

# Topics in HIV Medicine®

A publication of the International AIDS Society–USA

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Update of the Drug Resistance Mutations in HIV-1: 2007 119

*International AIDS Society–USA Drug Resistance Mutations Group*

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## About This Issue

This issue features 4 *Perspectives* articles taken from presentations given at recent International AIDS Society–USA Continuing Medical Education courses, as well as a *Review* article and a special contribution.

Bruce D. Walker, MD, discussed elite control of HIV infection and implications for vaccines and treatments at the New York course in March 2007. Daniel Douek, MD, PhD, offered a discussion on HIV disease progression and the role of CD4 + T lymphocytes in the gut, as presented at the San Francisco course in May 2007. Joel Palefsky, MD, discussed human papillomavirus and associated anogenital disease in HIV-infected men and women, as presented at the Chicago course in May 2007. Also from the Chicago course, Eric S. Daar, MD, presented a review of issues surrounding incorporating novel virologic tests into clinical practice.

This issue also contains a review article written by Erika Z. Aaron, MSN, CRNP, and Shannon M. Criniti, MPH, which outlines issues in preconception health care for HIV-infected women. Finally, in a special contribution, the International AIDS Society–USA Drug Resistance Mutations Group has updated the current list of HIV-1 mutations associated with antiretroviral drugs and the accompanying footnotes.

## Topics in HIV Medicine®

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**Perspective****HIV Disease Progression: Immune Activation, Microbes, and a Leaky Gut**

*Recent findings indicate that the majority of all CD4+ T lymphocytes are lost during acute HIV infection, with mucosal compartments being most severely affected. The frequency of infection is very high in gut CD4+ T cells, and depletion of these cells persists into the chronic phase of infection. Infection is associated with increased gut permeability, with microbial translocation being evidenced by increased circulating lipopolysaccharide (LPS) levels. Plasma LPS levels correlate with systemic immune activation, which drives chronic HIV infection. Antiretroviral therapy reduces plasma LPS, and greater CD4+ T cell reconstitution is associated with lower LPS levels. These findings have a number of implications for therapeutic strategies. This article summarizes a presentation on HIV disease progression made by Daniel Douek, MD, PhD, at an International AIDS Society–USA Continuing Medical Education course in San Francisco in May 2007. The original presentation is available as a Webcast at [www.iasusa.org](http://www.iasusa.org).*

In its natural hosts (eg, sooty mangabeys, African green monkeys, chimpanzees), simian immunodeficiency virus (SIV) does not cause AIDS, other disease, or immunodeficiency, even in the presence of high viral loads. However, SIV infection in rhesus macaques—ie, an “unnatural” infection—results in rapid progression to AIDS, despite lower viral loads than in “natural” infection; similarly, SIV adapted to humans also causes rapid progression to AIDS. The mechanism for the progression to AIDS in HIV infection is depletion of CD4+ T lymphocytes, the target cells of the virus, with progression being clearly linked to a decrease in peripheral blood CD4+ cell numbers. In a series of studies investigators have sought to characterize how, where, and when HIV infection causes CD4+ T cell depletion, and to better understand how mechanisms underlying the disease process might differ in natural and unnatural infection.

**How, Where, and When?**

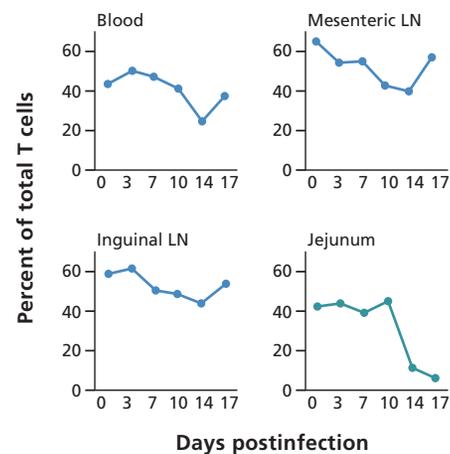
In the traditional view of HIV disease course, acute infection is accompanied

by a rapid transient decrease in peripheral blood CD4+ T cell count and a rapid partial recovery of this loss, with chronic infection being characterized by a gradual and profound decline in CD4+ T cell numbers. Thus HIV infection has been thought of as a relatively indolent disruption of CD4+ T cells eventually leading to collapse of immune function. This notion has been largely based on measurements of CD4+ T cell counts in peripheral blood.

In studies over the past several years, Dr Douek’s laboratory and others have shown that with HIV and in macaque and SIV infection, the earliest targets of infection are mucosal memory CD4+ T cells, which bear the CCR5 HIV coreceptor and which constitute the majority of CD4+ T cells. The greatest numbers of mucosal memory CD4+ T cells (indeed, the majority of all T cells) are found in the gastrointestinal (GI) tract, with this compartment harboring possibly 80% of the entire T cell population. In studies assessing mucosal CD4+ T cell depletion in acute macaque SIV infection in blood, mesenteric and inguinal lymph nodes, and the jejunum, loss was virtually complete in the GI tract within 17 days of infection, representing a profound loss in memory T cell population given the concentration of these cells in this compartment (see Figure 1). As can be seen by comparing the graphs for

peripheral blood with jejunum, neither the degree nor tempo of memory cell loss in the GI tract, the major reservoir of infected cells, could be predicted by the cell loss profile in the blood.

As shown in Figure 2, flow cytometric analysis in humans indicates an abundance of CCR5+ CD4+ T cells in the gut through which HIV could readily propagate. In a study in gut (terminal ileum) biopsies from more than 50 individuals with or without HIV infection, massive depletion of CCR5+ CD4+ T cells from the gut was found in HIV-infected patients (Figure 3, left). In HIV-infected patients, gut memory cells were more frequently infected than were peripheral blood memory cells, with the frequency differing by 10-, 100-, and occasionally as much as 1000-fold in individual comparisons (Figure 3, right). These findings, indicating both that the major reservoir of target and infected cells is the GI tract and that there is rapid and profound loss of cells in acute infection, provide a new model for HIV disease course (see Figure 4). On this model, the bulk of CD4+ T cell loss occurs within the first 2 to 3 weeks of acute infection.



**Figure 1.** CD4+ T cell depletion in peripheral blood, mesenteric lymph nodes (LN), inguinal LN, and jejunum after simian immunodeficiency virus infection of macaques.

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Daniel Douek, MD, PhD, is Chief of the Human Immunology Section in the Vaccine Research Center of the National Institute of Allergy and Infectious Diseases in the National Institutes of Health.

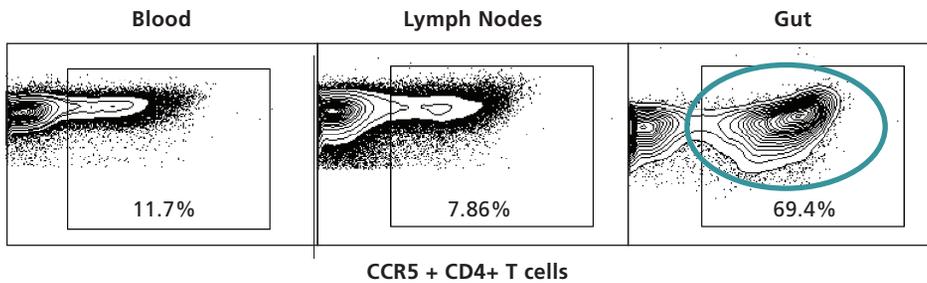


Figure 2. Flow cytometry results showing concentrations of CCR5+ CD4+ T cells in peripheral blood, lymph nodes, and gut in humans. There is an abundant supply of CCR5+ CD4+ T cells in gut through which HIV can rapidly propagate. Adapted from Brenchley et al, *J Exp Med*, 2004.

### Why Is HIV Disease Progressive?

The question then arises as to why there is progressive loss of CD4+ T cells beyond acute infection. To answer this question, aspects of immune activation in chronic HIV infection were investigated. Systemic immune activation has both beneficial and harmful effects in chronic infection and is in fact a strong predictor of disease progression, with recent findings indicating that it is a stronger predictor than peripheral plasma HIV RNA level. Immune activation in chronic HIV infection includes polyclonal B cell activation, increased turnover of T cells, a high frequency of “activated” phenotype T cells, and increased levels of cytokines, chemokines, and other proinflammatory mediators. The “good” effects of activation include restoration of memory CD4+ T cells and immunocompetence. The “bad” effects include lymph node fibrosis, retention of effector T cells in lymph nodes, thymic dysfunction, clonal exhaustion, drainage of memory T cell pools, and generation of more targets for HIV that permit ongoing HIV replication. What has been largely unclear is what is driving ongoing systemic immune activation in chronic HIV disease.

Given the massive depletion of memory T cells in the gut, microbial translocation from the gut was speculated to be involved in driving immune activation. In this process, gut-derived microbes or microbial products translocate to the systemic circulation in the absence of overt bacteremia. Microbial translocation is observed in

numerous settings, including graft versus host disease, inflammatory bowel disease, and gut surgery, and is correlated with systemic immune activation in some of these conditions. Enteropathy associated with HIV disease was initially reported as early as 1984 (Kotler et al, *Ann Intern Med*, 1984), with a number of other studies in subsequent years showing the presence of enteropathy, malabsorption, and increased intestinal permeability. It is now recognized that individuals with HIV infection can have the greatest increase in gut permeability among many of the GI epithelial barrier pathologies. Thus, both immunologic and structural defects in the GI tract in HIV infection can contribute to microbial translocation.

Microbial translocation can be quantified by measuring plasma levels of lipopolysaccharide (LPS; ie,

endotoxin). Plasma LPS levels were measured in approximately 300 subjects without HIV infection or with acute or early HIV infection, chronic infection, or AIDS (on the basis of CD4+ cell count <200/ $\mu$ L), with none of the subjects having any evident active infections other than HIV infection. Plasma LPS levels in HIV-seronegative subjects were similar to those in patients with acute or early HIV infection; however, patients with chronic HIV infection and those with AIDS (together termed “progressors”) had statistically significantly higher LPS levels than either HIV-seronegative subjects ( $P < .0001$  for both) or patients with acute or early HIV infection ( $P < .0001$  for both). A prior study by Suffedini and colleagues in which noninfected volunteers received injections of LPS showed that estimated plasma levels of as low as 14 pg/mL produced systemic immune activation measured as increased levels of inflammatory cytokines such as tumor necrosis factor, interleukin (IL)-1 receptor antagonist, IL-6, and IL-8 (Suffedini et al, *J Infect Dis*, 1999). In Dr Douek’s study, the median plasma LPS level in patients with progressors was 75 pg/mL, sufficient to stimulate systemic immune activation. That the source of circulating plasma LPS in HIV-infected individuals is predominantly the gut is suggested by studies showing dramatic reductions in LPS

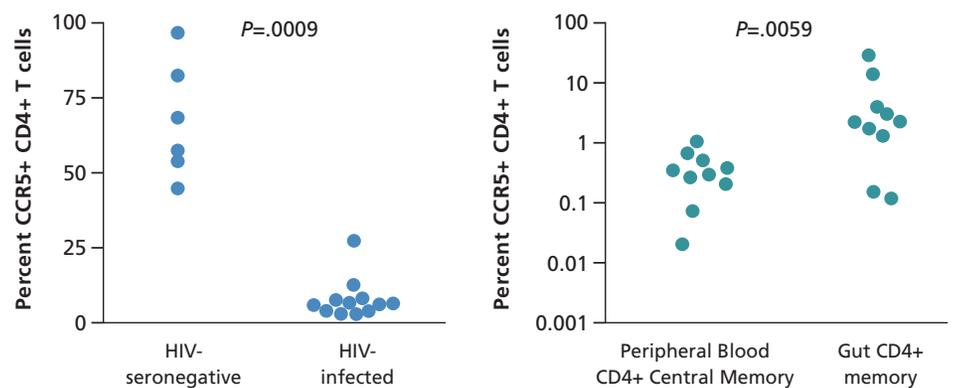


Figure 3. Left: Percentage CCR5+ CD4+ T cells in gut biopsies from HIV-seronegative and HIV-infected subjects. Right: Percentage of HIV-infected CD4+ T cells in the peripheral blood and in the gut. Gut biopsies are from more than 50 individuals. CCR5+ CD4+ T cells are massively depleted from the gut. There is a high frequency of infected cells at all stages of disease. Adapted from Brenchley et al, *J Exp Med*, 2004.

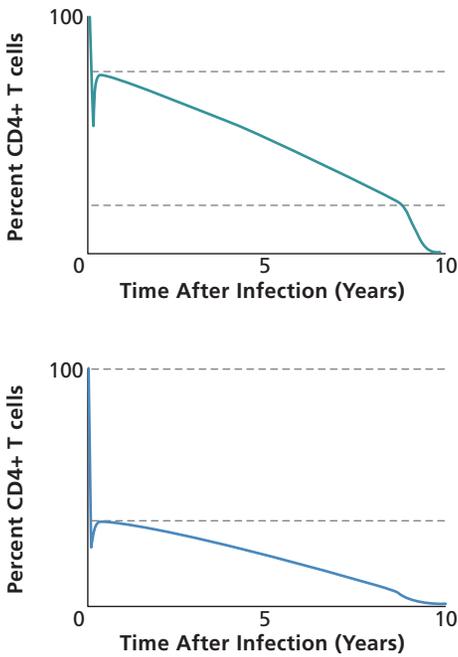


Figure 4. Top: “Old” view of HIV disease course. Blood CD4+ T cell counts: CD4+ T cell depletion occurs slowly over the course of the disease. Bottom: Revised view of HIV disease course, showing massive depletion of CD4+ T cells in acute infection.

levels in SIV-infected monkeys given large doses of “gut-sterilizing” antibiotics. Evidence that chronic LPS stimulation is occurring in HIV infection was then provided by studies showing substantially increased levels of soluble CD14 (sCD14) in plasma; LPS-stimulated monocytes secrete sCD14 and shed surface CD14. Both patients with acute or early HIV infection and those with progressive infection had markedly higher plasma sCD14 levels than HIV-seronegative subjects ( $P < .0001$  for both), indicating chronic in vivo LPS stimulation of monocytes and macrophages (Brenchley et al, *Nat Med*, 2006).

In these studies, LPS and LPS stimulation have been used as a marker for any immunostimulatory product that might be translocated from the gut to the circulation. Since LPS does not stimulate T cells directly, Dr Douek and colleagues investigated whether LPS levels correlate with other measures of non-LPS-mediated immune activation. An increased frequency of CD38+ HLA-DR+ CD8+ T cells is known to

occur in HIV infection and is associated with disease progression. It was found that the percentage of these cells correlated substantially with plasma LPS in the HIV-infected cohort (correlation [ $r$ ] = .5553;  $P = .0059$ ), indicating that other immunostimulatory factors are directly or indirectly activating these CD8+ T cells. Interferon alfa (IFN- $\alpha$ ) is produced by dendritic cells, which also are not stimulated by LPS. It was found that plasma IFN- $\alpha$  levels also correlated substantially with plasma LPS level in the cohort ( $r = .6244$ ;  $P < .0001$ ), again indicating that other immunostimulatory factors apart from LPS are active in HIV infection.

It is known that immune activation decreases with potent antiretroviral therapy, although it shows a much slower decline than does HIV RNA level and remains elevated above normal for at least a year after starting treatment. In a group of 28 HIV-infected patients, 24 had statistically significant decreases in plasma LPS after 48 weeks of full HIV RNA suppression on initial antiretroviral therapy ( $P = .0107$  for change from baseline in entire group). However, levels in the HIV-infected patients remained substantially higher than those in HIV-seronegative subjects ( $P = .0026$ ). As shown in Figure 5, CD4+ T cell count change was inversely correlated with LPS level after 48 weeks in these antiretroviral-treated patients ( $r = -.4628$ ;  $P = .0151$ ), suggesting that improved mucosal immunity reduces microbial translocation.

As noted, “natural” SIV infection is nonpathogenic and is associated with low immune activation despite high viral load. To determine whether such “natural” infection is associated with reduced microbial translocation, plasma LPS was measured in uninfected and SIV-infected macaques and in uninfected and SIV-infected sooty mangabeys. Whereas SIV infection was associated with a statistically significant increase in LPS in macaques ( $P = .002$ ), there was no difference in levels between uninfected and SIV-infected sooty mangabeys ( $P = .975$ ), suggesting absence of microbial translocation in “natural” infection.

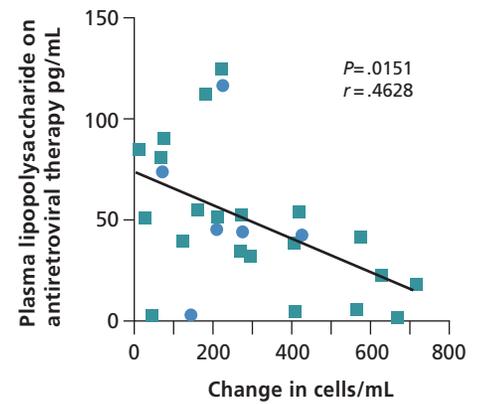


Figure 5. Inverse correlation of change in CD4+ T cell count and plasma lipopolysaccharide after 48 weeks of suppressive antiretroviral therapy. Immune activation decreases with antiretroviral therapy, but remains elevated above normal for at least 1 year. Adapted from Brenchley et al, *J Exp Med*, 2004.

## Summary and Implications

With HIV infection, the majority of all CD4+ T cells are lost during the acute phase of infection, with mucosal tissues being most severely affected. The frequency of infection is very high in gut CD4+ T cells, and depletion of these cells persists into the chronic phase of infection. Infection is associated with increased gut permeability and decreased enterocyte functionality, with increased circulating LPS levels indicating the occurrence of microbial translocation from the gut. Plasma LPS levels correlate with activation of innate and adaptive immunity in HIV infection, with suppressive antiretroviral therapy resulting in reduced plasma LPS levels and greater CD4+ T cell reconstitution being associated with reduced LPS levels. Measurement of LPS indicates that microbial translocation does not occur in nonpathogenic SIV infection.

These findings indicate that acute HIV infection is a very different disease state from chronic infection. Acute infection is characterized by massive and rapid CD4+ T cell loss, whereas chronic infection is characterized by persistent immune activation that drives viral replication and further CD4+ T cell depletion. The findings further indicate that the integrity of the

mucosal barrier is a paramount factor in disease progression.

Antiretroviral therapy is currently the best way to protect the gut and prevent microbial translocation and reduce chronic systemic immune activation. The ways in which these findings may alter our approach to treatment include perhaps changing the concept of “early” therapy to mean hours or days after exposure to HIV rather than weeks or months. Strategies for achieving early reduction of target cell infection need to be pursued—eg, by using microbicides applied mucosally. Further, pre-exposure and postexposure prophylaxis, if practically feasible, could be highly effective ways to prevent infection. Therapies to improve gut immune reconstitution (eg, cytokines) and to at-

tenuate mediators of inflammation (eg, antiseptics agents) could be pursued. In addition, there should be increased emphasis on the development of vaccines to prevent or reduce CD4+ T cell depletion at mucosal surfaces.

*Presented by Dr Douek in San Francisco in May 2007. First draft prepared from transcripts by Matthew Stenger. Reviewed and edited by Dr Douek in August 2007.*

*Dr Douek has no relevant financial affiliations to disclose.*

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### Suggested Reading

**Brenchley JM**, Price DA, Schacker TW, et al. Microbial translocation is a cause of systemic immune activation in chronic HIV infection. *Nat Med.* 2006; 12:1365-1371.

**Brenchley JM**, Schacker TW, Ruff LE, et al. CD+4 T cell depletion during all stages of HIV disease occurs predominantly in the gastrointestinal tract. *J Exp Med.* 2004; 200:749-759.

**Kotler DP**, Gaetz HP, Lange M, Klein EB, Holt PR. Enteropathy associated with the acquired immunodeficiency syndrome. *Ann Intern Med.* 1984;101:421-428.

**Suffredini AF**, Hochstein HD, McMahon FG. Dose-related inflammatory effects of intravenous endotoxin in humans: evaluation of a new clinical lot of Escherichia coli O:113 endotoxin. *J Infect Dis.* 1999;179:1278-1282.

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Established in 1992, the International AIDS Society–USA (IAS–USA) is a not-for-profit, HIV clinical specialist-education organization. The mission of the IAS–USA is to improve the treatment, care, and quality of life of persons with HIV and AIDS through balanced, relevant, innovative, and state-of-the-art education and information for practitioners who are actively involved in HIV and AIDS care. The organization’s educational activities are particularly intended to bridge clinical research and patient care.

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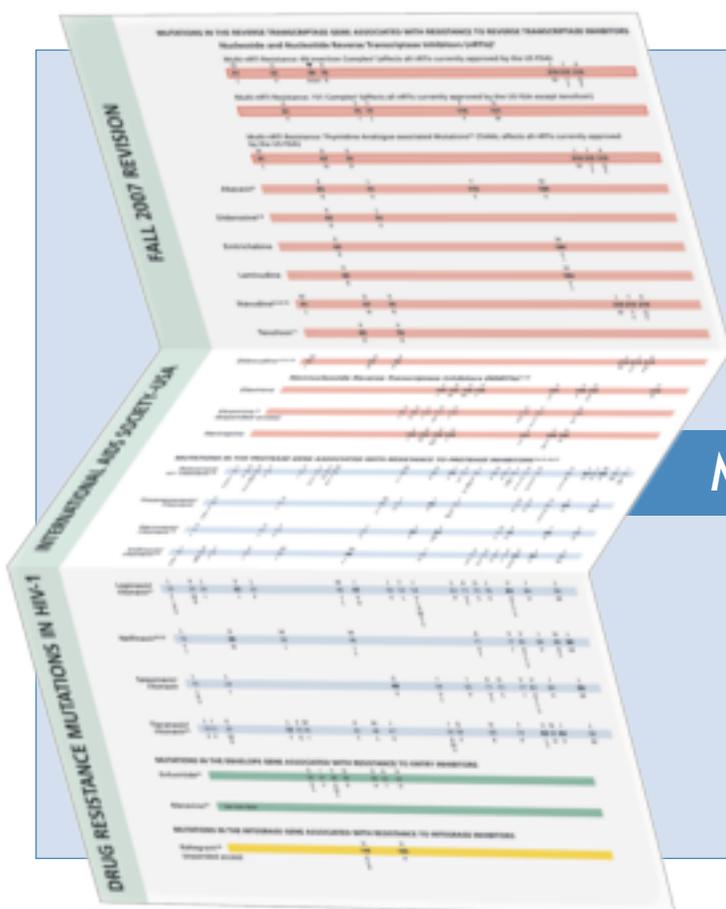
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## 2007 Updated Drug Resistance Mutations Figures

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# Update of the Drug Resistance Mutations in HIV-1: 2007

*Victoria A. Johnson, MD, Françoise Brun-Vézinet, MD, PhD, Bonaventura Clotet, MD, PhD, Huldrych F. Günthard, MD, Daniel R. Kuritzkes, MD, Deenan Pillay, MD, PhD, Jonathan M. Schapiro, MD, and Douglas D. Richman, MD*

This version of the International AIDS Society–USA (IAS-USA) Drug Resistance Mutations Figures updates the figures published in this journal in August/September 2006.<sup>1</sup> The IAS-USA Drug Resistance Mutations Group is an independent, volunteer panel of experts with the goal of delivering accurate, unbiased, and evidence-based information on these mutations to HIV clinical practitioners. As has been established for all IAS-USA panels, rotations of panel members has begun, where 1 or 2 panel members will periodically step down from panel participation and new members will join. The panel rotations are designed to ensure that all IAS-USA expert panels remain diverse in member affiliations and areas of expertise.

The figures are designed for use in identifying key mutations associated with viral resistance to antiretroviral drugs and in making therapeutic decisions. Care should be taken when using this list of mutations for surveillance or epidemiologic studies of transmission of drug-resistant virus. A number of amino acid substitutions, particularly minor mutations, represent polymorphisms that in isolation may not reflect prior drug selective pressure or reduced drug susceptibility.

The mutations listed have been identified by 1 or more of the following criteria: (1) in vitro passage experiments or validation of contribution to resistance by using site-directed mutagenesis; (2) susceptibility testing of laboratory or clinical isolates; (3) genetic sequencing of viruses from patients in

whom the drug is failing; (4) correlation studies between genotype at baseline and virologic response in patients exposed to the drug. The group reviews data that have been published or have been presented at a scientific conference. Drugs that have been approved by the US Food and Drug Administration (FDA) as well as drugs available in expanded access programs (EAPs) are included. They are listed in alphabetic order by drug class. User notes provide additional information as necessary. Although the Drug Resistance Mutations Group works to maintain a complete and current list of these mutations, it cannot be assumed that the list presented here is exhaustive. Readers are encouraged to consult the literature and experts in the field for clarification or more information about specific mutations and their clinical impact.

In the context of making clinical decisions regarding antiretroviral therapy, evaluating the results of HIV genotypic testing includes: (1) assessing whether the pattern or absence of a pattern in the mutations is consistent with the patient's antiretroviral history; (2) recognizing that in the absence of drug (selection pressure), resistant strains may be present at levels below the limit of detection of the test (analyzing stored samples, collected under selection pressure, could be useful in this setting); and (3) recognizing that virologic failure of the first regimen typically involves HIV-1 isolates with resistance to only 1 or 2 of the drugs in the regimen (in this setting, resistance most commonly develops to lamivu-

dine or the nonnucleoside reverse transcriptase inhibitors [NNRTIs]).<sup>2-7</sup> The absence of detectable viral resistance following treatment failure may result from the presence of drug-resistant minority viral populations, nonadherence to medications, laboratory error, drug-drug interactions leading to subtherapeutic drug levels, and possibly compartmental issues, indicating that drugs may not reach optimal levels in specific cellular or tissue reservoirs.

## Revisions to the Figures for the August/September 2007 Update

### Nucleoside (or Nucleotide) Reverse Transcriptase Inhibitors (nRTIs)

Recent work regarding thymidine analogue-associated mutations (TAMs) and mutations in the connection domain<sup>8</sup> have been described in user note 4, the first revision in this year's update. Because the clinical significance of mutations in the connection domain has not been determined, they have not been listed on the figure bars.

### Nonnucleoside Reverse Transcriptase Inhibitors (NNRTIs)

A bar has been added to this section for etravirine, an investigational drug newly available via an EAP, and is accompanied by user note 13, which gives additional information on the associated mutations.<sup>9</sup> The delavirdine bar has been removed because of its limited clinical role.

(continued, page 124)

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MUTATIONS IN THE REVERSE TRANSCRIPTASE GENE ASSOCIATED WITH RESISTANCE TO REVERSE TRANSCRIPTASE INHIBITORS

**Nucleoside and Nucleotide Reverse Transcriptase Inhibitors (nRTIs)<sup>1</sup>**

Multi-nRTI Resistance: 69 Insertion Complex<sup>2</sup> (affects all nRTIs currently approved by the US FDA)

M	A	▼ K				L	T	K
<b>41</b>	<b>62</b>	<b>69 70</b>				<b>210 215 219</b>		
L	V	Insert R				W	Y	Q
						F	E	

Multi-nRTI Resistance: 151 Complex<sup>3</sup> (affects all nRTIs currently approved by the US FDA except tenofovir)

	A	V	F	F	Q			
	<b>62</b>	<b>75 77</b>		<b>116</b>	<b>151</b>			
	V	I	L	Y	M			

Multi-nRTI Resistance: Thymidine Analogue-associated Mutations<sup>4,5</sup> (TAMs; affects all nRTIs currently approved by the US FDA)

M	D	K				L	T	K
<b>41</b>	<b>67</b>	<b>70</b>				<b>210 215 219</b>		
L	N	R				W	Y	Q
						F	E	

		K	L	Y	M			
Abacavir <sup>6</sup>		<b>65</b>	<b>74</b>	<b>115</b>	<b>184</b>			
		R	V	F	V			

		K	L					
Didanosine <sup>7,8</sup>		<b>65</b>	<b>74</b>					
		R	V					

		K			M			
Emtricitabine		<b>65</b>			<b>184</b>			
		R			V			
					I			

		K			M			
Lamivudine		<b>65</b>			<b>184</b>			
		R			V			
					I			

M	D	K				L	T	K
Stavudine <sup>4,5,9,10</sup>		<b>67</b>	<b>70</b>			<b>210 215 219</b>		
L	N	R				W	Y	Q
						F	E	

		K	K					
Tenofovir <sup>11</sup>		<b>65</b>	<b>70</b>					
		R	E					

M	D	K				L	T	K
Zidovudine <sup>4,5,9,10</sup>		<b>67</b>	<b>70</b>			<b>210 215 219</b>		
L	N	R				W	Y	Q
						F	E	

**Nonnucleoside Reverse Transcriptase Inhibitors (NNRTIs)<sup>1,12</sup>**

			L	K	V	V	Y	Y	G	P
Efavirenz			<b>100 103 106 108</b>				<b>181</b>	<b>188 190</b>		<b>225</b>
			I	N	M	I	C	L	S	H
							I	A		

		V	A	L	K	V	V	Y	G	
Etravirine <sup>13</sup>		90	98	100	101	106	179	181	190	
(expanded access)		I	G	I	E	I	D	C	S	
					P		F	I	A	
							V	V		

		L	K	V	V	Y	Y	G	
Nevirapine		<b>100 103 106 108</b>				<b>181</b>	<b>188 190</b>		
		I	N	A	I	C	C	A	
				M		I	L	H	

MUTATIONS IN THE PROTEASE GENE ASSOCIATED WITH RESISTANCE TO PROTEASE INHIBITORS<sup>14,15,16,17</sup>

Atazanavir +/- ritonavir <sup>18</sup>	L	G	K	L		V	L	E	M		M	G	I	F	I	D	I	I	A	G		V	I	I	N	L	I	
	10	16	20	24		32	33	34	36		46	48	<b>50</b>	53	54	60	62	64	71	73		82	<b>84</b>	<b>85</b>	<b>88</b>	90	93	
	I	E	R	I		I	I	Q	I		I	V	L	L	L	E	V	L	V	I	C		A	V	V	S	M	L
	F		M			F	F		L		L			Y	V		M		I	S		T	V					
	V		I			V	V		V								V		T	A		F						
	C		T	V											A			L				I						
Fosamprenavir/ ritonavir	L					V					M	I		I						G	L		V	I		L		
	10					32					46	47	<b>50</b>		54					73	76		82	<b>84</b>		90		
	F					I					I	V		V	L				S	V		A	V			M		
	I										L				V							F						
	R														M							S						
	V																					T						
Darunavir/ ritonavir <sup>19</sup>	V					V	L				I		I						G	L		I			L			
	11					32	33				47		<b>50</b>	<b>54</b>					73	<b>76</b>		<b>84</b>			89			
	I					I	F				V		V	M				S	V		S	V			V			
														L												V		
Indinavir/ ritonavir <sup>20</sup>	L	K	L			V		M			M			I					A	G	L	V	V	I		L		
	10	20	24			32		36			<b>46</b>				54				71	73	76	<b>77</b>	<b>82</b>	<b>84</b>		90		
	I	M	I			I		I			I			V				V	S	V	I	A	V		M			
	R										L							T	A			F						
	V																					T						
Lopinavir/ ritonavir <sup>21</sup>	L	K	L			V	L				M	I		I	F	I			L	A	G	L	V	I		L		
	10	20	24			<b>32</b>	33				46	<b>47</b>		50	53	54			63	71	73	76	<b>82</b>	84		90		
	F	M	I			I	F				I	V		V	L	V			P	V	S	V	A	V		M		
	I										L	A				L							F					
	R															A			T				T					
	V															S						S						
Nelfinavir <sup>20,22</sup>	L					D		M			M								A		V	V	I	N	L			
	10					<b>30</b>		36			46								71		77	82	84	88	<b>90</b>			
	F					N		I			I							V		I	A	V	D	M				
	I										L							T			F		S					
																					T							
Saquinavir/ ritonavir	L		L									G		I		I			A	G		V	V	I		L		
	10		24									<b>48</b>			54		62	71	73		77	82	84		<b>90</b>			
	I		I								V			V		V		V	S		I	A	V		M			
	R													L				T				F						
	V																					T						
Tipranavir/ ritonavir <sup>23</sup>	L	I	K			L	E	M			K	M	I		I	Q		H	T		V	N	I		L			
	10	13	20			<b>33</b>	35	36			43	46	47		54	58		69	74		<b>82</b>	<b>83</b>	<b>84</b>		90			
	V	V	M			F	G	I			T	L	V		A	E		K	P		L	D	V		M			
			R												M						T							
															V													

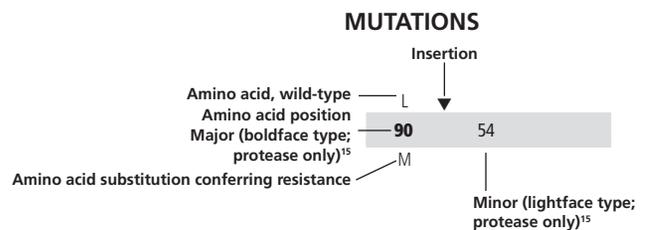
MUTATIONS IN THE ENVELOPE GENE ASSOCIATED WITH RESISTANCE TO ENTRY INHIBITORS

Enfuvirtide <sup>24</sup>	G	I	V	Q		Q	N	N
	36	37	38	39		40	42	43
	D	V	A	R		H	T	D
	S		M					
			E					
Maraviroc <sup>25</sup>	See User Note							

MUTATIONS IN THE INTEGRASE GENE ASSOCIATED WITH RESISTANCE TO INTEGRASE INHIBITORS

Raltegravir <sup>26</sup> (expanded access)	Q	N
	<b>148</b>	<b>155</b>
	H	H
	K	
	R	

**Amino acid abbreviations:** A, alanine; C, cysteine; D, aspartate; E, glutamate; F, phenylalanine; G, glycine; H, histidine; I, isoleucine; K, lysine; L, leucine; M, methionine; N, asparagine; P, proline; Q, glutamine; R, arginine; S, serine; T, threonine; V, valine; W, tryptophan; Y, tyrosine.



*The International AIDS Society–USA Drug Resistance Mutations Group reviews new data on HIV drug resistance in order to maintain a current list of mutations associated with clinical resistance to HIV. This list includes mutations that may contribute to a reduced virologic response to a drug.*

*The mutations listed have been identified by 1 or more of the following criteria: (1) in vitro passage experiments or validation of contribution to resistance by using site-directed mutagenesis; (2) susceptibility testing of laboratory or clinical isolates; (3) genetic sequencing of viruses from patients in whom the drug is failing; (4) correlation studies between genotype at baseline and virologic response in patients exposed to the drug. In addition, the group only reviews data that have been published or have been presented at a scientific conference. Drugs that have been approved by the US Food and Drug Administration (FDA) as well as drugs available in expanded access programs are included (listed in alphabetic order by drug class). User notes provide additional information as necessary. Although the Drug Resistance Mutations Group works to maintain a complete and current list of these mutations, it cannot be assumed that the list presented here is exhaustive. Readers are encouraged to consult the literature and experts in the field for clarification or more information about specific mutations and their clinical impact.*

## User Notes

1. Numerous nucleoside (or nucleotide) reverse transcriptase inhibitor (nRTI) mutations, such as the M41L, L210W, and T215Y mutations, may lead to viral hypersusceptibility to the nonnucleoside reverse transcriptase inhibitors (NNRTIs) in nRTI-treated individuals. The presence of these mutations may improve subsequent virologic response to NNRTI-containing regimens in NNRTI treatment-naïve individuals (Shulman et al, *AIDS*, 2004; Haubrich et al, *AIDS*, 2002; Tozzi, *J Infect Dis*, 2004; Katzenstein et al, *AIDS*, 2003). NNRTI hypersusceptibility can be conferred by 2 distinct phenotypes: increased enzyme susceptibility to NNRTI (eg, V118I/T215Y) or decreased virion-associated levels of reverse transcriptase (eg, H208Y/T215Y and V118I/H208Y/T215Y). The viruses that contained less reverse transcriptase replicated less efficiently than those with wild-type levels of reverse transcriptase. (Clark et al, *Antivir Ther*, 2006). The clinical relevance of all these mutations has not been assessed.

2. The 69 insertion complex consists of a substitution at codon 69 (typically T69S) and an insertion of 2 or more amino acids (S-S, S-A, S-G, or others). The 69 insertion complex is associated with resistance to all nRTIs currently approved by the US FDA when present with 1 or more thymidine analogue-associated mutations (TAMs) at codons 41, 210, or 215 (Miller et al, *J Infect Dis*, 2004). Some other amino acid changes from the wild-type T at codon 69 without the insertion may also be associated with broad nRTI resistance.

3. Tenofovir retains activity against the Q151M complex of mutations (Miller et al, *J Infect Dis*, 2004).

4. Multi-nRTI resistance mutations, also known as nucleoside analogue-associated mutations (NAMs), are associated with resistance to numerous nRTIs. The M41L, D67N, K70R, L210W, T215Y/F, and K219Q/E are known as TAMs. TAMs are a subset of NAMs that are selected by the thymidine analogues zidovudine and stavudine and are associated with cross-resistance to all nRTIs currently approved by the US FDA (Larder et al, *Science*, 1989; Kellam et al, *Proc Natl Acad Sci USA*, 1992; Calvez et al, *Antivir Ther*, 2002; Kuritzkes et al, *J Acquir Immune Defic Syndr*, 2004). Mutations at the C-terminal reverse transcriptase domains (amino acids 293–560) outside of regions depicted on the figure bars may prove to be important for HIV drug resistance. Mutations in the connection (A371V) and RNase H (Q509L) domains of reverse transcriptase are coselected on the same genome as TAMs and increase significantly zidovudine resistance when combined with TAMs. They also increase, although to a much lesser extent, cross-resistance to lamivudine, abacavir, and tenofovir but not to stavudine or didanosine (Brehm et al, *Antivir Ther*, 2006). When the polymerase domain contains TAMs, mutations in the connection domain (E312Q, G335C/D, N348I, A360I/V, V365I, and A376S) increase resistance to zidovudine from 11-fold to as much as 536-fold over wild-type reverse transcriptase (Nikolenko et al, *Proc Natl Acad Sci USA*, 2007). Three mutations (N348I, T369I, and E399D) in the reverse transcriptase C-terminus are associated with the increased resistance to zidovudine and to NNRTIs. Mutations at this level could modulate NNRTI resistance by affecting dimerization of p66/p51 heterodimers (Gupta et al, *Antivir Ther*, 2006). Since the clinical relevance of these mutations has

not been demonstrated, they are not depicted on the figure bars.

5. The E44D and the V118I mutations increase the level of resistance to zidovudine and stavudine in the setting of TAMs, and correspondingly increase cross-resistance to the other nRTIs. The significance of E44D or V118I when each occurs in isolation is unknown (Romano et al, *J Infect Dis*, 2002; Walter et al, *Antimicrob Agents Chemother*, 2002; Girouard et al, *Antivir Ther*, 2002).

6. The M184V mutation alone does not appear to be associated with a reduced virologic response to abacavir in vivo (Harrigan et al, *J Infect Dis*, 2000; Lanier et al, *Antivir Ther*, 2004). When present with 2 or 3 TAMs, M184V contributes to reduced susceptibility to abacavir and is associated with impaired virologic response in vivo (Lanier et al, *Antivir Ther*, 2004). The M184V plus 4 or more TAMs resulted in no virologic response to abacavir in vivo (Lanier et al, *Antivir Ther*, 2004).

7. The K65R mutation may be selected by didanosine and is associated in vitro with decreased susceptibility to the drug (Winters et al, *Antimicrob Agents Chemother*, 1997). The impact of the K65R mutation in vivo is unclear.

8. The presence of 3 of the following—M41L, D67N, L210W, T215Y/F, and K219Q/E—has been associated with resistance to didanosine (Marcelin et al, *Antimicrob Agents Chemother*, 2005). The K70R and M184V mutations are not associated with a decreased virologic response to didanosine in vivo (Molina et al, *J Infect Dis*, 2005).

9. The presence of the M184V mutation appears to delay or prevent emergence of TAMs (Kuritzkes et al, *AIDS*, 1996). This effect may be overcome by an accumulation of TAMs or other mutations. The clinical significance of this effect of M184V is not known.

10. The T215A/C/D/E/G/H/I/L/N/S/V substitutions are revertant mutations at codon 215, conferring increased risk of virologic failure of zidovudine or stavudine in antiretroviral-naïve patients (Riva et al, *Antivir Ther*, 2002; Chappey et al, *Antivir Ther*, 2003; Violin et al, *AIDS*, 2004). In vitro studies and preliminary clinical studies suggest that the T215Y mutant may emerge quickly from one of these mutations in the presence of zidovudine or stavudine (Garcia-Lerma et al, *J Virol*, 2004; Lanier et al, *Antivir Ther*, 2002; Riva et al, *Antivir Ther*, 2002).

11. The K65R mutation is associated with a reduced virologic response to tenofovir in

vivo (Miller et al, *J Infect Dis*, 2004). A reduced response occurs in the presence of 3 or more TAMs inclusive of either M41L or L210W (Miller et al, *J Infect Dis*, 2004). Slightly increased treatment responses to tenofovir in vivo were observed if M184V was present (Miller et al, *J Infect Dis*, 2004).

12. The long-term virologic response to sequential NNRTI use is poor, particularly when 2 or more mutations are present (Antinori et al, *AIDS Res Hum Retroviruses*, 2002; Lecossier et al, *J Acquir Immune Defic Syndr*, 2005). The K103N or Y188L mutation alone prevents the clinical utility of all NNRTIs currently approved by the US FDA (Antinori et al, *AIDS Res Human Retroviruses*, 2002). The V106M mutation is more common in HIV-1 subtype C than in subtype B, and confers cross-resistance to all currently approved NNRTIs (Brenner et al, *AIDS*, 2003; Cane et al, *J Clin Micro*, 2001).

13. The impact of most mutations depends on the simultaneous presence of Y181C; Y181C has impact only when present with 1 or more of these mutations. Substantial virologic response was still seen in clinical trials despite the presence of single mutations (Vingerhoets et al, *Antivir Ther*, 2007).

14. The same mutations usually emerge whether or not PIs are boosted with low-dose ritonavir, although the relative frequency of mutations may differ. Data on the selection of mutations in antiretroviral-naive patients in whom a boosted PI is failing are very limited. Numerous mutations are often necessary to significantly impact virologic response to a boosted PI. Although numbers vary for the different drugs, 3 or more mutations are often required.

15. Resistance mutations in the protease gene are classified as either “major” or “minor,” if data are available.

Major mutations in the protease gene are defined in general either as those selected first in the presence of the drug; or those shown at the biochemical or virologic level to lead to an alteration in drug binding or an inhibition of viral activity or viral replication. Major mutations have an effect on drug susceptibility phenotype. In general, these mutations tend to be the primary contact residues for drug binding.

Minor mutations generally emerge later than major mutations, and by themselves do not have a significant effect on phenotype. In some cases, their effect may be to improve

replicative fitness of the virus containing major mutations. However, some minor mutations are present as common polymorphic changes in HIV-1 nonsubtype B clades, such as K201/R and M36I in protease.

16. Ritonavir is not listed separately as it is currently used at therapeutic doses as a pharmacologic booster of other PIs. At higher doses tested previously in humans, ritonavir administered as monotherapy produces mutations similar to those produced by indinavir (Molla, *Nature Med*, 1996).

17. HIV-1 Gag cleavage site changes can cause PI resistance in vitro. It has been observed that mutations in the N-terminal part of *gag* (MA: E40K; L75R; K113E and CA: M200I; A224A/V), outside the cleavage site, contribute directly to PI resistance by enhancing the overall Gag processing by wild-type protease. (Nijhuis et al, *PLoS Med*, 2007). The clinical relevance of these mutations has not been assessed.

18. In most patients in whom an atazanavir/ritonavir-containing regimen was failing virologically, accumulations of the following 13 mutations were found (L10F/I/V, G16E, L33F/I/V, M46I/L, I54L/V/M/T, D60E, I62V, A71I/T/L, V82A/T, I84V, I85V, L90M, and I93L). Seven mutations were retained in an atazanavir score (L10F/I/V, G16E, L33F/I/V, M46I/L, D60E, I84V, I85V); the presence of 3 or more of these mutations predicts a reduced virologic response at 3 months, particularly when L90M was present (Vora et al, *AIDS*, 2006; <http://www.hivfrenchresistance.org/2006/tab2.html>). A different report (Bertoli et al, *Antivir Ther*, 2006) found that the presence of 0, 1, 2, or greater than or equal to 3 of the following mutations were associated with 92%, 93%, 75%, and 0% virologic response to atazanavir/ritonavir: L10C/I/V, V32I, E34Q, M46I/L, F53L, I54A/M/V, V82A/F/I/T, I84V; presence of I15E/G/L/V, H69K/M/N/Q/R/T/Y, and I72M/T/V improved the chances of response. For unboosted atazanavir, the presence of 0, 1, 2, or greater than or equal to 3 of the following mutations was associated with 83%, 67%, 6%, and 0% response rates: G16E, V32I, K201/M/R/T/V, L33F/I/V, F53L/Y, I64L/M/V, A71I/T/V, I85V, I93L/M.

19. Darunavir (formerly TMC-114), boosted with ritonavir, was approved by the US FDA in June 2006. Resistance data are therefore still preliminary and limited. HIV RNA response to boosted darunavir correlated with baseline susceptibility and the presence of multiple specific PI mutations. Reductions in response were associated with increas-

ing numbers of the mutations indicated in the bar. Some of these mutations appear to have a greater effect on susceptibility than others (eg, I50V versus V11I). Further study and analysis in other populations are required to refine and validate these findings.

20. The mutations depicted on the chart bar cannot be considered to be comprehensive since little relevant research has been reported in recent years to update the resistance and cross-resistance patterns for this drug.

21. In PI-experienced patients, the accumulation of 6 or more of the mutations indicated on the bar is associated with a reduced virologic response to lopinavir/ritonavir (Masquelier et al, *Antimicrob Agents Chemother*, 2002; Kempf et al, *J Virol*, 2001). The product information states that accumulation of 7 or 8 mutations confers resistance to the drug. In contrast, in those in whom lopinavir/ritonavir is their first PI used, resistance to this drug at the time of virologic rebound is rare. However, there is emerging evidence that specific mutations, most notably I47A (and possibly I47V) and V32I are associated with high-level resistance (Mo et al, *J Virol*, 2005; Friend et al, *AIDS*, 2004; Kagan et al, *Protein Sci*, 2005).

22. In some nonsubtype-B HIV-1, D30N is selected less frequently than other PI mutations (Gonzalez et al, *Antivir Ther*, 2004).

23. Accumulation of more than 2 mutations at positions 33, 82, 84, and 90 correlate with reduced virologic response to tipranavir/ritonavir, although an independent role for L90M was not found. Detailed analyses of data from phase II and III trials in PI-experienced patients identified mutations associated with reduced susceptibility or virologic response. These include: L10V, I13V, K20M/R, L33F, E35G, M36I, K43T, M46L, I47V, I54A/M/V, Q58E, H69K, T74P, V82L/T, N83D, and I84V. Accumulation of these mutations is associated with reduced response. Subsequent genotype-phenotype and genotype-virologic response analyses determined some mutations have a greater effect than others (eg, I84V versus I54M). Refinement and clinical validation of these findings are pending (Schapiro et al, CROI, 2005; Mayers et al, *Antivir Ther*, 2004; Hall et al, *Antivir Ther*, 2003; McCallister et al, *Antivir Ther*, 2003; Parkin et al, CROI, 2006; Bacheler et al, European HIV Drug Resistance Workshop, 2006).

24. Although resistance to enfuvirtide is associated primarily with mutations in the first heptad repeat (HR1) region of the gp41 envelope gene, wild-type viruses in the

depicted HR1 region vary 500-fold in susceptibility. Such pretreatment susceptibility differences were not associated with differences in clinical responses (Labrosse et al, *J Virol*, 2003). Furthermore, mutations or polymorphisms in other regions in the envelope (eg, the HR2 region or those yet to be identified) as well as coreceptor usage and density may affect susceptibility to enfuvirtide (Reeves et al, *Proc Natl Acad Sci USA*, 2002; Reeves et al, *J Virol*, 2004; Xu et al, *Antimicrob Agents Chemother*, 2005). Thus, testing to detect only the depicted HR1 mutations may not be adequate for clinical management of suspected failure (Reeves et al, *J Virol*, 2004; Menzo et al, *Antimicrob Agents Chemother*, 2004; Poveda et al, *J Med Virol*, 2004; Sista et al, *AIDS*, 2004; Su, *Antivir Ther*, 2004).

25. Maraviroc activity is limited to patients with only CCR5 (R5)-using virus detectable; CXCR4 (X4)-CCR5 mixed tropic viruses and X4-using viruses do not respond to maraviroc treatment. Some cases of virologic failure during maraviroc therapy are associated with outgrowth of X4 virus that pre-exists as a minority population below the level of assay detection. Mutations in the HIV-1 gp120 molecule that allow the virus to bind to R5 receptors in the presence of drug have been described in viruses from some patients whose virus remained R5 at the time of virologic failure. A number of such mutations have been identified, and the phenotypic manifestation of this drug resistance is a reduction in the maximal percentage inhibition (MPI) rather than the increase in the 50% inhibitory concentration (IC<sub>50</sub>; defined by fold increase) that is characteristic of resistance to other classes of antiretrovirals. The resistance profile for maraviroc is too complex to be depicted on the figures. The frequency and rate at which maraviroc resistance mutations emerge are not yet known.

26. Raltegravir failure was associated with integrase mutations in 2 distinct genetic pathways defined by 2 or more mutations including: (1) a signature (major) mutation at either Q148H/K/R or N155H; and (2) 1 or more minor mutations unique to each pathway. Minor mutations described in the Q148H/K/R pathway include L74M + E138A, E138K, or G140S. The most common mutation pattern in this pathway is Q148H + G140S; this Q148H + G140S pattern exhibits the greatest loss of drug susceptibility. Mutations described in the N155H pathway include this primary mutation plus either L74M, E92Q, T97A, E92Q + T97A, Y143H, G163K/R, V151I, or D232N (Hazuda et al, *Antivir Ther*, 2007).

(continued from page 119)

### Protease Inhibitors

The L76V mutation has been added to the fosamprenavir/ritonavir, indinavir/ritonavir, and lopinavir/ritonavir bars. In the protease inhibitor (PI) category, user note 20 has been updated to reflect the relative lack of recent data regarding indinavir/ritonavir and nelfinavir.

### Entry Inhibitors Resistance

The section formerly called “Mutations in the GP41 Envelope Gene Associated with Resistance to Entry Inhibitors” has been relabeled as “Mutations in the Envelope Gene Associated with Resistance to Entry Inhibitors” and includes a bar for the recently FDA-approved drug maraviroc. The resistance profile for maraviroc is too complex to be depicted on this graphic (see the user note). The enfuvirtide bar has been revised so that the mutations of the first heptad repeat (HR1) region of the gp41 envelope gene appear similarly to those in other bars.

### Integrase Inhibitors

A bar for the investigational drug raltegravir has been placed in the newly added category of “Mutations in the Integrase Gene Associated with Resistance to Integrase Inhibitors.” Raltegravir is available via an EAP. User note 26 discusses the 2 distinct genetic pathways seen in which integrase mutations are associated with raltegravir failure.<sup>10</sup>

### Comments?

The IAS-USA Drug Resistance Mutations Group welcomes comments on the mutations figures and user notes.

Please send your evidence-based comments, including relevant reference citations, to the IAS-USA at **resistance2007“at”iasusa.org** or by fax at 415-544-9401. Please include your name and institution.

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## References

- 1. Johnson VA**, Brun-Vézinet F, Clotet B, et al. Update of the drug resistance mutations in HIV-1: Fall 2006. *Top HIV Med.* 2006;14:125-130.
- 2. Descamps D**, Flandre P, Calvez V, et al. Mechanisms of virologic failure in previously untreated HIV-infected patients from a trial of induction-maintenance therapy. Trilege (Agence Nationale de Recherches sur le SIDA072) Study Team. *JAMA.* 2000;283:205-211.
- 3. Havlir DV**, Hellmann NS, Petropoulos CJ, et al. Drug susceptibility in HIV infection after viral rebound in patients receiving indinavir-containing regimens. *JAMA.* 2000;283:229-234.
- 4. Maguire M**, Gartland M, Moore S, et al. Absence of zidovudine resistance in antiretroviral-naïve patients following zidovudine/lamivudine/protease inhibitor combination therapy: virological evaluation of the AVANTI 2 and AVANTI 3 studies. *AIDS.* 2000;14:1195-1201.
- 5. Gallego O**, Ruiz L, Vallejo A, et al. Changes in the rate of genotypic resistance to antiretroviral drugs in Spain. *AIDS.* 2001;15:1894-1896.
- 6. von Wyl V**, Yerly S, Böni J, et al. Emergence of drug resistance on first-line combination therapy in the Swiss HIV cohort study: a comparison between boosted PI and NNRTI. [Abstract 667] 14th Conference on Retroviruses and Opportunistic Infections. February 25-28, 2007; Los Angeles, CA.
- 7. Walmsley S**, Bernstein B, King M, et al. Lopinavir-ritonavir versus neftinavir for the initial treatment of HIV infection. *N Engl J Med.* 2002;346:2039-2046.
- 8. Nikolenko GN**, Delviks-Frankenberry KA, Palmer S, Maldarelli F, Fivash MJ, Coffin JM, Pathak VK. Mutations in the connection domain of HIV-1 reverse transcriptase increase 3'-azido-3'-deoxythymidine resistance. *Proc Natl Acad Sci USA.* 2007;104:317-322.
- 9. Vingerhoets J**, Buelens A, Peeters M, et al. Impact of baseline NNRTI mutations on the virological response to TMC125 in the phase III clinical trials DUET-1 and DUET-2. *Antivir Ther.* 2007;12:S34.
- 10. Hazuda DJ**, Miller MD, Nguyen BY, and Zhao J for the P005 Study Team. Resistance to the HIV-integrase inhibitor raltegravir: analysis of protocol 005, a Phase II study in patients with triple-class resistant HIV-1 infection. *Antivir Ther.* 2007;12:S10.

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## Perspective

# Incorporating Novel Virologic Tests into Clinical Practice

*Virologic assays continue to evolve in order to meet the needs of HIV-infected patients and their health care providers. Genotypic and phenotypic assays for resistance to reverse transcriptase inhibitors, protease inhibitors, and fusion inhibitors have clear roles in disease management, with both types of assay having advantages and disadvantages. The failure of current assays to identify or measure the presence of minority resistant variants has clinical implications, since presence of such variants is associated with increased risk of virologic failure. Viral fitness may be relevant to disease management, but clinical role of available assays has not been determined. HIV coreceptor tropism assays will also be a crucial tool in the use of coreceptor antagonists, and data are emerging that will define pathways to treatment failure when using these new agents and the new integrase inhibitors. One clear finding for all antiretroviral drugs is that they select for resistance and must be used with good optimized background therapy to avoid virologic failure. This article summarizes a presentation on viral assays made by Eric S. Daar, MD, at an International AIDS Society–USA Continuing Medical Education course in Chicago in May 2007. The original presentation is available as a Webcast at [www.iasusa.org](http://www.iasusa.org).*

## Drug Resistance Testing for Antiretroviral Agents

Genotypic testing currently provides a report of antiretroviral resistance mutations within the HIV reverse transcriptase and protease genes. These reports can be difficult to interpret in terms of deciding on disease management when numerous resistance mutations are present. Drugs that have only recently become available may not be included in the reports. In addition, minority HIV variant populations constituting less than approximately 25% of the total viral pool in the individual patient are not detected in genotypic testing. Mixtures of wild-type and mutant variants present in proportions greater than this threshold, however, are reported.

Phenotype resistance tests are available for reverse transcriptase inhibitors, protease inhibitors, and the fusion inhibitor enfuvirtide. Results of phenotyping are reported as fold

change from wild-type virus with an indication provided as to whether the virus is susceptible, intermediate-susceptible, or resistant to each antiretroviral. Phenotyping provides an average of susceptibility of the viral population in the individual patient and does not account for how mixtures of wild-type and mutant variants separately affect susceptibility. Thus, for example, susceptibility to a particular antiretroviral might be reported on phenotyping when genotyping shows mixtures of resistant and wild-type virus to the agent. In this case, the presence of the mutant variants might well argue against use of this drug. Phenotypic testing has the advantage of providing quantitative information in the setting of complex resistance patterns. Clinical cut-off values, reporting likelihood of virologic response based on phenotype, are increasingly being investigated for individual antiretrovirals and reported in phenotypic testing. As with genotypic testing, information on susceptibility values and clinical cut-offs may not be available for newer drugs.

Discordance between genotype and phenotype results can occur because of the presence of mixtures of resistant and wild-type variants that has not yet sufficiently decreased average sus-

ceptibility of the entire infecting pool of virus. Discordance may also occur because of interactions of mutations, since mutations for some antiretrovirals can increase or decrease overall susceptibility to other antiretrovirals. For example, the M184V mutation may improve susceptibility to zidovudine and tenofovir and reduce susceptibility to abacavir and didanosine.

Virtual phenotyping testing is an approach to analyzing genotypes that assigns an expected fold change in susceptibility by comparing the patient's actual genotype with a database with matching genotypes and phenotypes. In cases in which mixtures of resistant and susceptible virus are present on genotyping, virtual phenotyping will report the sensitivity of both the resistant and susceptible variants, unlike routine phenotyping.

Thus far, there are insufficient data comparing different resistance testing techniques to determine relative usefulness in predicting response and guiding treatment. In general, all have advantages and disadvantages that must be kept in mind for optimal interpretation of results, and all provide information that is helpful in making clinical decisions (see also: Johnson et al, this issue). However, the inability of these tests to identify minority variants is likely to be relevant. A recent report showed that allele-specific polymerase chain reaction (PCR), which is capable of detecting minority variants in frequencies as low as 0.4% to 2.0%, detected minority variants in 30 of 205 samples shown as wild-type on standard genotyping. Use of allele-specific PCR to detect K103N, Y181C, and M184V minority mutations in baseline samples from patients in the CNA 30021/30024 trial examining efavirenz/lamivudine plus abacavir or zidovudine (95 with virologic failure and 220 with viral suppression) showed that virologic failure occurred in 7 of 9 patients with minority resistant variants. Logistic regression anal-

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ysis showed that the presence of the minority variants was associated with an 11-fold increased risk of virologic failure ( $P = .004$ ; Johnson, 14th CROI, 2007). Allele-specific PCR is expensive, labor intensive, has limitations, and not available for clinical use. Nevertheless these findings underscore the need to closely monitor patient response to therapy, regardless of findings in routine genotyping or phenotyping.

### Viral Fitness

Numerous studies have provided at least indirect evidence that development of multiple resistance mutations to antiretrovirals can reduce the ability of mutant HIV to replicate. For example, some studies have shown continued immunologic benefit in patients with documented drug resistance and virologic failure, and others have shown precipitous declines in CD4+ cell counts when treatment to which resistance has developed is discontinued. A true measure of viral fitness is one that analyzes the ability of the patient's population of diverse viral variants to grow in the in vivo milieu subjected to immunologic and drug pressures. Such measures are not readily available. One assay under study assesses replicative capacity—ie, how the virus replicates in the absence of drug by determining how a virus derived from the polymerase gene of the patient's virus replicates relative to a reference strain. There is some indication that measures of fitness or replication capacity can relate to what happens in clinical practice. For example, in the E-184V study, patients on lamivudine-containing therapy with HIV RNA levels above 1000 copies/mL and CD4+ counts above 500 cells/ $\mu$ L who requested a treatment interruption were randomized to interruption or lamivudine monotherapy (Castagna et al, *AIDS*, 2006). Reduction in CD4+ count to below 350 cells/ $\mu$ L or development of an opportunistic infection (which occurred in 2 patients on treatment interruption) occurred in 68% of the interruption group and 44% of the lamivudine monotherapy group. Overall, mean CD4+ cell count de-

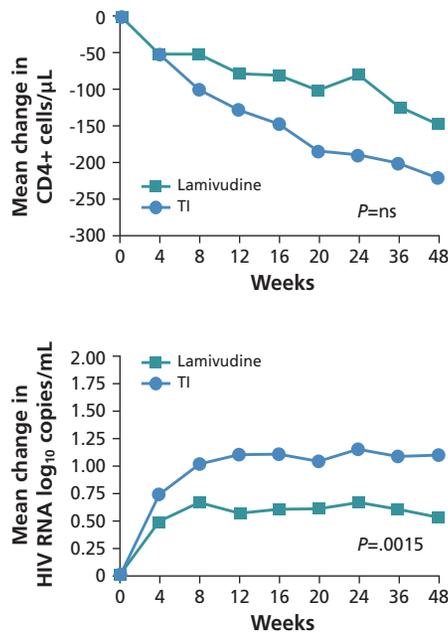


Figure 1. Change in CD4+ cell count (top) and HIV RNA level (bottom) with lamivudine monotherapy or treatment interruption (TI) in the E-184V study. Adapted from Castagna et al, *AIDS*, 2006. ns indicates nonsignificant.

clined in both groups with the reduction being nonsignificantly smaller in the lamivudine group than in the interruption group; viral load increased in both groups, with the increase being substantially smaller in the lamivudine group (Figure 1). Use of a replication capacity assay showed a 2.36-fold increase in the lamivudine group versus a 9.75-fold increase in the treatment interruption group ( $P = .013$ ). The differences in outcome between the 2 groups are likely partially explained by a reduction in viral fitness associated with the persistence of the M184V mutation under lamivudine selection pressure, which appears to be reflected by the difference in the replication capacity findings.

### Coreceptor Tropism

An antagonist of the CCR5 coreceptor was recently approved by the US Food and Drug Administration (FDA). The advent of this new class of drugs provides additional motivation for being able to identify and measure levels of CCR5-using (R5) virus with and CXCR4-using

(X4) virus (see next section).

It has long been recognized that the phenotypes of nonsyncytium-inducing (NSI) virus, typically present in early infection, and syncytium-inducing (SI) virus, typically emerging in later infection, are associated with different rates of progression in HIV infection, with the presence of SI virus being associated with more rapid immunologic deterioration. It remains unclear, however, whether there is in fact emergence of a more virulent strain of HIV or whether disease progression itself allows for emergence of the SI virus. It is now recognized that for the most part these different phenotypes correspond to coreceptor use in viral binding: the R5 viruses are the NSI viruses, which account for most transmitted variants and are prevalent in early disease; the X4 viruses are the SI viruses found more frequently in later disease and associated with rapid immunologic decline. There are also dual-tropic viruses that can use both coreceptors and some people have mixtures of X4 and R5 viruses. Phenotypic assays for receptor tropism have been developed, with one currently being widely used in clinical trials. This assay, which amplifies the entire viral envelope, identifies virus as R5 only, X4 only, or dual or mixtures of these variants. The assay, which detects X4 virus and thus nonresponsiveness to an R5 inhibitor, has a turnaround time of 16 days, a screen failure rate of 4% to 6% (based on 15,000 clinical trial samples), is 100% accurate at detecting minority variants at a 10% mixture and 83% accurate at a 5% mixture, and has 94% sensitivity at HIV RNA levels of 500 copies/mL to 1000 copies/mL. A more sensitive assay for detection of CXCR4-utilizing minority variants is in development. Investigators are also attempting to develop and validate other phenotypic assays as well as genotypic algorithms based on mutations associated with coreceptor-tropism in the gp120 V3 loop; however, the sensitivity of the latter approach may be limited.

Three studies using the phenotypic assay in treatment-naïve patients (evaluating from 325 to 979 patients) have shown R5-only virus in 81% to 88%

of samples, dual and mixed virus in 12% to 19%, and X4-only virus in 0% to 0.1%. In treatment-experienced patients, 5 studies (evaluating from 117 to 1076 subjects) have found R5-only in 49% to 67%, dual and mixed in 22% to 48%, and X4-only in 2% to 5% of samples. A study by Dr Daar's group in a hemophilia cohort using a single assessment of coreceptor tropism at baseline showed that the presence of dual and mixed-tropic virus was associated with a greater than 4-fold increased risk of clinical progression compared to those with only R5 virus.

### Resistance to Novel Targets: CCR5 Coreceptor Antagonists and Integrase Inhibitors

#### CCR5 Coreceptor Antagonists

The rationale for developing CCR5 coreceptor antagonists is that homozygosity and heterozygosity for the CCR5 Δ32 gene deletion, resulting in absence and reduction, respectively, of CCR5 receptors on the cell surface, are associated with protective benefits. Homozygotes (~1% of the white population) appear to be protected from infection, with some cases of acquisition due to X4 virus being observed. These individuals appear to be otherwise healthy (although there is some evidence of greater viremia in hepatitis C virus

infection and greater severity of West Nile virus encephalitis in this setting). Heterozygotes (~15% of whites) have delayed progression of HIV disease compared with HIV-infected individuals with wild-type CCR5.

A concern with treatment with CCR5 antagonists is the potential selection or enrichment for X4 or dual and mixed virus. In a recent study, treatment-experienced patients who had X4-only or dual and mixed or nonphenotypable virus were randomized to optimized background therapy alone or with once- or twice-daily maraviroc for 24 weeks (Mayer et al, 16th IAC, 2006). Overall, maraviroc was well tolerated and appeared to be safe. Virologic outcomes were similar in all groups, although there appeared to be a trend to better response at the higher maraviroc dose; CD4+ cell count increases were somewhat greater with maraviroc, and included increases with maraviroc versus decreases with placebo in a small number of patients with X4 virus who had virologic failure. Although these findings suggest that patients with dual and mixed virus are not likely to derive the greatest benefit from CCR5 antagonist treatment, they also suggest that the potential enrichment of dual and mixed or X4 virus was not associated with immunologic decline in the relatively short term.

In the MOTIVATE 1 and 2 studies treatment-experienced patients

with R5-only virus received once- or twice-daily maraviroc or placebo plus optimized background therapy consisting of 3 to 6 antiretrovirals. Rates of virologic response to less than 400 copies/mL and less than 50 copies/mL, were approximately twice as high in the maraviroc groups than in the placebo groups in both studies, with MOTIVATE 1 data shown in Figure 2 (Lalezari et al, 14th CROI, 2007). Analysis of changes in tropism showed that approximately 8% of patients in the combined study populations had a shift between screening and baseline, reflecting patients who had mixed populations that initially were below limits of assay detection. Among all treatment failures, 64% (63 of 98) of those receiving maraviroc had a shift from CCR5 tropism to dual and mixed tropism, compared with only 5% (4 of 84) of those receiving placebo (Nelson et al, 14th CROI, 2007). Although CD4+ cell counts increased in subgroups of patients with virologic failure, increases were smaller in the maraviroc patients who had tropism change (increases of 37/μL and 56/μL in the 2 dosage groups) than those with failure who maintained the CCR5 tropism (increases of 61/μL and 138/μL in the 2 dosage groups). The significance of these findings remains somewhat unclear. Results of in vivo resistance testing for the agent suggest that a small number of individuals do have failure with R5 virus with phenotypic resistance to the CCR5 antagonist.

MOTIVATE 1-Week 24

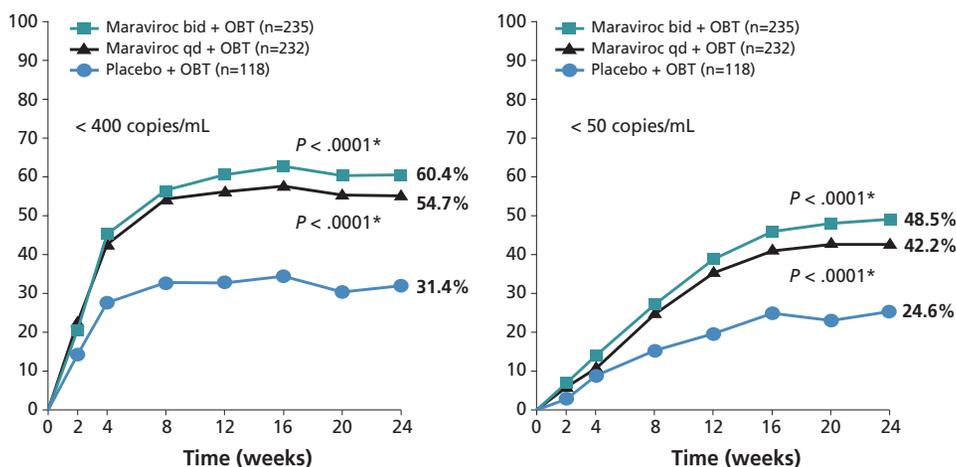


Figure 2. Virologic response in MOTIVATE 1 study. Adapted with permission from Lalezari et al, 14th CROI, 2007. OBT indicates optimized background therapy; bid, twice daily; qd, daily \*HIV-1 RNA value imputed as baseline if missing or if patient discontinued before 24 weeks versus placebo + OBT.

#### Integrase Inhibitors

In the BENCHMRK 1 and 2 trials, the addition of the investigational integrase inhibitor raltegravir to optimized background therapy in treatment-experienced patients was associated with markedly increased rates of virologic response (Cooper et al, 14th CROI, 2007; Steigbigel et al, 14th CROI, 2007). Overall, virologic failure occurred in 16% of raltegravir patients and 51% of placebo patients. Partial analysis based on genotyping in 41 patients in whom raltegravir was failing showed integrase changes in 32 cases and no consistent changes in 9. Two potential primary genetic pathways to resistance were identified: N155H (with the additional

mutations E92Q, V151I, T97A, G163R, and L74M) and Q148K/R/H (with additional mutations G140S/A and E138K). Another potential pathway was Y143R/C (with additional mutations L74A/I, E92Q, T97A, I203M, and S230R). The observed mutations were proximal to the catalytic center and similar to those selected for in in vitro testing.

## Summary

Novel virologic assays continue to evolve in order to optimally meet the needs of HIV-infected patients and their health care providers. Genotypic and phenotypic assays for resistance to reverse transcriptase inhibitors, protease inhibitors, and fusion inhibitors have clear roles in disease management, and both types of assays have advantages and disadvantages. Viral fitness appears to be relevant to disease management, but how to use available assays in clinical practice is still being explored. Tropism assays will be crucial tools in the use of coreceptor antagonists, and data are emerging that will define pathways to treatment failure when using these agents and integrase inhibitors. One clear finding for all antiretrovirals is that resistance develops for all of them, and all must be used with good optimized background therapy to avoid virologic failure.

*Presented by Dr Daar in May 2007. First draft prepared from transcripts by Matthew Stenger. Reviewed and edited by Dr Daar in August 2007.*

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## Suggested Reading

**Castagna A**, Danise A, Menzo S, et al. Lamivudine monotherapy in HIV-1-infected patients harbouring a lamivudine-resistant virus: a randomized pilot study (E-184V study). *AIDS*. 2006; 20:795-803.

**Cooper D**, Gatell J, Rockstroh J, et al. Results of BENCHMRK-1, a phase III study evaluating the efficacy and safety of MK-0518, a novel HIV-1 integrase inhibitor, in patients with triple-class resistant virus. [Abstract 105aLB.] 14th Conference on Retroviruses and Opportunistic Infections. February 25-28, 2007; Los Angeles, CA.

**Johnson J**. Clinical implications of low-frequency HIV-1 variants. [Abstract 61.] 14th Conference on Retroviruses and Opportunistic Infections. February 25-28, 2007; Los Angeles, CA.

**Johnson VA**, Brun-Vézinet F, Clotet B, et al. Update of the drug resistance mutations in HIV-1: 2007. *Top HIV Med*. 2006; 15:123-129.

**Lalezari J**, Goodrich J, DeJesus E, et al. Efficacy and safety of maraviroc plus optimized background therapy in viremic ART-experienced patients infected with CCR5-tropic HIV-1: 24-week results of a phase 2b/3 study in the US and Canada. [Abstract 104bLB.] 14th Conference on Retroviruses and Opportunistic Infections. February 25-28, 2007; Los Angeles, CA.

**Mayer H**, van der Ryst E, Saag M, et al. Safety and efficacy of maraviroc (MVC), a novel CCR5 antagonist, when used in combination with optimized background therapy (OBT) for the treatment of antiretroviral-experienced subjects infected with dual/mixed-tropic HIV-1: 24-week results of a phase 2b exploratory trial. [Abstract ThLB0215.] 16th International AIDS Conference. August 13-18, 2006; Toronto, Canada.

**Nelson M**, Fatkenheuer G, Konourina I, et al. Efficacy and safety of maraviroc plus optimized background therapy in viremic, ART-experienced patients Infected with CCR5-tropic HIV-1 in Europe, Australia, and North America: 24-week results. [Abstract 104aLB.] 14th Conference on Retroviruses and Opportunistic Infections. February 25-28, 2007; Los Angeles, CA.

**Steigbigel R**, Kumar P, Eron J, et al. Results of BENCHMRK-2, a phase III study evaluating the efficacy and safety of MK-0518, a novel HIV-1 integrase inhibitor, in patients with triple-class resistant virus. [Abstract 105bLB.] 14th Conference on Retroviruses and Opportunistic Infections. February 25-28, 2007; Los Angeles, CA.

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## Perspective

# Human Papillomavirus Infection in HIV-infected Persons

*Rates of cervical and anal human papillomavirus (HPV) infection and abnormal cytology are high in HIV-infected women, as are rates of anal HPV infection and abnormal cytology in HIV-infected men who have sex with men (MSM). Available evidence indicates that the incidence of anal cancer in HIV-infected MSM has increased in association with prolonged life expectancy achieved with antiretroviral therapy. Routine screening for cervical neoplasia is recommended for HIV-infected women. Routine screening is not yet universally recommended for anal neoplasia, although it should be considered for at-risk patients, particularly given recent improvements in local treatments. A preventive vaccine against cervical HPV infection is approved for use in young women before onset of sexual activity and acquisition of HPV infection. Its potential benefit in preventing anal infection in women and men has yet to be determined, and its potential utility in those with HIV infection remains unknown. This article summarizes a presentation on HPV infection in HIV-infected patients made by Joel Palefsky, MD, at an International AIDS Society–USA Continuing Medical Education course in Chicago in May 2007. The original presentation is available as a Webcast at [www.iasusa.org](http://www.iasusa.org).*

Infection with human papillomavirus (HPV) poses risk of cervical and anal cancer in women and anal cancer in men who have sex with men (MSM). There are high rates of cervical and anal HPV infection in HIV-infected women and high rates of anal HPV infection in HIV-infected MSM. Infection in the cervix primarily occurs in the transformation zone where the squamous epithelium of the exocervix and columnar epithelium of the endocervix meet. The anal canal has a similar zone where the squamous epithelium of the anus meets the columnar epithelium of the rectum. This latter structure is more exteriorized in women than in men, which may account for the apparent higher rate of anal infection in women than in men. The virus must reach the basal cell layer to initiate infection, and it is currently thought that it takes approximately 24 hours for the virus to enter these cells. This window appears to allow time for vaccine-induced antibody response that has been shown to be effective in preventing infection (see below).

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### Risk of Human Papillomavirus Infection and Associated Cancer

Figure 1 shows rates of HPV cervical infection and abnormal cervical cytology in HIV-seronegative women and HIV-infected women according to CD4+ cell count, and rates of anal infection and abnormal cytology in MSM according to HIV status and CD4+ cell count. Rates of infection and abnormal cytology increase as CD4+ cell count decreases, as do the number of oncogenic HPV types involved in infection. As shown in Figure 2, data from a single study population (the Women's Interagency HIV Study) show that anal infection in women is more common than cervical infection. Studies in the general population have also shown that rates of anal infection are at least as high as rates of cervical infection in women.

HPV infection is the most common sexually transmitted infection in the general population, with it being estimated that some 75% of all sexually active adults acquire a genital HPV type during their lifetime. In immunocompetent individuals, HPV replication is usually suppressed, with relatively low rates of progression of low-grade squamous intraepithelial lesions (LSIL), which are not thought to be a cancer precursor, to high-grade squamous in-

traepithelial lesions (HSIL), which are believed to be a cancer precursor. In HIV infection, immune suppression is associated with higher rates of progression of LSIL to HSIL. However, it appears that immune response is not as important a determinant of progression from HSIL to cancer, with slow accumulation of genetic changes appearing to account for the prolonged

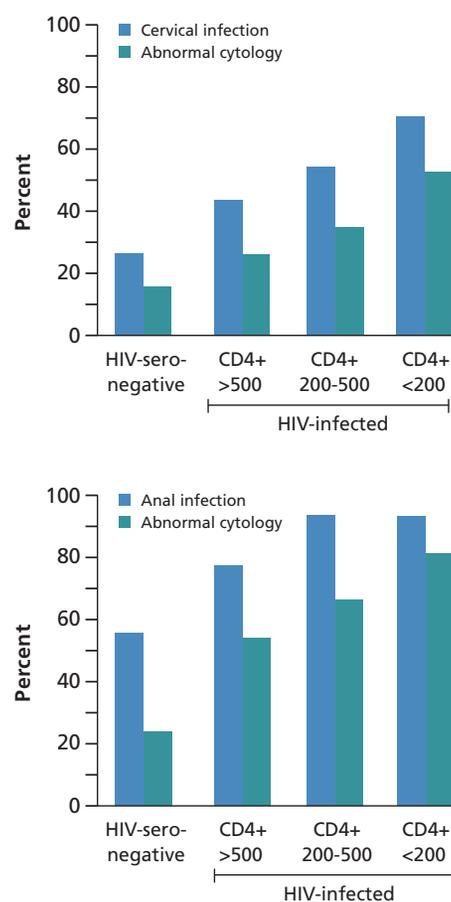


Figure 1. Top: Rates of human papillomavirus (HPV) cervical infection and abnormal cytology in HIV-seronegative women and HIV-infected women according to CD4+ count (cells/μL). Bottom: Rates of HPV anal infection and abnormal cytology in HIV-seronegative men who have sex with men (MSM) and HIV-infected MSM according to CD4+ count (cells/μL). Adapted from Palefsky et al, *J Natl Cancer Inst*, 1999; Massad et al, *JAIDS*, 1999; Palefsky et al, *JAIDS*, 1998; and Palefsky et al, *J Infect Dis*, 1998.

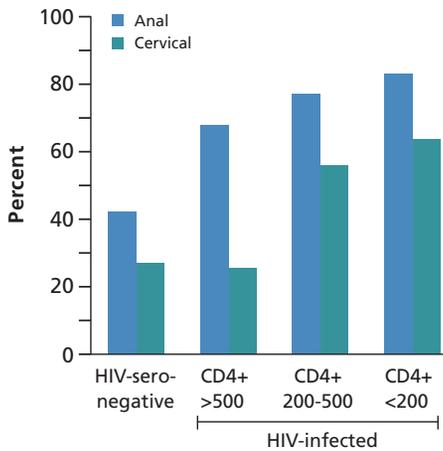


Figure 2. Rates of cervical and anal human papillomavirus infection in HIV-seronegative women and HIV-infected women according to CD4+ count (cells/ $\mu$ L) in the Women's Interagency HIV Study. Adapted from Palefsky et al, *J Infect Dis*, 2001.

progression (eg, 20 years) to cancer. This is supported by the finding of studies of the effect of antiretroviral therapy on cervical and anal lesions, with available data suggesting only a minor positive effect or no effect of antiretroviral therapy-associated immune reconstitution on regression of HSIL. In fact, numerous data indicate that the rate of anal cancer is increasing in HIV-infected individuals since the advent of potent antiretroviral therapy, with the prolonged life span associated with antiretroviral therapy appearing to permit progression to cancer in more patients. One recent study found that the rate of anal cancer in the general population increased from 0.6 per 100,000 population in the pre-HIV era of 1973 to 1981 to 0.8 per 100,000 in 1982 to 1995, and to 1 per 100,000 during the potent antiretroviral therapy era of 1996 to 2001 (Chiao et al, *JAIDS*, 2005). At the same time, the ratio of females to males decreased from 1.6 to 1, to 1.2 to 1. It is believed that these changes reflect an increasing incidence of anal cancer in the HIV-infected MSM population. Another study in 8640 HIV-infected MSM in London (Bower et al, *JAIDS*, 2004) showed that the rate of anal cancer increased from 35 per 100,000 person-years in the pre-antiretroviral therapy era to 92 per 100,000 person-years af-

ter introduction of antiretroviral drugs. A study matching the San Francisco AIDS Surveillance Registry and the California Cancer Registry showed that among more than 14,000 adults diagnosed with AIDS from 1990 to 2000, the risk of anal cancer increased nearly 3-fold after 1995, with likelihood of death from anal cancer also increasing somewhat during this period (Hessol et al, *Am J Epidemiol*, 2007).

### Assessment Guidelines

The guidelines for assessing cervical intraepithelial neoplasia (CIN) in HIV-infected women are currently being rewritten but are expected to undergo little change. The guidelines call for a Pap test at initial evaluation and a repeated Pap test at 6 months. If both are negative, Pap testing can be repeated annually. Physicians should maintain a low threshold for performing colposcopy for findings of atypical squamous cells of unknown significance (ASCUS) or higher grades of dysplasia.

The 2005 Centers for Disease Control and Prevention (CDC), National Institutes of Health (NIH), HIV Medicine Association of the Infectious Diseases Society of America guidelines for assessment of anal intraepithelial neoplasia (AIN) in HIV-infected patients state: "Although formal guidelines recommending anal Pap smear screening have not been adopted, certain specialists recommend anal cytologic screening for HIV-1-infected men and women. High-resolution anoscopy should be considered if the anal Pap smear indicates ASCUS or ASC-H [atypical squamous cells—cannot rule out HSIL] and should be performed if a person has LSIL or HSIL on anal Pap smear. Visible lesions should be biopsied to determine the level of histologic changes and to rule out invasive cancer." These guidelines are also being rewritten, but the new guidelines also will not recommend routine screening. The recommendation for routine screening awaits evidence that screening and treatment of lesions reduces risk of progression to anal cancer.

However, some local authorities are recommending screening when it is available. For example, the New York

State Department of Public Health AIDS Institute guidelines for the Primary Care Approach to the HIV-Infected Patient state: "Although this is a new practice that may not be routinely available, screening for cellular dysplasia is prudent and recommended, particularly in persons at high risk for infection with papilloma virus." The guidelines recommend that at baseline and as part of the annual physical examination of all HIV-infected adults, clinicians should: inquire about anal symptoms, such as itching, bleeding, diarrhea, or pain; perform a visual inspection of the anal region; and perform a digital rectal examination. It is further recommended that clinicians perform anal cytology at baseline and annually in MSM, in any patient with a history of anogenital condylomas, and in women with abnormal cervical or vulvar histology, and that patients with abnormal anal Pap test findings be referred for high-resolution anoscopy or examination with biopsy.

Infrastructure for treating abnormal anal findings in routine screening is currently lacking in most locales, and the value of screening in the absence of such infrastructure is questionable. However, such infrastructure should be developed, since earlier identification of abnormalities or cancer itself can result in improved outcomes. Identification of lesions early in the natural history of disease would increase the proportion of patients identified with smaller lesions; small lesions can be treated with application of trichloroacetic acid, which is well tolerated and very inexpensive. Identification of lesions during later progression—eg, the middle range of natural history of the disease—would increase the proportion of patients who could have treatment by other local modalities, avoiding the morbidity and expense associated with surgical removal. For example, infrared coagulation, an office-based treatment performed with an anoscope, has been found to result in freedom from disease in 65% of patients after median follow-up time of 413 days (Goldstone et al, *Dis Colon Rectum*, 2005).

Earlier identification of disease late in the course of progression would fa-

**Table 1.** Analysis of Efficacy of Human Papillomavirus (HPV) L1 Protein Virus-like Particle Vaccine Against HPV Types 16- and 18-related Cases of Cervical Intraepithelial Neoplasia Grades 2 and 3 or Worse

	Vaccine N=10,268			Placebo N=10,273			Efficacy (95% confidence interval)
	No. of subjects	No. of cases	Incidence	No. of subjects	No. of cases	Incidence	
Per-protocol	8487	0	0	8460	53	0.4	100% (92.9–100)
Modified intent-to-treat	9831	122	0.6	9896	201	0.9	39.0% (23.3–51.7)

Per-protocol population included only women with no prior evidence of HPV infection. Modified intent-to-treat population included women with prior HPV infection. Adapted from Human Papillomavirus Quadrivalent (Types 6, 11, 16, and 18) Vaccine, Recombinant, Package Insert, 2006.

cilitate earlier diagnosis and treatment of cancer. As with cervical cancer, survival in anal cancer is better with earlier diagnosis and treatment. Treatment for anal cancer currently involves use of chemotherapy and radiation therapy, which are associated with substantial morbidity.

It must be emphasized, however, that it remains to be demonstrated that screening and treatment of AIN can reduce the incidence of anal cancer. It is likely that routine screening will not be universally recommended until such evidence is available. Even in the absence of infrastructure for screening and treatment of AIN, all at-risk men and women should be screened for anal cancer by digital rectal exam.

### Human Papillomavirus Vaccines

The currently available preventive HPV vaccine is a quadrivalent virus-like particle vaccine covering HPV types 16 and 18, implicated in approximately 70% of cervical cancers worldwide, and covering types 6 and 11, which cause genital warts. In women with no serologic or DNA evidence of HPV type 16 or 18, the vaccine was 100% effective in preventing CIN grades 2 and 3 or worse due to these HPV types (Garland et al, *N Engl J Med*, 2007). In an intent-to-treat analysis including women with prior exposure to HPV, vaccine efficacy was 39% (see Table 1). The vaccine is currently recommended in the United States for young women prior to onset of sexual activity (eg, ages 11 or

12 years), who thus are unlikely to have had prior HPV infection. The vaccine was also 99% effective in preventing genital warts. In intent-to-treat analysis including women with prior exposure to HPV, vaccine efficacy was 12.2% in preventing CIN grades 2 and 3 or worse due to any HPV type. Although, again, the study population was not similar to the current target population of young women, the finding emphasizes the fact that infection with HPV types not covered by the vaccine is still possible, and that it is still necessary for vaccinated individuals to receive annual Pap screening. Thus far, the vaccine appears safe. It is currently unknown whether booster doses are needed. Other preventive HPV vaccines are in development, including one designed to protect against infection with HPV 16 and HPV 18, and in phase III trials has been shown to be highly effective to prevent initial infection with these HPV types as well as to prevent development of CIN grades 2 to 3 associated with HPV 16 and HPV 18 in women who had not yet been exposed to these types (Paavonen et al, *Lancet*, 2007). The vaccine is currently being reviewed for approval by the US Food and Drug Administration (FDA).

With regard to potential use in HIV-infected individuals, several issues need to be clarified, including safety, whether there are sufficient numbers of HIV-infected individuals who are not HPV-infected already to provide for vaccine efficacy, and whether HIV-infected individuals can mount and maintain

protective antibody titers against HPV. It is currently unclear whether the vaccine protects against anal HPV infection and AIN, although studies regarding this are underway. It should also be considered whether all men should be vaccinated before the start of sexual activity to prevent penile infection (and thus transmission to sexual partners, be they men or women) and to prevent anal infection in MSM, as well as to interrupt the cycle of transmission.

### Conclusion

The incidence of AIN and anal cancer is high among HIV-infected women and MSM. Antiretroviral therapy has a limited positive effect on HPV-related neoplasia, and there is mounting evidence that the incidence of anal cancer will continue to rise among HIV-infected MSM. At-risk men and women should be considered for screening and treatment of AIN, particularly since local treatments are improving. All at-risk men and women should be screened for anal cancer by digital rectal exam, since real benefits accrue from early detection of cancer. Data are awaited to determine whether HPV vaccines can prevent anal HPV infection, AIN, and anal cancer.

### Suggested Reading

**Bower M,** Powles T, Newsom-Davis T, et al. HIV-associated anal cancer: has highly active antiretroviral therapy reduced the incidence or improved the outcome? *J Acquir Immune Defic Syndr*. 2004;37:1563-1565.

**Chiao EY**, Krown SE, Stier EA, Schrag D. A population-based analysis of temporal trends in the incidence of squamous anal canal cancer in relation to the HIV epidemic. *J AIDS*. 2005;40:451-455.

**Gardasil** [Package Insert]. White Station, NJ; Merck & CO., Inc. 2006

**Garland SM**, Hernandez-Avila M, Wheeler CM, et al. Quadrivalent vaccine against human papillomavirus to prevent anogenital diseases. *N Engl J Med*. 2007;356:1928-1943.

**Goldstone SE**, Kawalek AZ, Huyett JW. Infrared coagulator: a useful tool for treating anal squamous intraepithelial lesions. *Dis Colon Rectum*. 2005;48:1042-1054.

**Hessol NA**, Pipkin S, Schwarcz S, Cress RD, Bacchetti P, Scheer S. The impact of highly active antiretroviral therapy on non-AIDS-defining cancers among adults with AIDS. *Am J*

*Epidemiol*. 2007;165:1143-1153.

**Paavonen J**, Jenkins D, Bosch FX, et al. Efficacy of a prophylactic adjuvanted bivalent L1 virus-like-particle vaccine against infection with human papillomavirus types 16 and 18 in young women: an interim analysis of a phase III double-blind, randomised controlled trial. *Lancet*. 2007;369:2161-2170.

**Palefsky JM**, Gillison ML, Strickler HD. HPV vaccines in immunocompromised women and men. *Vaccine*. 2006;24(Suppl 3):S140-S146.

**Palefsky JM**, Holly EA, Efrdc JT, et al. Anal intraepithelial neoplasia in the highly active antiretroviral therapy era among HIV-positive men who have sex with men. *AIDS*. 2005;19:1407-1414.

**Palefsky JM**, Holly EA, Ralston ML, Da Costa M, Greenblatt RM. Prevalence and risk factors for anal human papillomavirus infection in

human immunodeficiency virus (HIV)-positive and high-risk HIV-negative women. *J Infect Dis*. 2001;183:383-391.

**Palefsky JM**, Minkoff H, Kalish LA, et al. Cervicovaginal human papillomavirus infection in human immunodeficiency virus-1 (HIV)-positive and high-risk HIV-negative women. *J Natl Cancer Inst*. 1999;91:226-236.

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## Perspective

# Elite Control of HIV Infection: Implications for Vaccines and Treatments

*Spontaneous and sustained (“elite,” or aviremic) control of HIV infection (ie, maintaining HIV RNA to <50 copies/mL in the absence of therapy) appears to occur in approximately 1 in 300 HIV-infected persons, and represents a distinct phenotype among HIV-infected individuals. Through a recently established international collaboration called the HIV Controller Consortium, over 300 elite controllers have been identified and blood samples collected. These ongoing studies will not only examine the immune responses to HIV that elite controllers generate, but will also make use of a newly available approach to defining the genetic basis of disease. Specifically, the consortium is attempting to determine the genetic basis underlying spontaneous control by performing whole genome analysis scans together with functional immunology studies in a large population of elite controllers. The goal of these studies is to provide insights that will help define the crucial parameters present in persons who are able to control HIV infection, similar to the control most people have with Epstein-Barr virus and varicella, namely by holding the virus in check. These findings could assist in the development of vaccines and new therapies. This article summarizes a presentation on spontaneous control of HIV infection and its implications for vaccine development made by Bruce D. Walker, MD, at an International AIDS Society–USA Continuing Medical Education course in New York in March 2007. The original presentation is available as a Webcast at [www.iasusa.org](http://www.iasusa.org).*

Progress toward a preventive HIV vaccine has been slow, and after 20 years of focused vaccine research an effective vaccine remains elusive. The greatest hindrance may be the inability to identify immunogens that can generate broadly cross-neutralizing antibodies capable of recognizing the extremely wide variation in target HIV envelope (Env) proteins; indeed, it is doubtful whether a single vaccine could produce a response that protects against the wide variation in Env proteins among potentially infecting HIV strains present within a single individual, much less within a large population. Given that a preventive vaccine is unlikely, important current goals in vaccine development include an alternate approach: identifying vaccines that do not prevent infection but reduce risk of transmission and prevent disease progression should a vaccinated person become infected.

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A variety of data indicate that risk of transmission of HIV is markedly reduced at plasma HIV RNA levels below 2000 copies/mL, a level of viremia at which progression of disease is also slowed. Such a level of viremia could serve as a target for vaccines to reduce risk of transmission and disease progression. Importantly, spontaneous control of HIV infection has been maintained at or below the relative threshold of an HIV RNA level of 2000 copies/mL in some individuals for up to 28 years or longer. Further understanding of these mechanisms could assist in the development of vaccines that augment control of infection, and may also serve to identify new targets for pharmacologic intervention. To this end, there has been a growing focus on understanding host, viral, and immunologic parameters that are associated with effective containment of HIV infection, and a collaborative effort, the International HIV Controller Consortium, involving patients, health care providers, and scientists has been established to focus an international effort to recruit

these patients and uncover the mechanisms that account for control.

## Working Definitions of Elite and Viremic Controllers

The HIV Controller Consortium focuses on the tail end of the spectrum of viral load in untreated HIV infection, with a particular emphasis on those with undetectable viral loads, an HIV RNA level of below 50 copies/mL by the currently available assays. Individuals who have maintained HIV RNA levels below 50 copies/mL for at least 1 year in the absence of antiretroviral therapy are identified as elite, or aviremic, controllers, and those maintaining levels of 50 copies/mL to 2000 copies/mL are identified as viremic controllers (see Table 1). Using these criteria, the median duration of infection for persons identified thus far is more than 12 years.

## Host and Viral Factors in HIV Controllers

With regard to potential host genetic factors in viral control, the HLA class I allele HLA-B\*57 and to a lesser extent the HLA-B\*27 allele are over-represented among elite controllers and viremic controllers compared with individuals with progressive infection. Among the first 60 elite controllers recruited in Boston, less than half express HLA-B\*57, and this is holding true as the effort is expanded with the HIV Controller Consortium. HLA-B\*27 is present in approximately one-fifth of patients. Although these findings clearly suggest a host genetic factor in viral control, the HLA-B\*57 allele, for example, is still absent in half or more of individuals with elite control. It is not yet understood how presence of these HLA types is mechanistically related to augmented viral control, but data suggest a link between HLA-B\*57 and expression of certain receptors of the innate immune system, suggesting the possibility

**Table 1. HIV Controller Consortium Definitions of Elite and Viremic HIV Controllers.**

#### Elite Controllers

- Maintain HIV RNA levels below 50 copies/mL
- No antiretroviral therapy for 1 year or longer
- Episodes of viremia are acceptable as long as there are not consecutive episodes

#### Viremic Controllers

- Maintain HIV RNA levels below 2000 copies/mL
- No antiretroviral therapy for 1 year or longer
- Episodes of viremia are acceptable as long as they represent the minority of all available determinations

Additional information is available at [www.elitecontrollers.org](http://www.elitecontrollers.org).

that innate immune mechanisms mediated via natural killer cells might contribute to long-term containment.

With regard to potential virus factors, a report several years ago documented slowed progression of disease in association with *nef*-deleted HIV mutants in an Australian cohort infected via transfusion from a single donor. Although earlier studies of *nef* in elite controllers have yielded conflicting results, data thus far using full genome sequencing in elite controllers has not suggested an association of augmented control with attenuated virus due to *nef* deletions or other specific viral polymorphisms. In addition, investigators at the Johns Hopkins University have succeeded in showing that replication-competent HIV can be isolated from aviremic controllers. These are promising findings since they suggest that the state of augmented control is not due merely to “wimpy” virus, but that there should be identifiable and potentially replicable mechanisms in humans that permit spontaneous control of replication-competent HIV.

#### Host Immune Factors

Cytotoxic CD8+ T lymphocytes (CTLs) act to kill HIV-infected host cells during

the period when the virus has become uncoated and its components are in the host cell cytoplasm, with these cells being recognized via presentation of viral antigen in the context of HLA class I molecules. HIV-specific CTLs are effective in eliminating infectious virus in tissue culture. The activity of these T cells and the enrichment for particular HLA class I molecules in elite controllers caused speculation that controllers might exhibit increased magnitude or breadth of CTL response to HIV. However, Dr Walker and colleagues found that magnitude of HIV-specific CTL response (as measured by interferon gamma enzyme-linked immunospot [ELISPOT] assay) was actually lower in elite controllers than in chronically infected progressors, with viremic controllers having an intermediate magnitude of response. Breadth of response (number of peptides to which there was response) was also statistically significantly smaller in elite controllers. Preliminary data suggest preferential targeting of Gag in elite and viremic controllers, consistent with the preferential targeting of Gag recently reported in persons with chronic untreated infection who experience augmented control of viremia. In this same cohort, preferential targeting of Env by HIV-specific CD8 T cells was associated with higher viral load.

These findings make sense from an immunologic perspective. The viral envelope may function as a decoy to evade immune response, accommodating extensive genetic heterogeneity that does not cause a replicative fitness disadvantage to the virus. Thus, immune responses directed against Env proteins may have relatively lesser effect in reducing viral populations. Gag is less flexible in this regard, since Gag mutations can come at the cost of reduced replicative fitness for the virus. In addition, as noted, CTLs recognize processed viral proteins generated in the target cell cytoplasm. When HIV infects a cell, the Env proteins are left on the cell surface and the preformed core proteins, including Gag, are injected into the cell. These proteins are then processed and sent back to the cell surface where they are presented

for immune recognition, likely before production of new virions has occurred within the cell. Studies from Dr David Watkin’s lab have shown that infected cells become targets for Gag-specific CTL response within hours of infection. In contrast, Env proteins will be similarly processed and presented only after production of new virions within the cell. Env-specific CTL responses do not occur for 24 hours after infection. Many efforts at developing vaccines intended to protect against disease progression have focused on generating Env-specific responses. However, driving the immune response toward Env-specific responses may turn out to favor the virus; driving the immune response toward Gag-specific responses may be a better strategy in terms of achieving long-term control.

Additional studies are needed to characterize the antiviral effect of Gag-specific CTL response. For the study of neutralizing antibodies, the gold standard is the ability to limit virus replication in vitro. Dr Walker and colleagues compared Gag-specific CTL lines with Env-specific CTL lines from elite controllers on the effects of HIV replication in tissue culture, and found that inhibition is consistently better with Gag-specific responses than inhibition with Env-specific responses. It remains to be determined why some individuals have such effective Gag-specific responses and many do not. One potential factor may involve function of CD4+ T helper cells. Comparison of aviremic controllers, viremic controllers, and progressors has shown that aviremic controllers have statistically significantly higher percentages of CD4+ T helper cells and CTLs that produce both interleukin-2 and interferon gamma (IFN- $\gamma$ ) than do either viremic controllers or progressors.

With regard to humoral immune response, studies of autologous and heterologous neutralization of virus derived from aviremic controllers, viremic controllers, and progressors using plasma from subjects in each group showed that levels of neutralizing antibodies were lowest for aviremic controllers.

## Next Steps

Elite controllers of HIV infection appear to be a distinct phenotype, and such control appears to involve the cellular immune response. Dr Walker and colleagues believe that the next key advances for vaccine design will come from dissection of the immune responses that account for spontaneous control of HIV. Lessons and advances from the Human Genome Project can contribute to progress in this regard. Based on techniques of the Human Genome Project, the whole genome analysis scan (WGAS) is a new technique that permits rapid automated analysis for short gene segments using single nucleotide polymorphisms as genetic signatures. This type of analysis has permitted definition of disease-related genes with study of as few as 100 individuals. The HIV Controller Consortium has a goal of performing WGAS on 1000 elite controllers and 1000 viremic controllers, along with 1000 progressor controls, to determine the influence of genetic factors on innate and adaptive immunity and durable suppression of HIV.

The estimated frequency of elite controllers is 1 in 300 infected individ-

uals. On the assumption that approximately 600,000 infected individuals in the United States know their infection status, it is estimated that approximately 2000 infected individuals know of their elite controller status. We hope that practitioners who know of such individuals will contact the consortium or encourage patients to contact the consortium for potential inclusion in this project. Participant blood samples will undergo evaluation by WGAS and functional immunology studies.

## Conclusion

In Dr Walker's studies, elite control of HIV infection has been associated with a number of factors—expression of HLA-B\*57, dominant CTL targeting of Gag, lower total magnitude and breadth of HIV-specific CTL response, variable inhibition of viral replication by CTL of different specificities, increased functionality of CD4+ and CD8+ T cells, and weak neutralizing antibodies. None of these factors alone predicts elite control. Yet, we know that such a level of spontaneous control of apparently infectious virus does occur, and determining what immunologic factors are suffi-

cient to produce such a state appears to be an achievable task. Identifying these immunologic factors could lead to development of vaccines that could prevent disease progression and control viremia in infected individuals to levels associated with a markedly reduced risk of transmission. Such an effect could have a profound impact in containing and contracting the global HIV epidemic. It is hoped that the HIV Controller Consortium studies will lead to identification of the factors underlying elite control. Readers of this article are encouraged to refer patients for this study, through [elitecontrollerstudy@partners.org](mailto:elitecontrollerstudy@partners.org), or by contacting Dr Florencia Pereyra at [fpereyra@partners.org](mailto:fpereyra@partners.org). Additional information on this study is available at [www.elitecontrollers.org](http://www.elitecontrollers.org).

*Presented by Bruce D. Walker, MD, in New York in March 2007. First draft prepared from transcripts by Matthew Stenger. Reviewed and edited by Dr Walker in August 2007.*

*Dr Walker has served as a consultant to Bristol-Myers Squibb, GlobeImmune, and Merck.*

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## Review

## Preconception Health Care for HIV-infected Women

Erika Z. Aaron, MSN, CRNP, and Shannon M. Criniti, MPH

*The advent of potent antiretroviral therapy coupled with the dramatic reduction in mother-to-child transmission seen over the past decade has allowed women with HIV infection to live longer, healthier lives and has affected their fertility desires. As a result, preconception health care for HIV-infected women should be a routine part of primary health care. Such counseling includes health recommendations and counseling for safer sexual practices, contraception, and pregnancy planning, identifies individual risks and corresponding interventions, provides personalized and nonjudgmental education, and provides access to integrated services that address all of a woman's health and psychological needs. The goals are: (1) to improve the health of the woman before conception; (2) to identify risk factors for adverse maternal or fetal outcomes and initiate interventions to optimize outcomes; and (3) to prevent transmission of HIV to infants as well as sexual partners. This article will review the components of preconception health care for HIV-infected women.*

Care for women with HIV infection has specific challenges before conception and during pregnancy that should be addressed in a comprehensive, standardized plan of care. Preconception care for HIV-infected women includes health recommendations and counseling for safer sexual practices, contraception, and pregnancy planning. The goals are: (1) to improve the health of the woman before conception; (2) to identify risk factors for adverse maternal or fetal outcomes and initiate interventions to optimize outcomes; and (3) to prevent transmission of HIV to infants as well as sexual partners. Preconception care is not a single clinical visit, but rather ongoing care that is integrated into primary care to address the needs of women with HIV infection during the different stages of reproductive life.<sup>1</sup> This article will review the components of preconception health care for HIV-infected women, emphasizing a process of ongoing care throughout the reproductive years.

### Background

The advent of potent antiretroviral therapy coupled with advances in the

understanding and treatment of HIV infection has improved the life expectancy for women with HIV infection. Similarly, the use of antiretroviral drugs during pregnancy has resulted in a dramatic reduction in mother-to-child transmission of HIV.<sup>2</sup> As HIV-infected women live longer and healthier lives, many express fertility desires and choose to become pregnant, and others face unintended pregnancies. Studies have estimated that 70% of HIV-infected women are sexually active<sup>3</sup> and 25% to 30% of HIV-infected women receiving medical care in North America express desires to have children.<sup>4,5</sup> Because one-half of all pregnancies in the United States are unintended,<sup>6</sup> preconception care must be initiated before patients express fertility desires in order to be effective. Clinicians should routinely discuss fertility desires as part of comprehensive primary care services for HIV-infected women.

It is crucial that a new model of health care be implemented that incorporates preconception care across the reproductive lifespan. This model should address the realities of living with HIV infection, such as its implica-

tions for reproductive health, fetal well being, and the prevention of transmission to infants and sexual partners. This model should be based on an expanded view of preconception health care, one that encompasses primary care, recommendations for safer sexual practices, contraception, and pregnancy planning, as well as one that calls for the collaboration of multidisciplinary health care providers.

### Components of Preconception Health Care for HIV-infected Women

The Centers for Disease Control and Prevention (CDC), the American College of Obstetrics and Gynecology (ACOG), and other national organizations recommend offering all women of childbearing age the opportunity to receive preconception counseling and care as a component of routine primary medical care.<sup>7,8</sup> The purpose of such care is to improve the health of every woman before conception by: (1) providing education and counseling targeted to the individual's needs; (2) identifying risk factors for adverse maternal or fetal outcomes; and (3) initiating interventions to optimize outcomes. The fundamental principles of preconception counseling and care have been outlined recently in a report by the CDC and the Agency for Toxic Substances and Disease Registry (ATSDR) Preconception Care Work Group.<sup>7</sup> Preconception care involves more than a single clinical visit; it is a process of ongoing care and interventions integrated into primary care that addresses the needs of women during the different stages of reproductive life.

In addition to the general components of preconception counseling and

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care that are appropriate for all women of reproductive age, HIV-infected women have specific needs that must be addressed by their primary care providers. Components of this HIV-specific preconception care have been outlined by the United States Public Health Service (USPHS) Perinatal HIV Guidelines Working Group.<sup>1</sup> Guidelines for integrating preconception care and HIV testing into a comprehensive reproductive health care model are shown in Table 1.

Assessing the need and options for family planning, implementation and follow-up of family planning and safer sex strategies, as well as preconception counseling, are routine components of primary health care for women with HIV infection.

### Contraception and Preventing Transmission of HIV and Other Sexually Transmitted Infections

HIV-infected women should use safe and reliable means of contraception until ready to conceive. A clinician's recommendations regarding contraceptive options for HIV-infected women need to take into account the convenience and safety of the method, efficacy in preventing pregnancy, prevention of transmission of HIV and other sexually transmitted infections (STIs), and potential interactions with antiretroviral drugs and other medications. For a review of contraceptive methods for HIV-infected women, consult the March 2005 issue of the International Planned Parenthood Federation *IPPF Medical Bulletin*.<sup>9</sup>

It is important to counsel HIV-infected women on the efficacy of barrier protection methods for decreasing the transmission of HIV and other STIs, and for providing protection from acquiring more virulent or drug-resistant HIV strains. There is strong evidence that male condom use reduces the risk of transmission of HIV, gonorrhea, chlamydia, and herpes simplex virus.<sup>10-12</sup> Experts estimate that consistent use of male condoms decreases the risk of HIV transmission by approximately 80% to 90%.<sup>13</sup> It must be considered, however, that condom use is associated with relatively higher pregnancy rates

**Table 1. Steps for Integrating Preconception Care and HIV Testing into a Comprehensive Reproductive Health Care Model**

- Provide continuous preconception counseling for women of reproductive age: ask about pregnancy intentions **every woman, every visit.**
- Provide family planning services integrated in HIV clinics.
- Provide rapid HIV testing of patients and their partners in obstetrics and gynecology and HIV clinics.
- Provide preconception education, evaluation, and risk assessment prior to pregnancy attempts.
- Provide integrated obstetrics and HIV services for HIV-infected pregnant women.
- Provide on-site case management, peer educators, and psychological services integrated into prenatal care.
- Provide state-of-the-art medical care to every woman.
- Provide rapid HIV testing in hospital delivery rooms for all unregistered or untested pregnant women.
- Provide linkages to HIV care for HIV-infected women and children by collaborating with pediatric services and family-centered clinics.

Adapted from the Perinatal HIV Guidelines Working Group, 2006.

than hormonal contraceptive methods or the intrauterine device (IUD).<sup>14</sup> With regard to female condoms, although research is more limited, there is some evidence that they provide effective protection against STIs in women.<sup>15</sup> Unfortunately, female condoms are not well known or their use is considered awkward by many women. Nevertheless, women who receive instruction in female condom use and have the opportunity to practice application and removal techniques on a pelvic model have an increased likelihood of using the method successfully and of viewing it favorably.<sup>16</sup>

The use of dual-protection contraception methods—hormonal contraception or IUD to prevent pregnancy and a barrier method to prevent HIV and other STIs—is ideal. However, research on the use of dual-protection methods is limited, and conclusions on promoting single methods versus dual methods are mixed. Clinicians need to tailor their contraception-counseling messages to respond to an individual woman's desires and motivations. Offering clear, positive, and reinforcing messages to women about the dual benefits of consistent condom usage,

and providing an array of contraceptive choices, may decrease the likelihood of HIV and STI transmission and unplanned pregnancy.<sup>17</sup>

### Preconception Education and Risk Assessment

It is advantageous for clinicians to counsel women about the importance of eliminating alcohol and substance use and about smoking cessation during preconception in order to prevent complications that can occur during pregnancy. Smoking during pregnancy is associated with several adverse outcomes including low-birth-weight, small-for-gestational-age neonates, placental abruption, premature rupture of membranes, an increase in sudden infant death syndrome, and neonatal respiratory problems.<sup>18</sup> Alcohol use by pregnant women is associated with fetal alcohol syndrome and fetal alcohol effects.<sup>19</sup> Cocaine use during pregnancy is associated with premature delivery, placental abruption, in utero fetal stroke, maternal stroke, and low-birth-weight neonates.<sup>20</sup> Hence, preconception referral to drug and alcohol counseling and rehabilitation services is advisable.

Preconception is also the time to optimize an HIV-infected woman's nutritional status. In particular, the initiation of preconception folic acid supplementation is highly effective in decreasing the occurrence of fetal neural tube defects (NTDs) both in low-risk women and in high-risk women who have had a previous pregnancy resulting in a neonate with a NTD.<sup>21</sup> Current practice recommends prenatal supplementation with 400 µg of folic acid daily for low-risk women of reproductive age.<sup>22</sup> For high-risk women, supplementation with 4 mg per day, started at least 1 month before conception and continued through the first trimester is recommended.<sup>23</sup>

Accurate reproductive, genetic, and psychiatric histories are essential components of preconception health care. They are particularly important for HIV-infected women, who often have psychiatric comorbidities, especially depression disorders.<sup>24</sup> We recommend screening all HIV-infected women with a simple screening test for depression, such as the Center for Epidemiologic Studies-Depression Scale or the Beck Depression Inventory, and referring those who test positive for formal psychological evaluation.

Preconception screening for infectious diseases should include HIV-1 genotype before starting antiretroviral therapy, as well as screening for varicella, rubella, rubeola, mumps, cytomegalovirus, toxoplasmosis, hepatitis B and C virus titers, and tuberculosis (TB). If appropriate, evaluation for prophylaxis for opportunistic infections and administration of vaccines for influenza, pneumococcal infections, and hepatitis A and B viruses should occur before pregnancy as well. Additionally, both partners should be screened for genitourinary tract infections and treated if indicated.

In addition to maintaining optimal HIV management, clinicians should attain medical control of other chronic illnesses such as diabetes, hypertension, or lupus in HIV-infected women before pregnancy is attempted. The risks and effects of pregnancy on each medical condition, and the effects of the medical condition on pregnancy outcomes,

should be explained so that the patient can make an informed decision about becoming pregnant. For complicated cases, it may be prudent to have the patient seen by a maternal fetal medicine specialist, who can provide detailed information about pregnancy and coexisting medical conditions.

### Pregnancy and HIV Infection

Education and counseling for HIV-infected women about perinatal HIV transmission risks, strategies to reduce those risks, the potential effects of HIV or its treatment on pregnancy, and the risk of transmission during breastfeeding, allows patients to be fully aware of the issues concerning HIV infection and pregnancy before conception. The use of potent antiretroviral therapy has reduced the risk of mother-to-child transmission to 1% to 2% when HIV infection is optimally managed,<sup>25</sup> and pregnancy has not been shown to have an adverse effect on the course of HIV disease.<sup>26</sup> Likewise, HIV infection and its treatment have not been demonstrated to adversely affect pregnancy outcomes, and overall, the benefits of antiretroviral therapy outweigh its risks.<sup>27</sup>

When pregnancy is desired by a woman who is maintained on antiretroviral therapy, a stable, maximally suppressed maternal HIV-1 RNA level before conception is necessary. If the woman does not meet the recommendations for antiretroviral therapy as indicated by the USPHS guidelines, therapy can be initiated in the second trimester of pregnancy, when organogenesis is complete, to prevent mother-to-child transmission.<sup>1</sup> When prescribing antiretroviral therapy for women of childbearing age, it is important to choose agents known to be effective in reducing the risk of perinatal HIV transmission and to avoid those with known teratogenicity. (For more information refer to the United States Department of Health and Human Services [DHHS] perinatal guidelines available at <http://www.aidsinfo.nih.gov/guidelines>.) Women of childbearing potential should avoid regimens that include efavirenz, owing to con-

cerns of teratogenicity that are based on animal studies and several case reports in humans describing NTDs in fetuses exposed to efavirenz in the first trimester.<sup>1, 28, 29</sup> Of the medications that have been adequately studied, with the exception of efavirenz, adverse pregnancy outcomes (preterm birth, low birth weight, and intrauterine growth retardation) and congenital abnormalities in antiretroviral-treated women are similar to those reported in uninfected women.<sup>1, 30, 31</sup> Therapy-associated adverse effects that may affect maternal-fetal health outcomes such as hyperglycemia, anemia, and hepatotoxicity should be reviewed. Short-term and potential long-term neonatal toxicities should be discussed.

Disclosure of HIV status to partners before pregnancy should be encouraged, but for some HIV-infected women, disclosure can be very unsettling. Fear of rejection, stigmatization, or domestic violence, and a history of childhood sexual abuse can be reasons behind a woman's reluctance to disclose her HIV status to a partner. Providing support through accessible psychological services can help women through this very sensitive process. Following disclosure, offering on-site rapid HIV testing of a partner and an infected woman's untested children can expedite the detection of their HIV status and, if necessary, their linkage to immediate care.

When all preconception issues have been identified and addressed and the patient is physically and emotionally prepared for pregnancy, the couple must have a plan for conceiving that minimizes the risk of transmission to the uninfected partner. Before any attempts at conception, the man should obtain a semen analysis to exclude azospermia and symptomatic epididymitis.

Interventions in planned pregnancies between serodiscordant couples using assisted reproduction methods can minimize the risk of HIV transmission to the uninfected partner. For serodiscordant heterosexual couples in which the woman is HIV-infected and the man is HIV seronegative, low-cost self-insemination techniques—such as using a needleless syringe or a diaphragm to

insert semen close to the cervix—are recommended. Timing these options to occur during ovulation can be calculated by using the simple and effective Spinbarkeit test: observing when the cervical mucous changes from a thick consistency to a thin, stretchy consistency. Alternatively, over-the-counter ovulation-prediction kits that detect the pre-ovulation surge of luteinizing hormone, or monitoring basal-body temperatures can also estimate the timing of ovulation. Information about these methods should be included as part of preconception counseling for couples interested in pregnancy. For a presentation that can be provided as patient information for serodiscordant couples, see “Thinking About Having a Baby? Preconception Counseling for HIV Discordant Couples” available at <http://www.womenchildrenhiv.org/>.

The successful use of timed conception with no unprotected intercourse outside of conception has been reported. The 4 cases of HIV transmission that occurred in this report resulted from exposure outside of conception attempts: 2 women seroconverted in their seventh month of pregnancy, and 2 seroconverted postpartum.<sup>32</sup>

It is the clinician’s responsibility to counsel the couple about the risks of attempting conception through unprotected intercourse. If, despite counseling, the couple decides to attempt pregnancy through unprotected intercourse, a discussion informing them that attaining an undetectable HIV RNA level through adherence to antiretroviral therapy before conception attempts has been shown to lower the risk of transmission is prudent.<sup>33</sup> In addition, it is important that couples are counseled to use condoms during all sexual encounters other than conception attempts during ovulation. In most cases, however, serodiscordant couples should be discouraged from conceiving naturally through unprotected intercourse owing to the cumulative risk of HIV transmission to the uninfected partner.<sup>34</sup>

When both partners are HIV seropositive, unprotected intercourse can increase the risk of transmitting new viral strains. However, a variety of as-

sisted reproduction techniques can mitigate this risk.<sup>35</sup> Those most commonly used include sperm preparation techniques such as sperm washing, column purification, and intracytoplasmic sperm injection. Sperm washing has shown substantial success in pregnancy outcomes with no HIV transmission to women.<sup>36,37</sup> Unfortunately, access to assisted reproduction techniques is limited and costly.<sup>35</sup>

### Effective Models of Care for HIV-infected Women

Health care models that emphasize collaboration between clinicians and agencies providing care and services to HIV-infected women of childbearing age have demonstrated effectiveness in improving health outcomes and reducing perinatal HIV transmission.<sup>38</sup> “One-stop-shopping” models of care provide primary health care, HIV specialist care, gynecology, prenatal care, nutrition services, pharmacy, case management, peer educator support, psychological services, and the provision of pediatric health care services and child day care in one location.<sup>39</sup> Over the past 5 years, this integrated-services model has enabled the Partnership Comprehensive Care Practice of Drexel University College of Medicine to achieve a 0% mother-to-child transmission rate among more than 130 pregnant women treated for HIV infection.

In addition, at our facility, the integration of a Title X Family Planning Program into an existing HIV clinic offers patients counseling and treatment for birth control, STIs, education about transmission of HIV, pregnancy options counseling, preconception counseling, and HIV testing for partners.

### Conclusion

The advent of potent antiretroviral therapy has allowed women with HIV to live longer and healthier lives. This circumstance coupled with the dramatic reduction in mother-to-child transmission seen over the past decade has changed the quality of life for HIV-infected women. It is crucial that a

new strategic plan for HIV prevention and a proactive approach to reproductive health care be implemented by health care providers who care for these women. Preconception counseling for HIV-infected women should be a routine part of primary health care. Such counseling identifies individual risks and corresponding interventions, provides personalized and nonjudgmental education so that a woman is fully informed of her choices, and provides access to integrated services that address all of a woman’s health and psychological needs. Collaborative partnerships between family planning, obstetrics and gynecology, pediatric, and HIV and AIDS services as part of an integrated system of care will likely improve our ability to provide high-quality care and be more responsive to the needs of HIV-infected women. Primary care providers for HIV-infected women have numerous opportunities to provide preconception care prior to pregnancy attempts. “Every woman, every visit” is a reminder to provide preconception health care as a routine component of primary care.

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### References

- 1. Perinatal HIV Guidelines Working Group.** Public Health Service Task Force Recommendations for Use of Antiretroviral Drugs in Pregnant HIV-1-Infected Women for Maternal Health and Interventions to Reduce Perinatal HIV-1 Transmission in the United States - Supplement: Safety and Toxicity of Individual Antiretroviral Agents in Pregnancy - October 12, 2006. Available at: <http://aidsinfo.nih.gov/Guidelines/GuidelineDetail.aspx?MenuItem=Guidelines&Search=Off&GuidelineID=9&CIassID=2>. Accessed: July 19, 2007.
- 2. Centers for Disease Control and Prevention (CDC).** Achievements in public health. Reduction in perinatal transmission of HIV infection—United States, 1985–2005. *MMWR Morb Mortal Wkly Rep.* 2006;55(21):592–597.
- 3. Hankins C, Tran T, Lapointe N.** Sexual behavior and pregnancy outcome in HIV-infected women. Canadian Women’s HIV Study Group. *J Acquir Immune Defic Syndr Hum Retrovirol.* 1998;18:479–487.
- 4. Chen JL, Philips KA, Kanouse DE, Collins RL, Miu A.** Fertility desires and intentions of

- HIV-positive men and women. *Fam Plann Perspect*. 2001;33:144-52, 165.
5. **Ogilvie G**, Palepu A, Remple VP, et al. Fertility intentions of women of reproductive age living with HIV in British Columbia, Canada. *AIDS*. 2007;21:s83-s88.
6. **Finer LB**, Henshaw SK. Disparities in rates of unintended pregnancy in the United States, 1994 and 2001. *Perspectives on Sexual and Reproductive Health*. 2006;38:90-96.
7. **Centers for Disease Control and Prevention**. Recommendations to improve preconception health and health care—United States. Available at: <http://www.cdc.gov/mmwr/preview/mmwrhtml/rr5506a1.htm>. Accessed: February 26, 2007.
8. **American College of Obstetricians and Gynecologists**. ACOG Committee Opinion number 313, September 2005. The importance of preconception care in the continuum of women's health. *Obstet Gynecol*. 2005;106(3):665-666.
9. **International Medical Advisory Panel (IMAP) of the International Planned Parenthood Federation (IPPF)**. IMAP statement on the management of HIV infection within sexual and reproductive health services. *IPPF Medical Bulletin*. 2005;39:1-5.
10. **National Institute of Allergy and Infectious Diseases**. Workshop Summary: Scientific Evidence on Condom Effectiveness for Sexually Transmitted Disease (STD) Prevention. Available at: <http://www3.niaid.nih.gov/research/topics/STI/pdf/condomreport.pdf>. Accessed: July 20, 2007.
11. **Warner L**, Stone KM, Macaluso M, Buehler JW, Austin HD. Condom use and risk of gonorrhea and chlamydia: a systematic review of design and measurement factors assessed in epidemiologic studies. *Sex Transm Dis*. 2006;33:36-51.
12. **Wald A**, Langenberg AG, Krantz E, et al. The relationship between condom use and herpes simplex virus acquisition. *Ann Intern Med*. 2005;143:707-713.
13. **Steiner MJ**, Cates W. Condoms and sexually-transmitted infections. *N Engl J Med*. 2006;354:2643.
14. **Cates W**, Steiner MJ. Dual protection against unintended pregnancy and sexually transmitted infections: what is the best contraceptive approach? *Sex Transm Dis*. 2002;29:168-174.
15. **Vijayakumar G**, Mabude Z, Smit J, Bekinska M, Lurie M. A review of female-contraception effectiveness: patterns of use and impact on protected sex acts and STI incidence. *Int J STD AIDS*. 2006;17:652-659.
16. **Van DeVanter N**, Gonzales V, Merzel C, Parikh N, Celentano D, Greeberg J. Effect of an STD/HIV behavioral intervention on women's use of the female condom. *Am J Public Health*. 2002;92:109-115.
17. **Davis KR**, Weller SC. The effectiveness of condoms in reducing heterosexual transmission of HIV. *Fam Plann Perspect*. 1999;31:272-279.
18. **Torpy JM**, Lynn C, Glass RM. Smoking and pregnancy. *JAMA*. 2005;293:1286.
19. **American Academy of Pediatrics Committee on Substance Abuse and Committee on Children with Disabilities**. Fetal alcohol syndrome and alcohol-related neurodevelopmental disorders. *Pediatrics*. 2000;106:358-361.
20. **Ebrahim SH**, Gfroerer J. Pregnancy-related substance use in the United States during 1996-1998. *Obstet Gynecol*. 2003;101:374-379.
21. **Cheschier N**, ACOG Committee on Practice Bulletins-Obstetrics. ACOG practice bulletin. Neural tube defects. Number 44, July 2003. (Replaces committee opinion number 252, March 2001). *Int J Gynaecol Obstet*. 2003;83:123-133.
22. **Standing Committee on the Scientific Evaluation of Dietary Reference Intakes**. *Dietary Reference Intakes for Thiamin, Riboflavin, Niacin, Vitamin B6, Folate, Vitamin B12, Pantothenic Acid, Biotin, and Choline*. Washington, DC: The National Academies Press; 1998
23. **Centers for Disease Control and Prevention**. Recommendation for the use of folic acid to reduce the number of cases of spina bifida and other neural tube defects. *MMWR Morb Mortal Wkly Rep*. 1992;41:RR-14.
24. **Morrison ME**, Petitto JM, Ten Have T, et al. Depressive and anxiety disorders in women with HIV infection. *Am J Psychiatry*. 2002;159:789-796.
25. **Cooper ER**, Charurat M, Mofenson L, et al. Combination antiretroviral strategies for the treatment of pregnant HIV-1-infected women and prevention of perinatal HIV-1 transmission. *JAIDS*. 2002;29:484-494.
26. **Alliegro MB**, Dorrucchi M, Phillips AN, et al. Incidence and consequences of pregnancy in women with known duration of HIV infection. Italian Seroconversion Study Group. *Arch Intern Med*. 1997;157:2585-2590.
27. **Tuomala RE**, Watts DH, Li D, et al. Improved obstetric outcomes and few maternal toxicities are associated with antiretroviral therapy, including highly active antiretroviral therapy during pregnancy. *JAIDS*. 2005;38:449-473.
28. **De Santis M**, Carducci B, De Santis L, Cavaliere AF, Straface G. Periconceptional exposure to efavirenz and neural tube defects. *Arch Intern Med*. 2002;162:355.
29. **Fundaro C**, Genovese O, Rendeli C, Tamburrini E, Salvaggio E. Myelomeningocele in a child with intrauterine exposure to efavirenz. *AIDS*. 2002;16:299-300.
30. **Townsend CL**, Tookey PA, Cortina-Borja M, Peckham CS. Antiretroviral therapy and congenital abnormalities in infants born to HIV-1-infected women in the United Kingdom and Ireland, 1990 to 2003. *JAIDS*. 2006;42:91-94.
31. **Lambert JS**, Watts DH, Mofensen L, et al. Risk factors for preterm birth, low birth weight, and intrauterine growth retardation in infants born to HIV-infected pregnant women receiving zidovudine. *AIDS*. 2000;14:1389-1399.
32. **Mandelbrot L**, Heard I, Henrion E, Henrion R. Natural conception in HIV-negative women with HIV-infected partners. *Lancet*. 1997;349:850-851.
33. **Barreiro P**, del Romero J, Leal M, et al. Natural pregnancies in HIV-serodiscordant couples receiving successful antiretroviral therapy. *JAIDS*. 2006;43:324-326.
34. **Williams CD**, Finnerty JJ, Newberry YG, West RW, Thomas TS, Pinkerton JV. Reproduction in couples who are affected by human immunodeficiency virus: medical, ethical, and legal considerations. *Am J Obstet Gynecol*. 2003;189:333-341.
35. **Stern JE**, Cramer CP, Garrod A, Green RM. Access to services at assisted reproductive technology clinics: a survey of policies and practices. *Am J Obstet Gynecol*. 2001;184:591-597.
36. **Sauer MV**, Chang PL. Establishing a clinical program for human immunodeficiency virus 1-seropositive men to father seronegative children by means of in vitro fertilization with intracytoplasmic sperm injection. *Am J Obstet Gynecol*. 2002;186:627-633.
37. **Semprini AE**, Vucetich A, Hollander L. Sperm washing, use of HAART and role of elective Caesarean section. *Curr Opin Obstet Gynecol*. 2004;16:465-470.
38. **Aaron E**, Levine AB. Gynecologic care and family planning for HIV-infected women. *AIDS Read*. 2005;15(8):420-423.
39. **Clark J**, Samson S, Simpson BJ, et al. Promising strategies for preventing perinatal HIV transmission: model programs from three states. *Matern Child Health J*. 2006;10:367-373.

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