500.

The study found no difference. Beginning AZT at a CD4 cell count above 500 did not delay progression to AIDS or improve survival[28].

A second study examined whether AZT was beneficial when started during acute HIV infection[29]. The study found AZT therapy improved CD4 cell counts and reduced infections; thus, it appeared to have benefit.

These studies may seem contradictory. However, they were done in much different patient populations. Also, as the authors of the first study point out, AZT has a relatively weak antiviral effect. When used against a virus which may take many years to cause disease even when untreated, it is not surprising that short-term beneficial effects may not translate into longer term benefit. Also, when testing a treatment in such high CD4 cell counts, very few people are likely to have disease progression regardless of treatment. This makes it is very difficult to find a treatment effect without a large number of patients, especially if it is a relatively small treatment effect.

Outlined below are the major clinical trials of AZT and a basic description of their results.

WHAT DOES THE RESEARCH SAY ABOUT AZT?

(Taken from Several "Lay-Langauge" Sources)

* **1987** -- Study of AZT verus placebo in persons with ARC or AIDS was stopped early because 19 people in the placebo group had died while only one taking AZT had.
* **1990** -- ACTG 019 study compared persons with no symptoms and a CD4 count less than 500 who got either AZT or placebo. Persons who received placebo were more than twice as likely to progress to AIDS as those who took AZT. A part of the study in CD4 cell counts higher than 500 continued.
* **1991** -- Nearly 1,000 persons with HIV were enrolled in a study comparing AZT to placebo in persons with a CD4 count greater than 400 who did not have symptoms. After two years, 38% of persons on placebo progressed to AIDS while only 20% of persons on AZT progressed. At a CD4 count between 500 and 750, 18% of those on placebo and 9% of those on AZT had progressed.
* **1992** --A French study confirmed benefit of AZT use in AIDS patients. Median survival in the group taking AZT was 21.2 months. In the untreated group, it was 9.6 months.
* **1992** -- A study conducted by the V.A. compared persons who began AZT early verus those that started AZT only after developing AIDS or having a CD4 count less than 200. The study found that AZT delayed the time it took to develop AIDS but did not result in a survival benefit.
* **1992** -- A U.S. cohort study found that persons who began AZT and a drug to prevent PCP before developing AIDS had a 57% reduced risk of death at 6 months and a 40% reduced risk at two years.
* **1992** -- The European Concord study compared starting AZT at 1,000 mg per day when there are no symptoms to waiting to begin AZT after symptoms or infections develop. During the study, persons in the delayed therapy group were allowed to switch to real AZT. A benefit to early AZT was found at one year; however, at three years no benefit was detectable.
* **1993** -- An European-Australian Collaborative study compared AZT to placebo in persons with greater than 400 CD4 cells. Persons on placebo were twice as likely to have disease progression as those taking AZT. The benefit from AZT lasted between two and one-half to three years. Survival was not studied.
* **1995** -- Another arm of the ACTG 019 study in which people began AZT when their CD4 count was above 500 were compared to people who waited to begin the drug found no difference in survival or progression to AIDS.
* **1995** -- A study comparing AZT to placebo in persons newly infected with HIV found benefit.