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Antiretroviral drug treatment interruption in human immunodeficiency virus–infected adults: Clinical and pathogenetic implications for the central nervous system

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Interruption of antiretroviral treatment provides a well-defined 'experimental' paradigm to study the dynamics of central nervous system (CNS) infection and host responses in relation to those of systemic infection. We review our experience with 12 subjects (9 who were viremic and three with suppressed infection at baseline) followed longitudinally with serial lumbar punctures and neurological evaluations after stopping their antiretroviral treatments. All but two subjects exhibited an increase in cerebrospinal fluid (CSF) HIV RNA. Approximately half of the cohort developed a substantial, though asymptomatic, CSF lymphocytic pleocytosis with CSF counts rising to 30–60 cells/μL in five of the subjects. Subjects with higher CSF cell counts exhibited higher CSF HIV concentrations. We interpret the relationship of CSF HIV concentrations and pleocytosis in the context of a simple model of virus and cell exchange between blood and CSF. The proportionally greater increase in CSF HIV after treatment interruption indicates that CSF HIV infection is often more effectively suppressed by combination antiretroviral therapy than is systemic infection. *Journal of NeuroVirology* (2004) **10(suppl. 1),** 44–51.

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This review focuses on the central nervous system (CNS) implications of the strategy of stopping antiretroviral treatment—variously termed *structured, strategic*, or *supervised* treatment interruption (STI) and here simply as treatment interruption—and particularly on two aspects: its direct clinical implications and some of the pathogenetic issues raised by our experience with repeated cerebrospinal fluid (CSF) analysis in individuals choosing this therapeutic option.

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Treatment interruptions have had at least three clinical rationales: (1) enhancing protective immune responses against human immunodeficiency virus (HIV) by reexposing the host to higher-titer virus (autoimmunization); (2) improving the response to a new therapeutic regimen in those failing therapy by allowing wild-type, drug-susceptible virus to outgrow drug-resistant strains; and (3) sparing the patient from the side effects, rigors, or expense of daily treatment (Lori and Lisziewicz, 2001). In general, the value of stopping treatment in order to augment anti-HIV immunity and attenuate the natural history of HIV infection seems limited; only in the setting where patients have been treated very early after initial infection does treatment interruption appear to sufficiently enhance host responses (Oxenius *et al*, 2002; Rosenberg *et al*, 2000). Similarly, there is limited evidence that stopping a drug regimen during virological failure and allowing wild-type virus to overgrow the predominant resistant strain alters the longer-term response to a subsequent salvage regimen; the resistant viruses persist at low levels and often reemerge quickly if not susceptible to the new drugs (Deeks

et al, 2001; Hance *et al*, 2001; Izopet *et al*, 2002). Thus, the third rationale is the one that now most commonly leads a larger number of patients and clinicians to consider treatment interruptions. They hope that interrupting therapy and thereby reducing overall drug exposure will lessen the alterations of lipid metabolism and other unwanted consequences of treatment (Dybul *et al*, 2001; Havlir, 2002). This can also reduce the expense of therapy and provide a welcome break to the difficulty of adhering to daily multidrug treatment. These important overall issues have been reviewed in the references cited above and will not be considered here. Rather we will describe our own observations on the effects of treatment interruption as they pertain to neurological issues, emphasizing both the potential clinical implications and the investigative opportunity that treatment interruption affords in characterizing the dynamics of exchange of HIV and inflammatory cells between the blood and the cerebrospinal fluid (CSF).

Effects of treatment interruption on the CSF and neurological function

Although observations of CSF changes in individuals stopping therapy are limited and show some variability, they also reveal some common features. We will briefly summarize here some of our experience to date from studies of 12 individuals who underwent serial lumbar punctures (LPs) before and after stopping treatment. All of these subjects were studied under an investigative protocol approved by the University of California San Francisco Committee on Human Research; decisions regarding treatment interruption were made independent of the CSF study protocol. They were all men with a mean age of 44 years. The general methods used have been previously described (Staprans *et al*, 1999; Price *et al*, 2001). Phenotypic and genotypic resistance and replication capacity assays were performed by ViroLogic (South San Francisco, CA) (Petropoulos *et al*, 2000; Barbour *et al*, 2002).

Subjects were studied in two contexts, virological success (plasma HIV RNA levels <50 copies/ml while on treatment; $n = 3$) and virological failure (persistent plasma or CSF HIV RNA levels >500 copies/ml on stable treatment; $n = 9$) (Figure 1). Results related to the first five of the subjects studied during virological failure have been published (Price *et al*, 2001); we have extended this experience with three more failure subjects and three additional

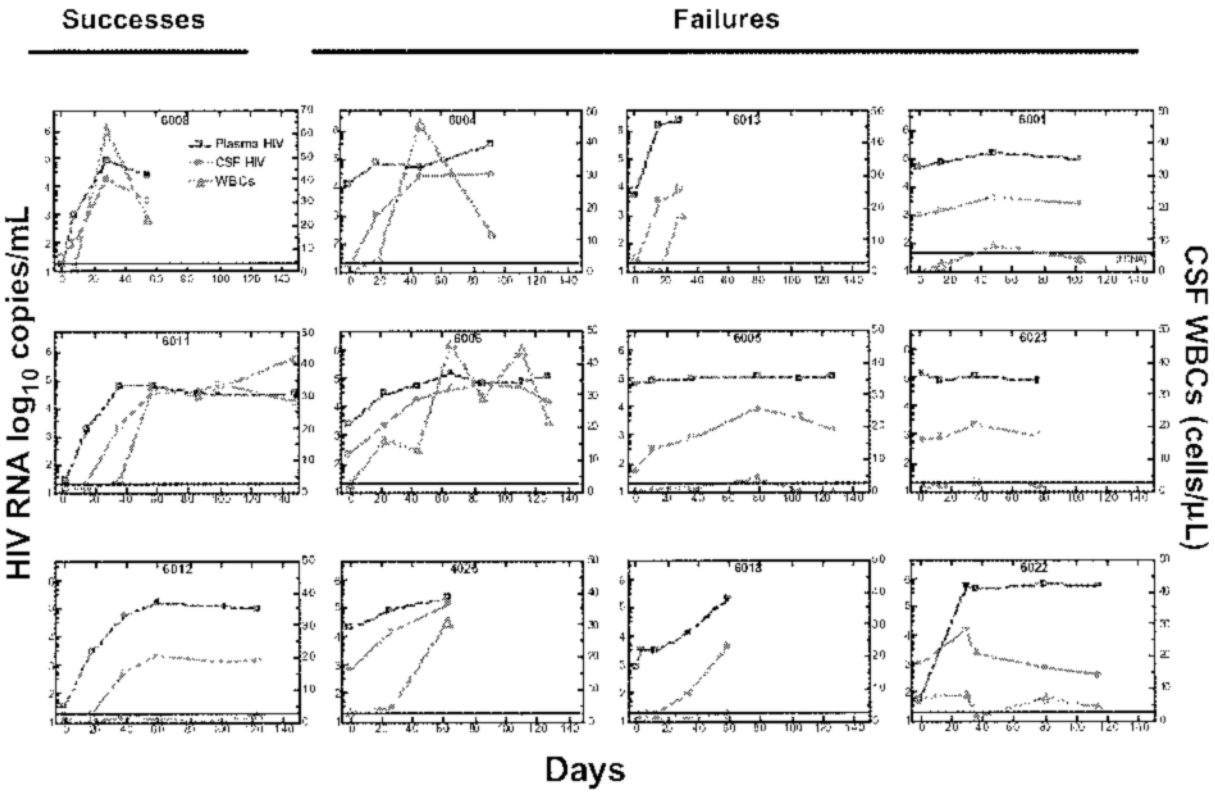

Figure 1 The course of changes in plasma and CSF HIV and in CSF WBCs during treatment interruption in 12 subjects. The three left panels depict the course in subjects in whom viral loads were below 50 copies/ml at baseline whereas the remaining nine panels show results in the subjects with baseline plasma or CSF viral loads above 500 copies/ml. Subject numbers are shown at the top of each panel and the symbols and lines are defined in the upper left panel. In all but one subject, HIV levels were measured using the Roche Amplicor PCR assay or its ultrasensitive modification and the lower limit of detection of 20 was used (and depicted as a dotted line in each panel); the exception was subject 6001 in whom the bDNA assay was used along with a lower detection limit of 50 copies/ml.

subjects with suppressed plasma HIV RNA levels before interrupting therapy.

All but two subjects exhibited an increase in CSF HIV RNA after stopping therapy; however, the timing and extent of this increase varied among individual subjects. Approximately half of the cohort developed a substantial CSF lymphocytic pleocytosis with CSF counts rising to 30 to 60 cells/μl in five of the subjects. In general, the extent of the pleocytosis correlated with the rise in CSF HIV RNA; those with the high CSF cell counts were also the subjects in whom the CSF viral load approached the levels of plasma virus. By contrast, those with little or no CSF cell increase exhibited lesser elevations in CSF HIV compared to plasma levels. The two subjects with little change in CSF HIV also showed no increase in plasma virus. Thus, these dynamic observations show an association between the rise in CSF cells and the increase in CSF HIV.

None of the subjects, either with or without pleocytosis, exhibited symptoms or signs of meningitis such as headache, photophobia, or stiff neck. Likewise, none complained of new or changing neurological symptoms or dysfunction, and none showed new abnormalities on examination. Quantitative neurological performance was assessed by a brief battery of tests and aggregated in the QNPZ-4 score, which is sensitive to changes in neurological capacity characteristic of acquired immunodeficiency syndrome (AIDS) dementia complex (ADC) (Price *et al*, 1999). This showed a slight overall improvement in group performance presumably related chiefly to practice effect (median increase in QNPZ-4 score of 0.24; intraquartile range −0.04 to 0.53 and overall range of −0.07 to 1.17). These data, although based on a small number of subjects, suggest that interrupting therapy has limited short-term neurological effect. The longer-term implications of cycling on and off therapy were not addressed.

CSF after treatment interruptions in virologically suppressed subjects

In the three subjects with viral suppression before treatment interruption, both the plasma and CSF HIV levels rose after a variable interval (Figure 1, left three panels). The increase in plasma HIV developed earlier than that in CSF in two (subjects 6011 and 6012) and showed a more rapid initial increment in the third (subject 6008). In the two subjects with pleocytosis, there was a similar temporal sequence with plasma HIV RNA levels rising first, followed by CSF HIV RNA levels and finally by a brisk pleocytosis. The subject without pleocytosis (6012) also showed a more blunted increase in CSF HIV. Indeed, subjects 6011 and 6012 provide an interesting contrast. The increase in plasma HIV in these two was similar in both magnitude and timing, yet they differed appreciably in the increase in the CSF HIV levels and in the appearance of increased cells in the CSF, emphasizing the interrelationship between CSF HIV and cell response. This contrast also highlights the question of what factors underlie the variability in CSF changes among individuals. To what extent are these differences associated with host genetics, with prior events in course of infection or with the character of the infecting virus?

CSF after treatment interruptions in patients with drug-resistant HIV (virological failure)

The nine panels to the right in Figure 1 show the plasma and CSF changes in the subjects with virological failure of combination antiretroviral therapy (eight with failure as detected in plasma and one unusual subject, 6022, with low plasma but elevated CSF HIV at baseline). In the four of these with robust CSF pleocytosis, this either was detected at the same time as (6006) or after (6004, 6026, and 6013) the increase in CSF HIV. In these four subjects, increases in plasma and CSF virus were detected over the same study interval as the appearance of elevated CSF cell counts. Because the CSF started at a lower level and approached or equaled plasma virus, the relative rise in CSF HIV was greater in three of these (observation of the fourth subject, 6013, was terminated early because of the very rapid increase in plasma virus). Two subjects (6005 and 6013) showed an increase in CSF HIV without pleocytosis, one without an appreciable change in plasma virus and the other with an earlier plasma rise, but thereafter parallel change in plasma and CSF HIV RNA levels. In two subjects (6001 and 6023), there was little or no appreciable change in plasma or CSF virus or in CSF cell count.

In the subjects in whom we previously reported results of antiviral resistance analysis (6004, 6006, 4026, 6005, 6001), the initial increase in CSF HIV RNA levels was predominated by resistant virus, and only subsequently supplanted by wild-type, susceptible virus (Price *et al*, 1999). This switch was detected during the same time interval during which wild-type also replaced resistant virus in the plasma (Deeks *et al*, 2001). In subject 6013, the increase in both plasma and CSF virus was predominated by quasispecies resistant to nnRTIs, without change in this resistance from baseline (baseline CSF resistance could not be studied due to the low virus titer, insufficient to amplify); presumably the study duration was too short for a switch to wild-type virus.

Two subjects (6018 and 6022) revealed unique patterns and warrant comment both as exceptions that emphasize the more common patterns and because they also may provide special insight into aspects of pathogenesis. In the case of subject 6018, baseline plasma showed high-level phenotypic and genotypic resistance to two of his drugs, 3TC and nevirapine, but susceptibility to his third drug, stavudine. Although at 33 days the plasma virus resistance was nearly identical to baseline, the now detectable CSF virus showed phenotypic susceptibility to all drugs. At the final sampling (59 days), viruses in both compartments were fully susceptible. Thus, less resistant

virus was detected first in the CSF. This observation is consistent with independent emergence of a more fit wild-type HIV in both plasma and CSF (when fitness is defined as the relative ability of the virus to replicate in the absence of drug). Theoretically, the wild-type variant was more readily detected in CSF because of the scarcity of the less-fit, drug-resistant variant.

Most individuals have higher levels of plasma HIV RNA, particularly when on treatment. However, one subject (6022) had notably lower plasma than CSF HIV RNA levels at baseline. During treatment interruption he developed a rapid and marked increase in plasma HIV RNA, but only a transient and much smaller increase in CSF HIV RNA. The plasma:CSF HIV ratio thus switched from a negative value to a high positive value during the period of treatment interruption. This unusual case contrasted with the other failure subjects in which this ratio narrowed or remained nearly equal during the period of observation. This observation illustrates that high virus in the plasma does not automatically 'spill over' into the CSF but rather that there are active processes determining HIV exchange or segregation between these two fluids. The results of phenotypic testing on this subject 3 months before the treatment interruption (not shown) indicated that plasma virus (concentration of 2.26 $\log_{10}$ copies/ml) had moderate resistance to one of his baseline medications, delavirdine, but no significant resistance to his other two medications, zalcitabine and ritonavir/lopinavir. The replication capacity of the plasma virus was 32% (the replication capacity assay used in these studies measures the relative ability of a recombinant virus containing the patient-derived *pol* and *gag* sequences to replicate *ex vivo* in the absence of therapy; the read-out for this assay is percent of a wild-type reference where a number less than 100% implies impaired replication capacity, or "fitness" (Huang *et al*, 2003)). Notably, CSF virus at this time (concentration of 3.39 $\log_{10}$ copies/ml) did not show this resistance and had 98% replication capacity. Subsequently, at the baseline interval, plasma virus was too low to amplify, whereas CSF virus now showed 4.2-fold resistance to delavirdine, though again no resistance to his other medications; however, replication capacity was not reduced (126%). Subsequent plasma and CSF samples during the treatment interruption showed loss of the phenotypic nnRTI resistance and emergence in plasma of a variant with high replication capacity (121% to 160%). Thus, the initial CSF 'failure' in the face of relative plasma suppression was likely due to poor drug penetration (none of the antiretroviral medications in his unusual regimen affords good CSF penetration) with inability to suppress a relatively fit virus compared to that of plasma which was either more (prior to interruption) or similarly (at the time of interruption) resistant but much less fit. After reversion to more wild-type virus, both compartments seemingly approached an equilibrium ('set point'), with a high level in plasma but a lower level in CSF, indeed a level similar to that at and before baseline.

Some clinical implications of changes in CSF after treatment interruption

Our observations of CSF after treatment interruption show that CSF pleocytosis is common, occurring in half of our subjects. Diagnostically, this means that when patients stop therapy and are found to exhibit a CSF lymphocytic pleocytosis, one need not invoke a secondary infection causing meningitis—rather this increase in cells is likely incidental to their coming off therapy. Although this same diagnostic consideration is also more broadly true for individuals who are not being treated with combination antiretroviral therapy (ART), the cell response may be emphasized in this setting and perhaps more readily misinterpreted.

These results also provide preliminary reassurance that short-term treatment interruption is usually neurologically benign. Despite the surge in CSF virus and the white blood cell (WBC) reaction, our subjects remained neurologically asymptomatic and without detectable CNS functional change. Hence, the increase in CNS virus does not appear harmful, at least in the near term. There are several important caveats regarding this tentative conclusion. First, our studies were for the most part of limited duration, and longer-term cessation of treatment might have greater neurological impact, although once subjects are off ART, they are similar to those without treatment in whom the likelihood of ADC depends upon more than the CSF viral load. Second, with one exception, our subjects were neurologically normal without prior evidence of ADC. For those with ADC, resurgence of viremia and CNS infection might be more problematic, and stopping treatment should be undertaken with caution. Third, our subjects were studied when their blood CD4+ counts were preserved and above the range of high susceptibility to ADC; stopping treatment is likely to be more hazardous generally, and to the CNS in particular, when systemic immune defenses are weak. Finally, the study sample size is clearly too small to confidently predict that treatment interruption is always neurologically safe.

To the extent that CSF HIV reflects or parallels brain infection (Price and Staprans, 1997), these studies show that systemic therapy can effectively suppress CNS virus even in the presence of drug resistance. In the 'virological success' subjects, both plasma and CSF virus populations usually rose rapidly after a delay, indicating that the drug regimens had effectively suppressed infection in both compartments. More interestingly, in the virological failure subjects, the CSF viral loads at times rose proportionally higher than those in plasma, indicating *more effective suppression* in this nervous system compartment than systemically. This finding was contrary to expectation

and runs counter to the frequently cited hypothesis that CSF infection will be more difficult to treat than systemic infection because of limited drug penetration. The superior suppression of CSF compared to plasma virus among patients with drug-resistant HIV agrees with our recent cross-sectional data showing that CSF HIV RNA levels are relatively lower in failing subjects than in untreated subjects in the face of similar plasma HIV RNA levels (unpublished). Thus, with respect to CSF infection, the *virological failure* subjects may better be regarded as *partial virological successes*. Subject 6023, showing higher CSF virus at baseline, is a notable exception to this 'rule' of superior CSF suppression, and the greater CSF infection in his case was not due to compartmentalized local resistance, but seemingly to inadequate local drug effect. His case argues that poor drug penetration and resultant insufficient local treatment effect can indeed be important in determining CNS treatment outcome in some, likely a minority, of patients.

Pathogenetic implications and speculations

These observations regarding the changes in CSF after treatment interruption underscore a number of questions regarding the pathogenesis of nervous system infection centered on the interchange and compartmentalization of infection and host inflammatory responses between blood and CSF (and, by implication, the brain). They show that there is a relationship between the magnitude of CSF HIV concentration and the CSF WBC response, though they do not immediately explain the mechanisms governing this relationship.

We have previously outlined alternative simple models for this relationship (Price *et al*, 2001). Figure 2 presents an extension of these models,

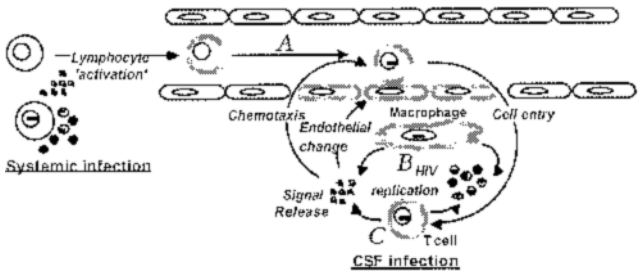

Figure 2 Models of CSF HIV infection and pleocytosis. The schematic depicts elements of systemic infection, CNS blood vessel with barrier endothelium, and infection within the CNS sustained by perivascular macrophages or CD4+ T lymphocytes. The three types of infection are described in the text, and include A, Push involving activation of T cells facilitating their entry into the CNS where a small minority of activated CD4+ cells support replication and release virus; B, pull involving locally resident infected cells, likely chiefly macrophages, producing HIV and also initiating signals that provoke inflammatory cell entry (chemokine attraction and endothelial alterations); C, amplification in which local autonomous infection is sustained by entering infected and uninfected T cells after initiation by either push or pull.

which now hypothesizes three 'types' of CSF infection, two primary and a third secondary, as related to CSF lymphocyte response:

A. *Push* whereby peripheral lymphocyte activation leads to an increase in entry of these cells into the CSF (and the brain perivascular spaces which can communicate with CSF). If some of these cells are infected (and activated) they will be capable of releasing virus into the CSF. This is infection *from without* and largely determined by systemic viremia and its effects on these leukocytes. It predicts that predominating viruses in the blood and CSF will more likely be identical when this is the major determinant of CSF infection.
B. *Pull* in which infection within the meninges (or adjacent perivascular spaces and brain) is the origin of CSF virus and secondarily attracts an inflammatory response. This infection *from within* is governed by local factors that may diverge from some of those determining systemic infection. Predominating viruses in the CSF are therefore more likely to differ from those in plasma. In this case, the lymphocytes found in the CSF will be reactive and not an appreciable source of fresh virus. Note that in this "pull" model it is possible that the blood and CSF infection might remain completely independent; in the setting of treatment interruption, both may be released from drug pressure and exhibit independent cycles of viral replication.
C. *Amplified* infection combines push and pull in a positive feedback loop. Once infection is initiated in the CSF by whichever of the above two primary mechanisms, local replication may add virus directly to the fluid and also recruit both infected and uninfected cells from blood, thereby bringing in viruses circulating in the blood as well as activated target cells that can then serve as substrate for further replication and increase in the local virus concentration. When amplification brings together a mixture of viruses, the more fit of these will eventually predominate.

Each of these mechanisms presumes that HIV enters the CSF within cells, either T lymphocytes or monocytes (evolving *in situ* to macrophages). Direct entry of cell-free virus (Liu *et al*, 2002) requires modifications of these models, but might involve similar push or pull mechanisms; for simplicity these are omitted from discussion.

The timing of events suggested by observations in two of the suppressed subjects (increase in plasma HIV, followed by increase in CSF HIV, and only subsequently by increased WBCs) and in three of the failure subjects (simultaneous increase in plasma and CSF, but again a delay in the initial increase in cells) might be interpreted to favor the pull hypothesis to explain the increase in cells after independent 'release' of virus in CSF. However, we are not wholly certain that measurements of the WBCs in the 'normal'

range are sensitive enough to detect a low level of cell influx that is still sufficient to bring the initial virus increment into the CSF from blood. Indeed, the earlier rise in the blood HIV than CSF HIV might argue for the push hypothesis, with early changes in immune cell activation and cell entry. Hence, our results might still be interpreted as compatible with either of the two primary hypotheses. Even in some of the subjects without abnormal CSF cell counts, there is suggestion of a small 'bump' in these counts.

The later development of more robust pleocytosis in those subjects in which the CSF HIV level approaches or equals that of plasma may signal the involvement of the amplification mechanism whereby there is both pull and push, leading to 'equilibration' between the two compartments, usually with wild-type, more fit virus. By contrast, in those without pleocytosis where CSF virus remains 10-fold or more below that of plasma, this amplification may be less active or absent.

How are these important mechanisms to be dissected further? Careful phylogenetic analysis of rebounding HIV in CSF and plasma may provide some insights into the relative importance of these proposed mechanisms. Viral strains would be expected to be similar or identical if the source of HIV in CSF is the peripheral blood—in the "push" model or after amplification mechanisms have superceded the initial events. In contrast, strains would more likely be discordant in the two compartments if removal of drug pressure results in independent increases in viral replication. The differences in resistance patterns of the viruses in plasma and CSF of subject 6018 at day 33 discussed above may argue for such an independent origin and early replication in the two compartments; in this case, there was no appreciable pleocytosis. On the other hand, analysis of both phenotypic and genotypic resistance patterns in the other subjects suggests that in the setting of treatment interruption plasma HIV and CSF HIV usually exhibit similar if not identical resistance, particularly after reversion to wild-type. This more general finding supports either the push or amplification models outlined here, though results of ongoing more detailed phylogenetic analysis need to be performed to address this more rigorously.

Potential CNS implications of continued therapy during "virological failure"

A premise of our work with CSF is that changes in this fluid either directly reflect (by diffusion from the interstitial or perivascular spaces into the CSF) or parallel (because of similar barriers to drug, virus and cell movement) changes in the brain, and that careful measurements of CSF may provide insights in HIV-mediated CNS neuropathogenesis (Price and Staprans, 1997). The mechanisms underlying the pathogenic effect of HIV on brain function remain to be precisely understood. As described in studies reviewed elsewhere in this symposium, experimental data using *in vitro* models and nonhuman primates suggest that HIV infects brain perivascular and other macrophages (Williams *et al*, 2001) and that such infection initiates a cascade of events leading to production of virus- and cell-coded signal and neurotoxic factors with resultant indirect injury of noninfected neural cells. Although the relative roles of various viral products, chemokines, and immunological reactants remain to be determined, it is important to keep in mind that HIV is the primary *driver* of this pathology, and hence the principal target of both secondary prophylaxis and therapy. To the extent that viral findings in CSF reflect those in brain, the results in this, albeit limited, sample suggest that combination antiretroviral therapy is generally effective in suppressing this potentially 'sequestered' infection, even when such therapy is unable to suppress other major sources of virus contributing to continued viremia. This likely explains the marked decline in ADC that characterizes the current era of HIV therapy in the developed world, although there remains some controversy over whether ADC incidence has been reduced to the same extent as the major opportunistic infections (Dore *et al*, 1999; Sacktor *et al*, 2001).

We are not yet certain why suppression of CSF infection appears to exceed that of plasma infection in most of those failing therapy. Reduced viral fitness of resistant virus in these individuals may be one factor. However, because fitness or replication capacity is measured in a lymphocyte-based assay (Petropoulos *et al*, 2000), the applicability of this assessment to CNS infection of macrophages remains uncertain. Further studies will need to examine fitness as it relates more definitely to replication in cells within the CNS. Some component antiviral drugs, particularly some of the nRTIs may be more effective in suppressing CNS virus than simple measurement of CSF drug levels suggest. More broadly, this is an area where the relationships of viral genotypes and viral physiology need to be more clearly understood.

Conclusions

Treatment interruption provides a well-defined experimental setting to study the dynamics of CSF infection and host responses in relation to those of systemic infection reflected in the blood. This is facilitated by the ease and low (although not absent) morbidity of repeated lumbar puncture. CSF changes rapidly in parallel with the blood, either reflecting direct exchange between these two fluids or similar processes in the respective compartments. Interruption of antiretroviral therapy likely recapitulates features of the initial seeding of the CSF during primary infection, although somewhat modified by previous host exposure and altered host responses. The accompanying pleocytosis provides evidence that the elevated CSF WBC counts recognized since the beginning of

the epidemic .(Appleman *et al*, 1988) do indeed indicate early and continued HIV infection of the CNS. This pleocytosis and the resurgence of HIV in CSF emphasize the virtually universal CNS exposure to HIV among those who are infected, an exposure that may eventually result in chronic encephalitis and ADC. Among the reassuring findings in our studies and others is that CSF HIV is usually suppressed by combination antiretroviral therapy, even without theoretically 'good' penetration of all the drugs in a regimen (though not always as illustrated by subject 6023), and often even when systemic infection is incompletely suppressed.

Before one is completely reassured by findings such as these showing that patients experiencing incomplete viral suppression as assessed in sampled blood appear to derive continued virological benefit in CNS, further studies are needed to confirm that this effect will be sustained over the longer term. In contrast to our general findings, it is still possible that in the longer term on-going viral replication in the presence of therapy may result in continued or enhanced viral evolution and the emergence over time of viral strains that are able to replicate effectively in the CNS even in the presence of drug. Although this has not yet been clearly shown to predispose to the development of ADC, careful longitudinal studies of patients with limited treatment options constrained by viral resistance will be needed to test this further. It also remains uncertain whether the pathogenic potential of drug-resistant variants in CSF (and by extension the CNS) is always predicted by the concentration of virus detected in samples obtained by lumbar puncture. In theory, maintaining a partially effective regimen may prevent progression to AIDS because of stable or increasing peripheral CD4+ T-cell counts and attenuation of the pathogenicity of circulating viruses. On the other hand, escaping viruses, despite representing only minority populations (in plasma or CSF or within circulating cells and undetected in either fluid), could still result in ongoing CNS injury.

That highly active antiretrovirus therapy (HAART) prevents or delays onset of neurological dysfunction is clear (Sacktor *et al*, 2001; Dore *et al*, 2002). What remains unclear is whether this benefit is related to a treatment-mediated decrease in viremia, difference in replication capacity and virulence, treatment-mediated increase in immunologic function, or all of these factors in concert. Careful analysis of changes in the quantity and character of viruses over the short term during treatment initiation and interruption and over the longer term of longitudinal cohort study should lead to a greater understanding of the ecology of HIV and the mechanisms contributing to its capacity for causing neurological morbidity and mortality.

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