Antibody Test Emerges as Best Way to Find JC Virus in MS Patients

BY RICHARD ROBINSON

ARTICLE IN BRIEF
Two studies indicate that while viral DNA in urine or blood does not correlate with PML risk, the presence of antiviral antibodies does, results that are likely to lead to development of a commercially available test. A third study shows that the virus persists long after acute PML symptoms subside, and may account for some of the ongoing neurologic deficits seen in patients following an episode of PML.

For multiple sclerosis patients (MS) contemplating natalizumab therapy, determining whether they are infected with the JC virus is critical, since treatment increases risk of progressive multifocal leukoencephalopathy (PML) in patients infected with the virus. Currently there is no accepted test for viral presence, and thus no way to stratify patients for risk of PML.

Two studies in the September Annals of Neurology indicate that while viral DNA in urine or blood does not correlate with PML risk, the presence of antiviral antibodies does, results that are likely to lead to development of a commercially available test. Meanwhile, a third study in the same issue shows that the virus persists long after acute PML symptoms subside, and may account for some of the ongoing neurologic deficits seen in patients following an episode of PML.

As of July 2010, more than 60 patients taking natalizumab have developed PML. The risk increases with treatment duration, from 1/100,000 in the first year of treatment, to 127/100,000 in the second year, to 171/100,000 in the third year, with an average risk among all exposed individuals of approximately 1/1000.

“We give patients a general risk of one in a thousand,” said Richard Rudick, MD, director of the Mellen Center in the Department of Neurology at the Cleveland Clinic Foundation, and lead author on the DNA study. “But it would be really nice if we had a way to identify someone at higher risk of developing PML, because then we would avoid the use of this drug, or would increase the threshold for using the drug even when the disease is so active, we can’t control it any other way. On the other hand, if we had a test that would show the risk is vanishingly low, let’s say one in one hundred thousand, we would lower the threshold, and not worry so much about those patients.”

A large, though still debated, proportion of the population is infected with the virus. After infection, the virus remains latent in the kidneys, but can reactivate; when that occurs, replicated viral DNA can be detected in the urine and blood. But could this be a valuable screening tool in MS patients?

BLOOD, URINE ANALYSES
To explore that question, Dr. Rudick and colleagues analyzed almost 13,000 blood and urine samples from almost 1,400 patients involved in the major natalizumab clinical trials. Samples included those from baseline, placebo and active treatment, drug suspension, and drug resuming periods. Viral DNA from samples was detected using quantitative polymerase chain reaction, either a commercially available version sensitive to 50 copies per mL, or an ultrasensitive assay able to detect 10 copies per mL.

JC virus DNA was found in plasma in four patients using the commercial test, and two more using the ultrasensitive assay, about a half percent of the entire study population. None of these patients went on to develop PML. Conversely, none of the five patients in the studies who developed PML had detectable virus in the blood at any time before they developed the disease. Urine was an equally poor measure of viral risk: In the 224 patients whose urine was analyzed, one quarter were positive for the virus at baseline, and one quarter were positive after 48 weeks of treatment, but the virus-positive patients changed — in some the virus appeared, and in others it vanished.

“Using existing technology,” Dr. Rudick said, virus in the blood or urine doesn’t appear to be a sensitive way to monitor the risk for PML. “It doesn’t appear to be a sensitive way to monitor the risk for PML. So in an individual patient in your practice, that means that a positive test doesn’t tell you much about risk, and a negative test doesn’t tell you much about risk. If you are a patient, you don’t really want that test.”

PROMISING ANTIBODY TEST
An antibody-based test, on the other hand, appears to be more promising. Leonid Gorelik, PhD, and colleagues at Biogen Idec, Inc., developed an enzyme-linked immunosorbent assay (ELISA) to detect antibodies to the virus in blood. Samples from patients with a positive urine test for viral DNA reacted, on average, much more strongly than those with negative urine tests. The team used the weakest reaction of the urine-positive responses as a cut-off point: anything below that was considered antibody-negative. Responses 2.5 times as strong were considered definitely positive, while responses in between were further evaluated in a second assay.

Tests of patient samples from the natalizumab resuming study, called STRATA, indicated that 53.6 percent of patients were antibody-positive. The authors found that many urine-negative patients nonetheless had positive antibody tests, which, they noted in their study, was “consistent with the assumption that a urinary JCV DNA test is unlikely to detect all JCV-infected individuals.”

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In samples from 17 patients who went on to develop PML, 100 percent tested positive, 16 to 180 months before symptoms developed. These data “suggest that patients without detectable levels of anti-JCV antibodies may have a lower risk of PML compared to patients who have detectable antibodies,” they concluded.

Commenting on the Biogen Idec study, Dr. Rudick said: “The likelihood of 17 of 17 being positive when only half the general population is positive is very low. That suggests that the risk of developing PML when you are negative is vanishingly small. That test might be useful.”

Joseph Berger, MD, professor and chairman of neurology at the University of Kentucky College of Medicine in Lexington, who was not involved with these studies, told Neurology Today: “The data they present are pretty compelling. It is a very good study, but there are some issues.”

More must be learned about the viral life cycle, he said, particularly whether it is resident in brain as well as the periphery, he said, and whether any of the cases of PML are from newly acquired, rather than latent, infections. Either situation would show a negative antibody test.

Most importantly, he said, more patients need to be studied to confirm these results. Dr. Berger suggested doing a urine test as a screen, and then an antibody test in those with a negative result, with a double negative the only sure indication of low risk. “Until we are comfortable that the antibody test by itself is the be-all and end-all, it would be best to do both.”

**VIRAL PERSISTENCE**

Meanwhile, it is now apparent that the JC virus can persist for years after stopping natalizumab treatment, according to Eugene O. Major, PhD, senior investigator at the NINDS in Bethesda, MD.

Dr. Major analyzed CSF samples from 35 MS patients with PML, including 13 with three or more samples taken weeks to months after diagnosis. Eleven of the 13 underwent plasma exchange, and two immunoadsorption. All 13 experienced the immune reconstitution inflammatory syndrome (IRIS). Despite this, seven of the 13 had viral DNA in the CSF. “This tells us that whatever the immune response is in some of these individuals, it may not be adequate to completely clear the infection,” Dr. Major said.

The persistence of the virus in the CNS may have clinical implications, he suggested. “We've never had the opportunity to look at longitudinal post-PML CSF samples. I am hoping that one of the values of this paper will be to make neurologists aware of neurologic deficits in post-PML patients. Some of those deficits may not be attributable to MS, but to an ongoing, smoldering PML.”

**REFERENCES:**