IVA. NUCLEOSIDE & NUCLEOTIDE ANALOGUES

i. Approved Nucleoside Analogues: Reevaluating Initial Therapy

by Jill Cadman

More and more people are embarking on the treatment merry-go-round following the aggressive stance outlined in the Public Health Service's antiretroviral treatment guidelines. There are numerous treatment options for antiretroviral-naïve patients. The sequence in which these drugs are used could be an important consideration for achieving maximum long- and short-term benefit. The choice of nucleoside analogue for an initial combination regimen may preserve future treatment options by averting the development of cross-resistance to other drugs that are currently available or in development. This is especially important since no one combination has yet proven invulnerable to virologic failure, requiring patients and their physicians to always plan ahead. Although there are not a great deal of hard data on this topic, new information is emerging from comparisons between several of the nucleoside analogues that gives some insight into potentially optimal sequencing.

What's on First?

In the battle to be nucleoside analogue of first choice in the foundation of a protease inhibitor-containing regimen, Bristol-Myers Squibb's upstart d4T has proved to be a formidable rival to Glaxo Wellcome's longstanding behemoth, AZT. As of December 1997, d4T has surpassed AZT in total prescription volume. (This according to the consulting firm that Bristol-Myers uses for market research. Not surprisingly, the company Glaxo uses shows AZT-containing regimens still holding a greater share of the market than those containing d4T.) The more important issue is whether d4T is as effective as AZT in treatment-naïve patients. A recently concluded trial, ACTG 306, compared the combinations of AZT/3TC and d4T/3TC as first-line therapy (Kuritzkes 1998). ACTG 306 enrolled 299 antiretroviral-naïve participants with CD4 counts of 200 to 600 between December 1995 and July 1996. All study subjects had low median baseline viral loads of about 10,000 (as measured with the bDNA assay). Participants were randomized to a d4T or ddI "limb" and then further randomized to one of three arms within the limb (d4T limb: d4T alone, d4T/3TC or AZT/3TC; ddI limb: ddI alone, ddI/3TC or AZT/3TC). After 24 weeks, participants receiving monotherapy had 3TC added to their regimens for a further 24 weeks.

Three-quarters of the trial participants remained on therapy through week 48, and 90% of those were available for follow-up evaluation:

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Effect of T-20 on HIV RNA and CD4 Cell Count

Dose (BID)	*Baseline VL*	*Day 14 VL*	*VL change*	*Baseline CD4*	*Day 14 CD4*	*CD4 change*
3 mg	4.82	4.7	-0.11	248	207	-41
10 mg	5.12	5.06	-0.06	357	344	-13
30 mg	4.95	4.47	-0.48	410	431	+21
100 mg	4.2	<2.70*	-1.50	322	374	+52

* All participants on 100 mg BID had plasma HIV RNA levels below 500 copies at day 14; VL = viral load

(Levin 1998)

TAG will continue to monitor novel antiretroviral agents as they make their way through the development pipeline.

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ABT-378. This is a new protease inhibitor currently in development by Abbott Laboratories. *In vitro*, it is approximately ten times more potent against HIV than ritonavir. Like available protease inhibitors, ABT-378 is processed by the cytochrome p450 hepatic enzyme system, and interacts strongly with ritonavir. Even small doses of ritonavir – 100 mg a day -- were sufficient to radically raise levels of ABT-378 in a recent dose-ranging study in healthy volunteers. The main side effects noted so far include loose stools or mild diarrhea. Resistance profiles so far suggest that people with extensive exposure to other inhibitors should be at least partially sensitive to ABT-378 (Japour 1998)

PNU-140690, Bristol Myers 232632, and Parke-Davis 178390. These protease inhibitors are the NNRTIs of protease inhibition -- that is to say, they are expected to block the HIV protease, but in a different fashion than currently marketed drugs. Because of this different mechanism, they could prove highly effective against strains of HIV that are resistant to other protease inhibitors.

FTC. This nucleoside analogue, which is now in Phase I/II trials is more potent than 3TC *in vitro*. Results from a small phase I study of FTC in HIV-infected patients found that patients treated with 25 mg FTC twice daily ha a 1.4 log drop in plasma HIV RNA levels after fourteen days, while patients treated with 200 mg FTC once daily experienced a 2.1 log drop. Both doses were reported to be well-tolerated. Unfortunately, FTC is completely cross-resistant with 3TC (Levin 1998).

Bis-Poc PMPA. This nucleotide analogue reverse transcriptase inhibitor, like adefovir (to which the drug is related) is manufactured by Gilead Sciences. Early studies have suggested that bis-poc PMPA may be more potent than adefovir. In a phase I dose-ranging study of bis-poc PMPA, thirty-six patients were treated with 75, 150 or 300 mg once daily. Most of the participants were antiretroviral experienced. These patients were treated for 28 days, and then had plasma HIV RNA levels measured:

Effect of BIS-POC PMPA on Plasma HIV RNA Levels

Treatment	*Viral load at day 28 (log)*
Placebo	-0.06
25 mg PMPA	-0.32
200 mg PMPA	-0.44
300 mg PMPA	-1.22

(Levin 1998)

The side effects most commonly observed in this study were elevated liver enzymes and high creatine kinase levels. Further studies of this drug are ongoing.

MKC-442. This NNRTI from Triangle Pharmaceuticals looks promising in very early phase I dose-finding studies. In a study of 48 HIV-infected patients, treated with 500 mg BID, median viral load reduction was 1.41 log after 8 days and 1.30 log after 15 days. Adverse events observed included headache, nausea, mild rash, and modest elevations in liver enzymes. Further studies are being conducted looking at 750 mg and 1000 mg twice a day. MKC-442 is likely to be cross resistant with Sustiva (Layne 1998).

T-20. This genetically engineered peptide is a fusion inhibitor (the process by which HIV attaches itself to the outside of a cell before hijacking the cell's genetic material). It will probably have to be given by a continuous infusion through a small computerized pump, about the size of a beeper, with a flexible catheter. Early studies suggest significant antiviral activity at higher doses:

Nelfinavir pre-treated patients. Agouron has suggested that, because the D30N mutation which arises most frequently following nelfinavir failure is not seen with other protease inhibitors, it may be easier to treat nelfinavir failures than people who have not responded to other protease inhibitors. To examine the effect of the ritonavir/saquinavir combination in patients with prior nelfinavir therapy, investigators conducted a study of patients who failed nelfinavir therapy in Agouron 506, which compared d4T to d4T/nelfinavir and in Agouron 511, which compared AZT/3TC to AZT/3TC/nelfinavir, both in treatment naive patients. Nelfinavir failure was defined as two consecutive viral load measurements of more than 500 copies on the bDNA assay. All patients defined as failing a nelfinavir-containing regimen were switched to a combination of d4T/3TC/RTV/SQV. Average duration of nelfinavir therapy while viral load was more than 500 copies was approximately 55 weeks. Baseline viral burden was approximately 40,000 copies, and baseline CD4 was 222 cells.

Seventy percent of study participants had the D30N mutation at baseline (the primary mutation selected for by nelfinavir in vitro), and 30% had the L90M mutation. Nineteen patients who have reached week 24 of the study are included in this analysis. Ninety percent of participants became undetectable at weeks four and twelve. At six months, 68% were undetectable (<500 copies on the bDNA assay). Of patients who have been treated for longer, 62.5% of eight patients treated to 32 weeks were undetectable.

Comparing patients who had the L90M mutation at baseline with patients who did not have the mutation at baseline, approximately 60% of both groups had viral load below the limit of detection at week twenty-four, suggesting that this mutation did not increase risk of virologic failure in response to this regimen. However, prior 3TC experience did suggest some risk of therapeutic failure with this 3TC-containing regimen (Forum for Collaborative HIV Research 1998).

Multiple Protease pre-treatment. Treatment of patients who have experienced virologic failure on multiple protease inhibitors is in a state of complete infancy. Only a few observational studies have been reported at conferences.

An Australian group reported on what they termed the "surprising" success of using six drugs (d4T, ddI, 3TC, nevirapine, nelfinavir, saquinavir) in patients who had already failed saquinavir, indinavir, ritonavir and multiple nucleoside analogues. By week sixteen, 9/12 patients who were able to tolerate the regimen had viral load counts below detectable levels (Workman 1998)

Another study from Stanford looked at patients with prior virologic failure on saquinavir and nelfinavir. These 16 patients were treated with nevirapine, indinavir, and two nucleoside analogues. Six of eleven patients reached undetectable viral loads using this regimen, however only three maintained suppression beyond week 20 (Lawrence 1998).

NEW DRUGS IN THE PIPELINE

A variety of new drugs currently in the pipeline may offer hope to people who have failed currently available treatments. Although little is known about these therapies, many of them show promise in either simplifying initial regimens, or in offering hope to people for whom current therapies have failed.

In a study of nelfinavir with two nucleoside analogues in sixteen patients experiencing failure on saquinavir regimens, the therapeutic switch produced only a transient viral load drop (0.59 log at two weeks) which rapidly returned to baseline by week 12 in most patients . Subsequently, however, 11 of these individuals were given indinavir (1,000 mg TID), nevirapine and two NRTIs after the nelfinavir failure. The median four-week viral load drop was 1.58 log, and 6/11 (55%) developed undetectable viral load (<400 copies), but only 3 (27%) stayed maximally suppressed beyond 20 weeks (Lawrence 1998).

Finally, a complicated study from Australia looked at three groups of patients:

* Group 1: 34 subjects who received therapy with Invirase and either added ritonavir at 400 mg or 600 mg BID while adjusting the dose of saquinavir
* Group 2: 14 individuals who first received Invirase and were then switched to indinavir, and
* Group 3: 14 protease inhibitor-naïve patients who initiated therapy with ritonavir/saquinavir

Median prior duration of saquinavir use in groups one and two was 31-35 weeks.

Effect of Adding Ritonavir or Switching to Indinavir in Saquinavir-Experienced Patients

	SQV to RTV/SQV	*SQV to IDV*	*1st RTV/SQV*
N	34	14	14
Baseline viral load	4.27 log	5.23 log	4.98 log
Baseline CD4	216	142	265
Prior SQV (wks)	31	36	--
N(%) changing 2nd drug	8(20%)	9(56%)	--
28 wk viral load change (log)	-1.28	-1.82	-2.32
28 wk viral load BLQ*	52%	50%	80%
28 wk CD4 change	+77	+76	+72

* BLQ=Below Limit of Quantification

(Bodsworth 1998)

In this study, baseline genotypic mutations did seem to predict outcome, though only five patients were available for genotypic analysis.

Indinavir pre-treated patients. Reports from a variety of investigators have examined use of ritonavir/saquinavir in patients with prior indinavir failure. Although these study have been small (usually looking at 10-20 patients), results have been almost uniformly negative, with initial reductions in viral load that quickly rebound to baseline. For instance, in one characteristic study, 19 patients at the San Francisco General Hospital who had failed indinavir were treated with ritonavir/saquinavir and, if possible, two new nucleoside analogues. Patients were switched a median of 2.1 months after treatment failure was detected. Despite an initial 1.6 log decline in viral load, by 24 weeks only two of 15 patients had undetectable viral loads (Forum for Collaborative HIV Research 1998).

Ritonavir pre-treated patients. In a retrospective chart review of 14 patients who, while experiencing virologic failure on ritonavir and two nucleoside analogues, added saquinavir. Virologic response was modest, and the authors conclude that this kind of therapeutic intensification strategy is not recommended (Hellinger 1997).

phenotypic resistance assay, suggested that 77% to 95% of isolates with ten-fold or greater resistance to once protease inhibitor had at least four-fold resistance to all three of the other marketed protease inhibitors:

Phenotypic Resistance to One PI is Associated With Cross Resistance to Three Others

> 10-fold resistance to:		*% Cross-Resistant To:*							
		IDV		*NFV*		*RTV*		SQV	
	N	4-fold	10-fold	4-fold	10-fold	4-fold	10-fold	4-fold	10-fold
IDV	224	--	--	86%	78%	95%	78%	83%	66%
NFV	277	87%	63%	--	--	90%	70%	77%	63%
RTV	261	93%	67%	87%	74%	--	--	78%	62%
SQV	220	90%	67%	89%	79%	95%	74%	--	--

(Hertogs 1998)

Preliminary data from studies of salvage therapy patients who have failed saquinavir have suggested that switching therapies early in the course of virologic failure may be more successful than continued treatment with a failing regimen:

Duration of Prior Saquinavir May Predict Response to Future Therapies

Duration of Prior SQV	*New treatment*	*Viral Load*	*Reference*
36 wks		-1.83	*Bodsworth 1998*
52 wks	IDV, SQV-SGC	-1.20	*Schapiro 1997*
112 wks	IDV, SQV-SGC	-0.58	*Para 1997*

(Forum for Collaborative HIV Research 1998)

However the benefits of such rapid changes must be weighed against the risks of rapidly using up therapies, and the preliminary reports of continued therapeutic efficacy despite virologic failure on HAART.

So far, as would be predicted by the genotypic and phenotypic resistance data, studies of HAART regimens in patients who have already failed at least one such regimen have been small and frankly discouraging. While several ongoing studies, including ACTG 359 and ACTG 372, are looking at new regimens for patients with prior indinavir failure, extensive cross resistance between currently available drugs is likely to limit the success of salvage therapy. Certainly that is the suggestion of a number of smallish salvage therapy studies that have already been reported.

Saquinavir pre-treated patients. A number of studies have suggested that, while response of saquinavir pre-treated patients to further HAART therapy is varied, median responses are significantly smaller than those which might be expected in therapy-naïve patients.

The most significant study of treatment in patients with prior saquinavir therapy was ACTG 333, (see below), which demonstrated cross resistance between saquinavir and indinavir.

In virologic data from ACTG 333, baseline genotypic mutations associated with resistance to saquinavir did not appear to predict virologic success or failure in switching therapies, raising the concern that genotypic analysis may be of little clinical utility.

ProCom. However, investigators have not yet abandoned the notion of maintenance therapies. In Europe, the British Medical Research Council is currently enrolling the ProCom study, in which patients with CD4 counts below 350 or viral load greater than 20,000 will be treated with d4T, ddI, saquinavir-soft gel capsule formulation, and nelfinavir for at least four months, with possible extensions to five or six months in patients who have not yet achieved undetectable viral load. After a maximum of 24 weeks of therapy, patients with undetectable viral load will be randomly assigned to continue all four drugs, reduce therapy to d4T/ddI, or to reduce therapy to saquinavir with nelfinavir.

ADAM is a similar study that will treat patients with d4T/3TC/nelfinavir/saquinavir for six months. Half of patients with undetectable viral load will then be randomly assigned to continue the quadruple-drug regimen, while half will be assigned to receive either d4T/3TC or nelfinavir/saquinavir. After twelve months in the study, those patients remaining on quadruple-drug treatment will be assigned to subtract either the nucleosides or the protease inhibitors from their treatment regimens. This study may allow us to determine whether a longer induction phase makes a difference in the success of maintenance therapy.

Investigators on these studies have not yet determined how the failures of ACTG 343 and Trilège will impact these studies.

In addition, the ACTG is continuing to work on studies that will evaluate strategies for reducing the number of anti-HIV drugs over the long term.

MANAGING PATIENTS IN WHOM ANTIRETROVIRAL THERAPY FAILS

At the 37th Annual Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC) in September 1997, Steven Deeks from the University of California/San Francisco presented data from a "real world" clinical setting looking at rates of virologic failure following initiation of HAART. The findings were shocking because they documented much higher rates of failure than had been noted in the setting of ongoing controlled clinical trials. Of 196 patients who began therapy, 136 patients stayed on treatment and were evaluable. Patients were treated with a protease inhibitor and at least one nucleoside analogue. Virologic success was defined as having plasma viral load below 500 during the two most recent visits. In this study, only 47% of patients had a successful response to HAART. It should be noted that this patient cohort included heavily pre-treated patients who were unable to combine a new nucleoside analogue with their protease inhibitor, but who instead recycled previously used therapies. As a consequence, clinical success using HAART in treatment-naïve patients would be expected to occur more frequently. Nonetheless, this study suggests that clinical management of therapeutic failure will continue to be important for some time to come (Deeks 1997).

More optimistically, however, Deeks' team also found that, despite the ongoing presence of replicating virus in patients with virologic failure on HAART, the median CD4 cell count remained 103 cells above baseline after eleven months of treatment. Although this was not a randomized, controlled study, Dr. Deeks reports that clinical events in patients who maintained CD4 responses were few (Deeks 1998).

The question of what to do with these virologic treatment failures is a vexatious one. In 1995, Condra and colleagues demonstrated extensive overlap in the genetic resistance mutations that arise in response to different protease inhibitors. More recent data from Hertogs and colleagues, using the PR-RT Antivirogram

Some researchers became excited about the possibilities of induction/maintenance regimen in HIV therapy after data were presented on two patients from the INCAS study who were treated with a combination of AZT/ddI/nevirapine. After these patients' viral load was reduced below the limits of detection, both patients chose to stop ddI therapy due to toxicities, but continued on AZT/nevirapine. Viral load in both patients remained undetectable for over a year. By contrast, patients who began treatment with AZT/nevirapine did not achieve undetectable viral RNA levels (Hall 1997).

ACTG 343. The AIDS Clinical Trials group randomized 509 patients with a median CD4 cell count of 450, and a median plasma HIV RNA count of 19,872 copies were treated with AZT, 3TC and indinavir for twenty-four weeks. Forty-three percent of participants had prior AZT experience, and all were naïve to 3TC and protease inhibitors. Of these patients, 316 completed the initial treatment phase and entered the maintenance period, using AZT/3TC or indinavir monotherapy or continued triple-drug therapy. However, six of these patients had detectable HIV RNA values prior to beginning maintenance therapy, and one subject had no HIV RNA values recorded during maintenance therapy. Consequently, the outcome analysis includes only 309 patients. The interim review was conducted at a median of seven weeks of maintenance therapy for all three arms. All patients included in the analysis had plasma HIV RNA values of less than 200 when entering the maintenance phase. Efficacy analysis was based on the time to first detectable HIV RNA measurement of more than 200. At the time of analysis, there were a total of 37 endpoints.

ACTG 343: Proportion with Viral Load Rebound During Maintenance Therapy

Maintenance Regimen	*N*	*Endpoints*
Indinavir	101	16 (15.8%)
AZT/3TC	104	18 (17.3%)
AZT/3TC/indinavir	104	3 (2.9%)

(ACTG 343 Team 1998)

The triple-drug combination arm was superior to the AZT/3TC maintenance regimen (p=0.0003) and to indinavir monotherapy (p=0.0012). Prior AZT experience did not predict failure in either the AZT/3TC/indinavir or indinavir monotherapy arms, however pre-treatment did seem to predispose failure in the AZT/3TC arm.

Trilège. In a similar study conducted in Europe and known as TRILÈGE, 371 people with an average 363 CD4 cells and about 30,000 plasma HIV RNA copies were treated with the triple-drug combination for three months and then 277 were randomly assigned to either AZT/3TC, AZT/indinavir or AZT/3TC/indinavir. After about two months of maintenance therapy, results were as follows:

Trilège: Proportion with Viral Load Rebound During Maintenance Therapy

Maintenance Regimen	*N*	*Endpoints*
AZT/indinavir	93	16 (17.2%)
AZT/3TC	92	22 (23.9%)
AZT/3TC/indinavir	92	6 (6.5%)

(Raffi 1998)

Again, the triple-drug combination outperformed both subtractive regimens (p=0.01) (Raffi 1998).

A: 800 mg amprenavir TID + 800 mg saquinavir-SGC TID
B: 800 mg amprenavir TID + 800 mg indinavir TID
C: 800 mg amprenavir TID + 750 mg nelfinavir TID
D: 800 mg amprenavir TID + AZT/3TC

Viral Load Response to Double-Protease Therapy Including Amprenavir

	Baseline VL	*Wk2*	*Wk4*	*Wk8*	*Wk16*	*RNA<400*	*RNA<20*
APV + SQV	40,700	-1.83	-2.53	-2.17	-2.94	5/5	2/5
APV + IDV	67,600	-2.18	-2.23	-2.21	-3.75	5/6	4/6
APV + NFV	53,700	-1.78	-2.04	-2.29	-1.84	3/6	3/6
APV/AZT/3TC	12,300	-1.53	-1.49	-2.44	-2.79	2/3	2/3

(Eron 1998)

With so few participants, it is not yet easy to make out adverse event data:

Amprenavir Double Protease Therapy: Adverse Events by Regimen

	APV/SQV	*APV/IDV*	*APV/NFV*	*APV/AZT/3TC*
N	7	8	7	4
Diarrhea	5	3	5	2
Oral Parasthesias	1	4	3	2
Nausea/Vomiting	3	3	1	2
Headaches	3	3	0	2
Abdominal pain or Flatulence	3	2	0	2
Cutaneous	2	1	1	2

(Eron 1998)

The relative success of double-protease regimens in combination with nucleoside analogues has encouraged patients and doctors to experiment with these regimens. Currently, most use of double-protease regimens has been confined to salvage therapy in patients who have already failed a single-protease-containing regimen. However, studies are beginning to suggest that, because of cross-resistance, salvage therapy may not be terribly effective using currently available double-protease regimens (see below). Recently, the US Public Health Service Treatment Guidelines were updated to include the combination of ritonavir and saquinavir with nucleoside analogues in naïve patients., and Hoffmann-LaRoche is pushing heavily to add the combination of nelfinavir and saquinavir. Ongoing studies will hopefully clarify the role of double-protease therapy in treating HIV-infected patients.

Induction/Maintenance Approaches to Therapy

Another method for simplifying anti-HIV therapy would be to use an induction/maintenance model, as is done with CMV and some serious fungal infections. Very potent, toxic therapy is given for a short time to limit the viability of the infectious agent, and then less potent, less toxic, easier to administer treatments are sustained for as long as necessary.

SPICE Study: Treatment Response at 32 Weeks

	Arm A	*Arm B*	*Arm C*	*Arm D*
Baseline N	26	26	51	54
HIV RNA change	-1.96 log	-1.77 log	-1.75 log	-1.86 log
CD4 change	+92	+73	+134	+161
%<400	70	55	83	69
%<50	55	50	70	39

(Hoffmann-LaRoche 1998)

These data suggest that the combination of nelfinavir and saquinavir may be potent, but should be administered in combination with two nucleoside analogues.

Indinavir and Nelfinavir. Early studies by Agouron suggested that the combination of nelfinavir and indinavir raised plasma levels of both drugs. This encouraged Agouron to think that the combination could allow patients to use both drugs twice a day rather than the currently-recommended three times per day.

Twenty-one protease-naive patients were enrolled in an open-label study of the safety and activity of nelfinavir and indinavir. Median baseline viral load was 50,500 copies, and median baseline CD4 was 259. The group was almost evenly divided between nucleoside experienced and naive patients.

Participants were assigned to receive indinavir, either at the standard 800 mg TID dose, or at a dose of 1,000 BID, and nelfinavir, either at the standard 750 mg TID dose, or at a dose of 750 mg BID. The nelfinavir BID dose resulted in substantially lower trough levels than did the nelfinavir TID dose, and so investigators raised the dose of nelfinavir in those patients to 1,000 mg BID.

Ten out of twenty-one patients (47%) achieved plasma HIV RNA levels of <400 copies. Six of these patients had plasma HIV RNA levels of <50 copies. No unexpected adverse events were seen. (Havlir 1998)

Ritonavir and Nelfinavir. Two small pilot studies are currently exploring different dosing regimens of this combination. Current doses under consideration are 400 mg ritonavir BID in combination with 1000 mg nelfinavir BID, 400 mg ritonavir BID in combination with 500 mg nelfinavir BID, and 400 mg ritonavir BID in combination with 750 mg nelfinavir BID.

Ritonavir and Indinavir. In early pharmacokinetic data generated by Abbott Laboratories in HIV-negative patients, combining indinavir with ritonavir seems to allow conversion to BID dosing with good pharmacokinetics. The combination seems to eliminate the food effects of indinavir as well. No data are available on the development of resistance using this combination, but as these two drugs have very similar resistance patterns, it could be a problem (Japour 1998).

Ritonavir and Amprenavir. In animals, there is a significant interaction between these drugs, resulting in major elevations of amprenavir plasma concentrations. Abbott claims that Glaxo Wellcome is delaying systematic study of this combination. Joseph Eron of the University of North Carolina recently presented preliminary findings from a small, open-label study of amprenavir in combination with other protease inhibitors. Patients were assigned to take:

Abbott 462: Virologic Response to RTV/SQV at Week 60

	N	*RNA <200*	*CD4*	*HIV RNA*
Arm A	30	80%	+210	-3.0 to 3.5 log
Arm B	27	90%	+150	-3.0 to 3.5 log
Arm C	21	89%	+175	-3.0 to 3.5 log
Arm D	26	89%	+160	3.0 to 3.5 log

(Hoffmann-LaRoche 1998)

Safety and tolerability data extend through week 48:

Abbott 462: Adverse Events in Patients Treated with RTV/SQV

Adverse Event	*Arm A*	*Arm B*	*Arm C*	*Arm D*
Circumoral parasthesia	1	3	1	4
Diarrhea	4	11	5	12
Fatigue	2	3	8	10
Nausea	4	7	4	11
Depression	1	3	1	4
Dizziness	1	0	5	4
Peripheral parasthesia	1	4	1	2

(Hoffmann-LaRoche 1998)

In addition, some mild elevations of LFTs and triglycerides were seen.

Nelfinavir and Saquinavir. Nelfinavir also interacts with saquinavir, raising saquinavir levels by about five-fold. Consequently, two studies of the combination have been performed.

In a small study of fourteen patients treated with 750 mg NFV TID and 800 mg SQV TID, no clinically significant drug-related abnormalities were seen. At month twelve, participants experienced a median decrease in HIV RNA of 2.4 logs, and a median increase in CD4 cells of about 100 cells. Approximately 80% of participants had HIV RNA levels of <500 at month 12.

In Hoffmann-LaRoche's SPICE study, participants were randomly assigned to receive:

A: Saquinavir in combination with two nucleoside analogues
B: Nelfinavir plus two nucleoside analogues
C: Saquinavir, nelfinavir and two nucleoside analogues
D: Saquinavir plus nelfinavir

Mean HIV RNA was about 63,000, and mean CD4 count was approximately 300 cells. Participants were evenly divided between nucleoside-naive and nucleoside experienced patients. Most participants chose AZT/3TC or d4T/3TC as their concomitant nucleoside analogues.

By week 32, 24 patients had crossed over to unblinded treatment with nelfinavir, saquinavir and two nucleoside analogues. The highest rate of crossovers due to virologic failure (n=11) came from arm D, the double-protease arm.

For the HIV-infected patient, it is probably early to convert these three-times-a-day drugs into twice-a-day drugs. More studies are now being conducted that should give us a clearer picture of the long-term efficacy of these simpler regimens. In the meantime, drug company spin should be taken with a big grain of salt.

Double Protease Combinations

Another strategy for improving patient outcomes on protease inhibitors is to increase the potency of therapy. Many different investigators are exploring combinations of two protease inhibitors. Because all currently-available protease inhibitors (as well as current and future non-nucleoside reverse transcriptase inhibitors) are processed through a pathway in the liver known as the cytochrome p450 system, interactions are expected between them that may change blood levels of the drugs. For instance, Norvir brand ritonavir raises levels of most of the other drugs, while Viramune brand nevirapine lowers levels of most of the other drugs. Following are some of the results so far.

Ritonavir and Saquinavir. Due to the poor oral bioavailability of the Invirase hard-gel capsule formulation of saquinavir, researchers attempted to exploit the drug's pharmacokinetic interaction with ritonavir, which raises plasma levels of Invirase dramatically. When several early studies suggested that the combination might be extremely potent, a sort of double-protease mania began to take hold, generating a whole slew of data from small pilot studies.

Although the need for increased plasma saquinavir levels has been addressed by the new Fortovase soft-gel capsule formulation of saquinavir, the ritonavir/saquinavir combination remains the best-studied double-protease combination, and was recently added to the PHS Guidelines for use in combination with nucleoside analogues as initial antiretroviral therapy. The combination is also proposed for use in treating patients who have failed an initial protease-containing regimen. Data in the Fortovase package insert suggest that plasma levels of both the hard- and soft-gel capsule formulations of saquinavir are similar when the drug is used in combination with ritonavir (Hoffmann-LaRoche 1998).

In Abbott 462, 142 protease-inhibitor naive patients were assigned to receive:

A:	400 mg BID RTV	+	400 mg BID SQV	(N=35)
B:	600 mg BID RTV	+	400 mg SQV	(N=36)
C:	400 mg TID RTV	+	400 mg TID SQV	(N=33)
D:	600 mg BID RTV	+	600 mg BID SQV	(N=37)

Median plasma HIV RNA at baseline was 4.6 logs, and baseline median CD4 cell count was about 275.

In a recent presentation, investigators presented sixty-week follow up from this study. Data presented were from an on-study analysis which excluded patients who had dropped out of the study, or who had discontinued study medication: interpretation of RNA data should be tempered by the substantial number of dropouts, particularly in arms C and D. By week 24 of the study, almost half of participants had switched their regimens to the 400 mg/400 mg BID regimen, usually due to toxicity.

Twice Daily Indinavir or Nelfinavir Regimens

In addition, manufacturers of protease inhibitors are looking for ways to simplify the dosing and administration requirements of their drugs. Merck and Agouron have both presented data on studies of their protease inhibitors in twice-a-day regimens. Current recommended dosing of both Crixivan and Viracept is three times a day. The companies, locked in a life-or-death struggle for market share, are determined not to let their rival gain any sort of competitive edge.

Viracept, which has a longer plasma half-life than Crixivan, has a promising pharmacokinetic profile with a dosing schedule of 1250 mg BID (twice daily). Drug levels stayed above the minimum concentration thought necessary for therapeutic effect, and were consistent with drug concentrations using the standard dose of 750 mg TID (three times daily). In a small six-month pilot study of BID dosing in combination with d4T and 3TC, viral load response was good, with fourteen out of fifteen patients taking 1250 mg of Viracept BID achieving plasma HIV RNA levels of less than 500 copies (Senseon 1998). Agouron's press release, distributed at the Fifth Retrovirus Conference in February 1998, also notes that twelve out of twelve patients treated with 1250mg Viracept TID who had HIV RNA levels that were undetectable by the standard assay were also undetectable by the ultrasensitive assay (<50 copies). The discrepancy between the numbers given (14/15 <500 copies versus 12/12 <50 copies) is unexplained. A larger study of Viracept twice daily is ongoing. (Agouron 1998).

For Merck, the situation is a little more complicated. Knowing that its drug's pharmacokinetics were likely to look bad in a BID dosing regimen, the company initiated pilot studies that focused on HIV RNA response rather than on drug levels. Three groups of patients who were naive to protease inhibitors and to 3TC were assigned to take 800 mg Crixivan TID, 1000 mg Crixivan BID or 1200 mg Crixivan BID. All patients were given AZT/3TC. About thirty patients were assigned to each group; participants had average CD4 counts of about 275, and average viral load of about 4.7log HIV RNA copies. After 32 weeks, 50% of patients treated with the standard 800 mg TID dose had viral load counts of less than 500 copies, and 40% had less than 50 copies. For patients treated with the BID doses, about 70% in both regimens had viral load counts of less than 500 copies, and 60% had viral load of less than fifty copies (Martin 1998). Sounds good, no?

However, sneaky Abbott Laboratories, still searching for some reason why patients might take Novir™ brand ritonavir, their notoriously toxic protease inhibitor, compared the pharmacokinetics of Crixivan BID with those of Crixivan BID in combination with Norvir. At the dose being tested by Merck (1200 mg BID), blood levels of drug dropped below what are commonly thought to be minimum necessary levels for several hours during each dosing regimen. In combination with Norvir, however, levels of Crixivan stay well above minimal concentrations during BID dosing, and the effects of food on Crixivan levels are eliminated (Japour 1998).

Now Merck responded to these concerns by noting -- accurately -- that blood levels of Crixivan have not been correlated to antiviral response (although they were very quick to note that low blood levels of Invirase™ brand saquinavir led to poor antiviral efficacy). Merck believes that the demonstrated antiviral potency of Crixivan BID dosing means that blood levels do not matter. If the drug works, who cares if the pharmacokinetics are bad? Furthermore, always reluctant to let a competitor get ahead, they note that 90% of a dose of Viracept™ is bound to plasma proteins in the bloodstream, and is therefore unavailable for inhibiting the HIV protease, while only 60% of a Crixivan dose is plasma protein bound. Consequently, blood levels of drug may not accurately reflect levels of active drug.

group to develop studies of these phenomena, and to define measurements of the syndrome's frequency and severity for use in other studies. Ongoing and currently planned studies include:

* ACTG 892, a superstudy across several antiretroviral protocols, will measure the effect of HAART on weight and lean body mass. This study is intended to rectify the fact that, at present, there are no good prospectively controlled data on the impact of HAART on weight and body composition. The study is currently open, and will enroll approximately 200 patients.

* ACTG 5005-S, a substudy of ACTG 384, which will compare protease inhibitor-based regimens to NNRTI-based regimens in naïve patients. ACTG 5005-S will try to ask if these metabolic and body compositional changes are protease inhibitor-specific, or if they occur with other HAART regimens. Approximately 300 patients will be enrolled, and investigators will follow fasting measurements of glucose and other metabolic indicators, body composition, and measurement of various body parts. Blood samples will also be banked for future use.

* ACTG 392 will evaluate the impact of high-quality protein supplementation on patients with mild to moderate weight loss. An exercise protocol is also planned.

A cardiovascular working group is also attempting to develop definitions of cardiovascular toxicity, including myocardial infarction for use in future ACTG studies. Finally, the group is also planning to test pharmacokinetic interactions between protease inhibitors & cholesterol-lowering drugs, some of which are metabolized by p450cyp3A isoform, and might therefore be expected to interact (Mulligan 1998).

In New York City, Dr. Kotler is working with the Community Research Initiative on AIDS (CRIA) to develop a study of human growth hormone for the treatment of truncal obesity. CRIA also has an ongoing study of glucose intolerance in patients treated with protease inhibitors.

Hopefully, data from these studies will help scientists and doctors to better characterize these side effects, understand their long-term clinical impact, and intervene effectively where necessary, enabling patients to maintain long-term therapy.

SIMPLIFYING HAART REGIMENS

Once Daily Dosing

One possibility for improving the ability of patients to adhere to medication for long periods of time would be the development of simpler regimens. Several new drugs, such as adefovir, efavirenz, PMPA and ABT-378 are being developed for once-daily therapy.

Similiarly, physicians in Frankfurt have released interim results from their study of seventy patients treated with ddI, 3TC and nevirapine, all used once daily. After 20 weeks of treatment, 90% of study participants'' plasma HIV RNA levels have fallen below the limit of quantification (400 copies/ml). Another study at Amsterdam University is comparing three different ddI/d4T-based regimens (the two nucleosides are given with either 3TC, nevirapine, or Crixivan). So far, the Crixivan group has the smallest percentage of patients below the limit of detection. "It's all compliance," says principal investigator Joep Lange (Cox 1998).

patients not taking a protease inhibitor: two reports are on patients treated with AZT, and one report is on a patient treated with d4T. Eight abstracts presented at the Fifth Conference on Retroviruses and Opportunistic Infections described symptoms thought to be associated with the syndrome.

In one study conducted at St. Vincent's Hospital in Sydney, Australia, researchers found that approximately sixty-five percent of patients treated with protease inhibitors developed symptoms of lipodystrophy after a mean of ten months. However, another study suggested an attack rate of only about five percent. Differences probably have to do with differing definitions of lipodystrophy. The Australian group performed molecular analysis to look for a reason why protease inhibitors might cause such metabolic changes. The researchers found that an amino acid sequence in the catalytic site of HIV protease was very similar to a low-density lipoprotein receptor-like protein (Carr 1998).

In a recent article on the syndrome, leading AIDS gastroenterologist Donald Kotler wrote:

> A major weakness in this hypothesis, that the changes represent a toxic, metabolic effect caused by protease inhibitors, is that some patients with the syndrome are not taking protease inhibitors. Investigators from San Francisco reported that the development of a Buffalo hump was not limited to patients receiving protease inhibitor therapy. In addition, we have observed several patients in New York with the "syndrome" who were not taking protease inhibitors, and their plasma viral burdens were low. We have been evaluating the ability of anthropometric measurements to detect the alterations in fat distribution, since data on a large number of HIV-positive control subjects studied since 1984 are available. Based on these data, there is preliminary evidence that altered fat distribution was a common phenomenon in HIV-infected subjects studied in the past, prior to the availability of protease inhibitors. Even among this study group, a few patients were seen who were identical to those being described as having the "syndrome."... It is therefore possible that both mechanisms are operative (Kotler 1998).

Dr. Kotler also observes that the lipodystrophy syndrome currently being described in HIV-negative patients is disturbingly similar to "Syndrome X, a collection of symptoms including truncal obesity and metabolic abnormalities that has been described in HIV-negative patients. Such similarity is "worrisome," Dr. Kotler writes, "since that syndrome is associated with accelerated cardiovascular disease."

Recently, in a letter to the *Lancet*, specialists from Minneapolis/St. Paul identified two patients who developed unusual coronary artery disease in young men being treated with protease inhibitors (a third case has since been reported). In a subsequent chart review of 124 patients receiving protease inhibitor treatment, 33% had lipid concentrations in the plasma that were high enough to suggest therapy with diet and exercise or with lipid-lowering drugs (Henry 1998).

At present, no therapy exists for the lipodystrophy syndrome, although some investigators are beginning to experiment with anabolic steroids and Human Growth Hormone. Medical treatment as indicated is appropriate for diabetes, hypertension, and significant hyperlipidemia. So far, clinically significant problems seem to be rare, however systematic study of the syndrome is really just beginning.

The Complications of HIV Research Agenda Committee of the ACTG has formed a metabolics focus

summarizing post-approval adverse event reporting, the Food and Drug Administration (FDA) has divided the syndrome into two categories: Fat Redistribution Syndrome/Lipodystrophy and Hyperglycemia/Diabetes. As of March 18th, 1998, the FDA had received a total of 62 reports of fat redistribution in more than 64 patients treated with protease inhibitors:

Reports of Fat Redistribution in People Treated with Protease Inhibitors

Drug	*Number of Reports*
Indinavir	45*
Ritonavir	10
Saquinavir	8
Ritonavir + Saquinavir	1

*One of the patients receiving indinavir was switched to nelfinavir with no improvement.

(Murray 1997)

Symptoms reported included both buffalo hump and Crix belly, alone and together. There were reports in both men and women that included gynecomastia or breast enlargement. Cushing's syndrome was ruled out in those patients who had a laboratory workup. The syndrome appeared in patients of all ages. Onset of the syndrome was noted an average of 5.6 months after initiation of therapy, with a range of five days to eleven months. FDA also has reports on 230 cases of hyperglycemia/diabetes in people treated with protease inhibitors, 26 of which are from foreign sources.

Reports of Hyperglycemia/Diabetes in People Treated with Protease Inhibitors

Drug	*Number of Reports*
Indinavir	141
Saquinavir	31
Ritonavir	23
Ritonavir+Saquinavir	24
Nelfinavir	15

(Murray 1997)

Forty reports described a loss of glucose control in patients with pre-existing diabetes, and thirty additional cases had a positive family history of diabetes. New onset diabetes was described in 94 patients, and diabetic ketoacidosis (DKA) in 24 patients. Among the 24 DKA patients, only three were documented as having preexisting diabetes. Ritonavir in combination with saquinavir represented approximately 10% of all hyperglycemia and/or diabetes cases, and was being used by 38% of patients who developed DKA. Onset of the syndrome was noted an average of 101 days after initiation of therapy, with a range of two to 390 days. The protease inhibitor was not necessarily discontinued for hyperglycemia; 104 patients were treated with insulin and 46 patients with oral hyperglycemic agents. Of the sixty-five patients who discontinued treatment, thirty-four reported improvement or recover, and in one case, a positive re-challenge was reported.

In addition, the FDA database contains two reports of symptoms associated with the syndrome in

triple-combination therapy with AZT, ddI and nevirapine in treatment-naïve subjects with moderately advanced HIV disease, patients who were able to successfully follow the triple-combination regimen for one year were much more likely to have undetectable plasma HIV RNA levels than patients who missed a total of 28 or more days of at least one drug during the course of the study. (Hall 1997)

Furthermore, we know from data developed by the federal government's AIDS Clinical Trials Group (ACTG) in a survey of 76 patients on combination therapies that missed doses of HIV drugs are frequent:

ACTG Survey:	Frequency of Missed Doses
Yesterday	11%
Day Before Yesterday	14%
Last Two Days (combined)	18%
Last Two Weeks	36%

(Chesney 1997)

Looking at a broader period of time, at least a third of patients missed at least one dose in the past two weeks. Due to suspected underreporting, investigators estimate that half of patients may be missing doses in a two-week time period. Patients were also asked why they missed doses of anti-HIV therapy:

ACTG Patient Survey: Self-Reported Reasons for Missing Doses	
Simply forgot	40%
Slept through the dose	37%
Away from home	34%
Change in routine	27%
Busy with other things	22%
Too sick	13%
Sick from side effects	10%
Depressed	9%

(Chesney 1997)

Clearly adherence will play a major role in clinical response to anti-HIV therapy.

LIPODYSTROPHY, HYPERGLYCEMIA & DIABETES

Furthermore, as patients are successfully treated with potent antiretroviral therapy for longer periods of times, new and puzzling side effects have emerged, about which neither the best clinical management, nor the long-term impact are yet understood. Most obviously, a troubling syndrome involving metabolic abnormalities and body composition changes has been noted in HIV-infected persons. The widely reported symptoms have included "Crix belly" (or "protease paunch" as the Merck public relations staff prefers to call it), "buffalo hump," peripheral wasting of the arms and legs, hyperglycemia and diabetes. The syndrome may also include mild elevations in serum cholesterol concentrations, hypertension, hypogonadism, subclinical insulin resistance, thinning of the skin on the arms and legs, increased wrinkling of the face, gynecomastia in men, and in women, enlargement of the breasts and narrowing of the hips (Murray 1997). The condition is said to resemble Cushing's Syndrome, a disorder of the adrenal gland that can cause central body obesity, glucose intolerance and hypertension. However, Cushing's Syndrome is characterized by overproduction of the powerful steroid hormone cortisol. In the syndrome seen in HIV-positive patients, cortisol levels in the bloodstream are normal. In

STATUS OF THE ERADICATION HYPOTHESIS

In July, 1996, at the Vancouver Conference, David Ho of New York's Aaron Diamond AIDS Research Center (ADARC) hypothesized that, with anti-HIV drugs that completely suppressed the replication of HIV throughout the body, the virus might be cleared from the body in two to three years, depending on the number of HIV-infected cells. (Harrington 97)

The successful implementation of this hypothesis using currently available therapies depended on a number of key assumptions:

First, Ho's estimate presupposed that there were only two cellular compartments harboring HIV: activated, HIV-infected CD4 cells, and latently infected CD4 cells or macrophages. By studying the decline in plasma virus levels after initiation of potent antiretroviral therapy, Ho estimated that the half-life of the first compartment was about 1.4 days and included more than 90% of the new virus population, while the second compartment had a half-life of about one to four weeks, and was producing less than ten percent of the new virus population.

Second, this estimate presumed that viral clearance in tissue compartments, such as the brain, lymph nodes, genital tract and gut, mirrored that in the plasma.

Finally, the estimate presumed that apparently complete suppression of HIV replication was in fact complete suppression of HIV replication. The most sensitive tests of plasma viral load currently available are unable to detect fewer than 20 copies of HIV RNA per milliliter of blood. If, as had been hypothesized, only one replicating virus could refuel the body with resistant virus, then the difference between 20 copies of virus and no copies per milliliter could be significant.

Based on this hypothesis, several studies were designed to explore therapeutic cessation in patients who have had undetectable plasma HIV RNA levels for a long time.

However, in late 1997, several studies found that there is a third cellular compartment harboring infectious HIV (Finzi 1997, Wong 1997, Chun 1997). This compartment consists of latently infected, resting, but previously activated memory CD4 cells with integrated HIV DNA in their nucleus. After two years of potent antiretroviral therapy and undetectable plasma HIV RNA levels, levels of HIV in this compartment still do not appear to have measurably declined.

Researchers are now working to find ways to stimulate these cells into activity, allowing HIV to destroy the cells without infecting new cells. However, until these efforts are successful, researchers warn, cessation of therapy would lead to complete re-seeding of the body with HIV: at present, anti-HIV therapy must be continued for the life of the patient.

THE CHALLENGE OF ADHERENCE

Lifetime therapy presents significant difficulties for patient adherence, in that current optimal therapeutic are complex and are associated with significant toxicity in the short term. These challenges to patient adherence are thought to be clinically significant: in the INCAS study, which looked at the effects of

IVA. NUCLEOSIDE & NUCLEOTIDE ANALOGUES

ii. Abacavir / Ziagen™ (Glaxo Wellcome)

by Theo Smart

Ziagen brand abacavir (formerly known as 1592U89 and commonly referred to as '1592') is a lipophilic carbocyclic guanosine analogue with good oral absorption and central nervous system (CNS) penetration. Limited preliminary data suggest it also may be the most potent nucleoside analogue antiretroviral yet tested in treatment-naive patients. The dramatic reductions in viral load (in the range of 1.4-1.8 logs below baseline) reported in one twelve-week dose-ranging study have led many activists to call for rapid development of and compassionate use access to the compound. The slow development of the drug stems from a number of factors, including Glaxo Wellcome's indecision over how it will market the drug, supply problems, and confusion about how to design of clinical studies of a new agent in the era or highly active antiretroviral therapy and utilization of viral load in the clinical management of people with HIV.

BACKGROUND

Several years ago, Glaxo used to own the rights to carbovir, a chemical antecedent to abacavir, but dropped the compound reportedly because of toxicity observed in animal studies. At this point, chemists from the old Burroughs Wellcome (BW) made slight structural modifications to carbovir that altered its toxicity profile, and took the new drug into clinic in late 1996. Shortly afterward, Glaxo and Wellcome (GW) merged, and the future of the compound plus two other nucleoside analogues under study by BW was in jeopardy, as the merged company already owned AZT and 3TC, a combination which would soon dominate the market. Some corporate decision-makers may have felt that there would be little advantage to GW in marketing yet another nucleoside analogue.

Indeed, GW wasted little time in dropping one of the old BW drugs, a cytosine analogue very similar to but reportedly more potent than 3TC, and then axed 935, a novel uridine analogue, when its *in vitro* antiviral activity -- roughly equivalent to AZT's -- was deemed disappointing. Abacavir was next on the chopping block, but the company found itself in a bind when the early data on abacavir far exceeded anyone's expectations -- the drug's potency seemed similar to that of a protease inhibitor.

What could the company do? It would have been unwise to sell the drug because in a competitor's hands it might cut into sales of AZT / 3TC. So GW instead chose to develop the compound slowly, emphasizing the clinical evaluation of abacavir in combination with its own drugs, AZT, 3TC and amprenavir. This was despite *in vitro* data suggestive of some cross resistance between abacavir and 3TC.

STRUCTURE & MECHANISM OF ACTION

Abacavir is a lipophilic carbocylic 2',3'-ene nucleoside analogue, activated intracellularly to a triphosphate (TP) carbocyclic guanine analogue, that acts as a reverse transciptase inhibitor. Guanine is one of the nucleotide bases of DNA. By acting as a defective decoy of this nucleotide, abacavir-TP is inserted by the HIV reverse transcriptase enzyme into the growing chain of HIV proviral DNA. No additional nucleotides can be placed next to this defective guanine which disrupts viral DNA chain synthesis, aborting infection of the cell.

ANTIRETROVIRAL POTENCY

In *in vitro* studies. Abacavir inhibits HIV clinical isolates in peripheral blood lymphocyte cultures with an average IC_{50} of 0.26 μM. This *in vitro* potency is similar to AZT's, which partly accounts for the surprise with which the clinical results were met. The increased *in vivo* potency is now explained by unusually efficient intracellular absorption and activation of the drug.

In vitro, abacavir is synergistic with the thymidine analogue AZT, the non-nucleoside reverse transcriptase inhibitor nevirapine, and Glaxo Wellcome's protease inhibitor amprenavir.

CLINICAL DATA

Data presented at Vancouver and at the Drug Therapy for HIV Infection Conference in Birmingham, England, established that abacavir has unprecedented potency. Those data were drawn from study 2001, in which patients were treated with 200 mg TID, 300 mg BID, 400 mg TID or 600 mg TID of abacavir (twenty patients per arm). After four weeks of monotherapy, viral load fell by 1.11-1.77 logs (from a baseline range of 4.5-5.1 logs), and CD4 cell counts increased by 63 to 83 cells (from a baseline of 356 to 396 cells). (Saag 96)

Patients then were randomized to continue monotherapy or to add AZT. At 12 weeks, viral load reductions ranged between 1.02-2.24 logs, and CD4 count increases were between 90-145 cells. There was no significant difference between the abacavir monotherapy and abacavir/AZT combination arm in absolute reduction of viral load, but a higher percentage of patients on combination therapy (60% versus 20%) had viral load reductions that fell below the limit of detection (200 copies).

In combination with amprenavir: In one arm of a dose-ranging study of GW's protease inhibitor amprenavir (formerly known as 141W94), nine patients received abacavir, 300 mg BID in combination with amprenavir 900 md BID. Two patients dropped out due to adverse events: one because of rash and dysarthria (difficultly speaking) and one because of nausea. After four weeks of therapy, the median decrease in viral load experienced by the remaining patients was 2.08 log (from a baseline of 4.19 log). CD4 counts increased by a median of 79 cells over a baseline of 223. Five of seven (71%) patients achieved viral load reductions that fell below 400 copies (Schooley 1997).

ONGOING STUDIES

In Protocol 2001, an early phase I study of abacavir, a small group of patients has now been treated fro up to 48 weeks. Patients may be taking abacavir either in combination with other nucleosides, or in combination with a protease inhibitor. After 24 weeks of treatment, 15/18 patients using abacavir in combination with nucleoside analogues had undetectable plasma HIV RNA (<400 copies), and 9/10 patients taking abacavir in combination with a protease inhibitor had less than 400 HIV RNA copies (Torres 1998).

Protocol 2002 is a 24-week dose-evaluation study currently underway in Europe comparing 100, 300 and 600 mg BID. The study has enrolled sixty antiretroviral naive patients with CD4 cell counts> 100, and viral loads above or equal to 30,000 copies. Subjects may elect to discontinue the blinded portion of the study and continue on AZT/ 3TC with abacavir if they reach one of a set of pre-defined criteria based on CD4 cell count, viral load or disease progression. Preliminary analysis of data from this study (out to week 4) and data from 2002 suggested that doses above 300 mg are roughly equivalent. Recently presented data from the Fifth Retrovirus Conference show a median reduction in plasma HIV RNA levels of about 2.5 log copies over the course of about 28 weeks. About seventy percent of these participants have plasma HIV RNA levels below 400 at week 28, and about 40% have plasma HIV RNA levels below 50 (Staszewski 1998).

Protocol 2003 is being conducted in patients with extensive prior nucleoside analogue therapy, CD4 cell counts above 100 and loads above 10,000 copies. The 40 patients enrolled have one of four treatment histories: 1) at least 6 months prior d4T monotherapy; 2) six or more months of ddI with or without AZT; 3) twelve or more months or prior AZT monotherapy; or 4) at least 12 months of AZT/ 3TC combination therapy. In recent analysis, the average viral load reduction from baseline at week four was 1.11 log copies, and the average reduction at week 22 was 1.30 log. For patients who did not have the 3TC M184V mutation at baseline, the reduction in viral load was 1.49 log copies at week four, and 1.48 log copies at week 22. For participants who did have the m184V mutation at baseline, some of whom also had additional mutations, the average viral load reduction was 0.52 log copies at week four, and 0.97 log copies at week 22. (Levin 1998)

Another study, which looks at the combination of abacavir with amprenavir, is presented below in the chapter on amprenavir. (Bart 1998)

RESISTANCE & CROSS RESISTANCE

Dr. Richard Harrigan from Glaxo Wellcome reported on the resistance profile of abacavir at the Fourth Conference on Retroviruses and Opportunistic Infections. *In vitro*, after four serial passages in the presence of increasing concentrations of drug, a mutation arises at the 184 codon on the reverse transcriptase enzyme. Dr. Harrigan noted that this is "the same mutation that causes high level resistance to 3TC, but interestingly, it only confers a marginal decrease in 1592 susceptibility." After several more passages, mutations at positions 65, associated with ddC resistance; 74 associated with ddI resistance; and/or 115 occur, which together with the 184 mutation confers a ten to twelve-fold decrease in susceptibility to abacavir. Such isolates remain susceptible

to AZT and d4T, however, and are only five-fold less susceptible to ddI and ddC. Any virus containing the 184 mutation is resistant to 3TC however (Harrigan 1997).

The resistance profile may be slightly different in humans, particularly when the drug is used in combination with other antivirals. In fact, when the virus was passaged in vitro in the presence of both abacavir and AZT, only the mutation at position 65 was observed. (This combinatorial effect on resistance did not extend to the combination of abacavir and 3TC, however, as the same mutations occurred when abacavir was sequenced alone.)

In study 2001, after twelve weeks nearly 60% of viral isolates that could be assessed from patients on abacavir monotherapy had some mutation or combination of mutations at positions 65, 74 or 184 compared to roughly 13% in the virus taken from patients treated with AZT/ abacavir. But perhaps the most promising observation from the study was that one patient who had the 184 mutation at baseline, experienced a one log reduction in viral load on abacavir, and an additional log reduction when AZT was added. This suggests that abacavir may be active in some patients who have become resistant to 3TC.

This single patient anecdote may be misleading, however. Randall Lanier has presented an analysis of the resistance data from study 2003, where abacavir was added to current failing nucleoside analogue therapy. (Lanier 1998) The data do not appear to be as negative as that reported by David Hardy, which suggested that abacavir had no activity at all in individuals with prior AZT experience. The picture that is evolving is more complex, however, GW is very craftily spinning the data to place abacavir in the best possible light.

Lanier correlated baseline genotypic and phenotypic data with subsequent response on abacavir. Response data have now been reported for both weeks four and twenty four. It is important to recognize that there can be an initial response to therapy that cannot be sustained because of the presence of viral isolates highly resistant to a drug that might be minor strains in one's overall viral population, and thus take slightly longer to grow out. An example of this might be ACTG 333, where patients on long term saquinavir had only muted responses to indinavir, despite the fact that indinavir-resistant viral isolates could not be detected at baseline. Lanier reported that with an increasing number of mutations in the reverse transcriptase enzyme, there was a decreasing abacavir-associated antiviral effect. One mutation by itself did not appear to decrease abacavir activity by much; two mutations decreased the activity by about half a log, three by 1 log, and so on until 5 or more mutations or so completely wiped out the drug's antiviral effect. The problem with this analysis is that it treats all mutations or combinations of mutations equally, and this is probably not the case.

Likewise, there was a decreasing antiviral effect with an increase in phenotypic resistance to abacavir at baseline. Two- to eightfold phenotypic resistance could reduce response at week 4 by almost a log. Eightfold resistance negated abacavir's effect. Again, the problem with such assays are that the predominant viral strain in circulation may be mostly sensitive, but highly resistant strains nonetheless may be present, and simply waiting to be selected.

The notion that GW is trying hardest to fight is that AZT/3TC pretreatment will make a person fail on abacavir. Data do suggest that simply having the 184 mutation does not wipe out abacavir, although it is interesting to note that the median loss of sensitivity to virus from patients in this study was ~4.4-fold, which is more than the 2-fold suggested by earlier in vitro data. The range was from between 2-fold and more than 7-fold. Meanwhile, dual resistant virus was 6.7 fold less sensitive to abacavir. Five out of 13 were fully (more than 8-fold) resistant.

Some have made much of the fact the eight of these AZT/3TC resistant patients were still sensitive to abacavir. Nevertheless, they were not very sensitive to it (5.5-6-fold resistant) and it seems unlikely that there is a significant clinical difference here.

Finally, one must come back to the clinical data — there was little or no sustained clinical response in these patients. What seems clear is that patients who have failed on AZT/3TC cannot rely on abacavir to anchor a HAART regimen.

As this study includes patients with a history of prior AZT, AZT/3TC, d4T, ddI (with or without AZT) treatment, final results may show whether pretreatment with any particular regimen prejudices against benefitting from abacavir.

The dreaded 151 mutation. A recently reported mutation in the reverse transcriptase enzyme at codon 151 that reportedly causes resistance to all marketed nucleoside analogues, apparently defeats abacavir as well. Thus, abacavir therapy will be of no avail in pretreated patients with this mutation. How often the 151 mutation might occur in people taking abacavir is unknown.

The 184 mutation. That abacavir and 3TC share the 184 mutation should be a cause of concern, particularly to Glaxo Wellcome. Although It is possible that the use of abacavir in combination with AZT/ 3TC may only serve to speed the development of resistance to 3TC, and loss of both drugs' antiretroviral activity, no evidence of such an effect was seen in Study 2002, which patients have been treated for more than 22 weeks with a combination of AZT, 3TC and abacavir. In the patients pretreated with 3TC, the 184 mutation, particularly when combined with other mutations, may render abacavir useless. And if abacavir commonly causes the 184 mutation when used as a first-line treatment, it will render subsequent 3TC therapy useless. GW may find itself marketing a drug that competes with 3TC, anyway, and will not increase the company's market share. This again helps to explain the slow development of the compound.

ADVERSE EVENTS

Animal and cell culture toxicology. The toxicity of abacavir in animal studies has been characterized by Dr. Steve Lafon of GW as "unremarkable." In laboratory studies, abacavir was relatively non-toxic to human bone marrow progenitor cells (BFU-E and CFU-GM cells; ICSO 110μM) and to human leukemic and liver tumor cell lines.

Human toxicology. Despite the positive press, abacavir treatment was associated with significant side effects in study 2001. These included nausea, headache, asthenia, diarrhea, insomnia, dizziness, vomiting, abdominal pain, rash and other conditions. Eleven of 80 patients experienced adverse events and laboratory abnormalities which were treatment-limiting or led to treatment discontinuation. These adverse events included suspected acute allergic reactions in two patients (symptoms included fever and rash, and in one subject, parasthesias); rash in one participant, dizziness, palpitations and photophobia in one patient; and nausea and/or vomiting in three subjects (one of whom also complained of fatigue). Laboratory abnormalities included one case of neutropenia, grade 3/4 ALT in one subject, decreased platelets in one patient and hypoglycemia in another. (Saag 96)

Potentially life-threatening hypersensitivity reaction. Recently, Glaxo-Wellcome distributed a letter to investigators using abacavir, warning that two to three percent of patients treated with abacavir had developed a hypersensitivity reaction, which, in several cases had become severe, including anaphylctic shock and one death. Early symptoms may include nausea (with or without vomiting), malaise and possibly an accompanying rash. Most serious cases occurred after patients had developed a reaction, been withdrawn from treatment, and re-challenged. The company warns that patients with hypersensitivity response that is severe enough to warrant discontinuation of therapy should be considered intolerant to abacavir (Joyner 1998). In the European study 2002, the safety profile has been reportedly similar to that seen in 2001. The most common adverse events have been nausea, headache, asthenia, diarrhea, and abdominal pain. Staszewski 1998)

PHARMACOKINETICS, FOOD & DRUG INTERACTIONS

In a placebo controlled phase I dose-escalation study (Protocol 131001), twelve out of eighteen patients were randomized to receive five escalating doses of abacavir (100, 300, 600, 900 and 1200 mgs) separated by a washout period of at least one week. The average T_{max} was 1.0-1.7 hours and the C_{max} was ranged between from 0.6-9.6 g. The intracellular halflife of the drug's active metabolite is presently unknown. Food decreases the abacavir AUC by 5% and the C_{max} by 35% (McDowell 95).

Central nervous system penetration. Abacavir was designed to enter the central nervous system. In animals, its CNS penetration is comparable to AZT's. In HIV-infected adults, the average CSF:plasma ratio was 0.19, one- hour post-dose. (Ravitch 1998)

Drug interactions. Unlike the protease inhibitors, abacavir is not metabolized by the cytochrome p450 liver enzyme system. It is therefore less likely to have significant interactions with other drugs, save, perhaps other nucleoside analogues, as the drug may effect the intracellular triphosphorylation of other nucleosides. (Ravitch 1998)

NEW & PLANNED STUDIES

A number of abacavir studies started recently, and a few others are slated to begin this month or in August. GW is still putting the final touches on several of the protocols. In particular, the total number of patients to be

enrolled, site selection and precise definition of virologic progression remain to be decided. In most of the studies, patients will have the option of going on open-label treatments if they meet this protocol defined criteria of progression after sixteen week on study. In some studies, this has been defined as two consecutive detectable viral loads (200 or 400 copies); other options under discussion have been consecutive viral loads over 5000 copies.

Antiretroviral-Naive Studies

Abacavir/Protease combination trial. Abacavir's potency and its CNS penetration provide clear rationale to evaluate it in combination with protease inhibitors. Study 2004 is a 48 week open label study in treatment-naive patients of abacavir plus the five major protease inhibitors: indinavir, ritonavir, saquinavir, nelfinavir and amprenavir. There will be sixteen patients randomized to each arm of the study at eight different sites in the US. Participants must have at least 100 CD4 cells and a viral load of above 5,000 copies to enroll. Patients who progress virologically, as defined by the protocol (see below), prior to week sixteen may continue on randomization or discontinue the study. Patients who meet failure criteria after this point have the option of continuing abacavir with any approved antiretroviral regimen of their choice in addition to the options of quitting or remaining in randomization.

The 'three is better than two study': AZT/3TC versus AZT/3TC/abacavir. Study 3003 is an international study in antiretroviral therapy-naive patients with at least 100 CD4 cells. Patients were randomized to receive AZT/3TC or the incestous combination of Glaxo's three nucleosides for forty-eight weeks in a blinded fashion. Patients who meet protocol defined switch criteria after sixteen weeks will have the option of continuing blinded in the trial (not bloody likely), quitting the study, or receiving abacavir in combination with any approved antiretroviral therapy.

Given that AZT/3TC/indinavir has proven superiority to AZT/3TC, and that resistance to AZT/3TC develops rapidly, the inclusion of the double-nucleoside arm would appear to be unethical, especially given the possibility that resistance-associated failure on abacavir could render subsequent therapy with most other nucleoside analogues useless.

Study 3005, the indinavir/combivir/abacavir trial. One of the more useful GW studies will try to show the equivalence of at least 48 weeks of indinavir plus combivir (the new combination AZT/3TC tablet) to the combivir/abacavir combination. To qualify patients must be over 16 years old, antiretroviral naive, with more than 100 CD4 cells and more than 10,000 copies of HIV RNA . Patients who meet the protocol defined criteria, (two consecutive HIV RNA counts of over 400 copies) after week sixteen will again be allowed to either drop out of the study, continue on blinded randomization, or receive open label study drugs plus any other currently licensed therapy.

Antiretroviral-Experienced Studies

The AIDS dementia complex trial. Study 3001 is a phase III international trial of abacavir in combination with antiretroviral therapy (stable for at least eight weeks prior to study entry). Subjects may not change or add to their background antiretroviral therapy during the randomized phase of the study. Patients will be randomly assigned to receive either abacavir (600 mg BID) or placebo for twelve weeks. CSF sampling is required at screening. Additional CSF sampling will be performed on consenting patients. Neurological, neuropsychological assessments as well as patient and caregiver questionaires will be collected for all participants. At the end of the 12 week randomization phase (or at the time of ADC progression, or after six weeks on study if the patient experiences severe drug toxicity related to background therapy and not abacavir) patients may continue open-label abacavir.

ACTG 320 rollover study: ACTG 368. This trial includes patients originally randomized to AZT/3TC on ACTG 320 or anyone with a CD4 cell count below 200 cells and more than three months prior therapy with AZT (or d4T) plus 3TC. All patients must be receiving AZT/3TC at the time of study entry and be protease and NNRTI-naive. Patients with over 50 CD4 cells will be randomized and followed for at least forty-eight weeks on:

* indinavir (TID or BID) + efavirenz, or
* indinavir (TID or BID) + efavirenz + abacavir

Patients with CD4 cell counts below 50 will be randomized only to either of the two indinavir TID regimens. Patients with confirmed detectable plasma HIV RNA (two consecutive viral loads above or equal 200 copies) will be offered open label indinavir/efavirenz/ abacavir.

The European antiretroviral-experienced study 3002. In this European study, patients with more than 12 weeks, and less than 18 months of prior antiretroviral therapy will be randomized to standard of care therapy with or without abacavir.

Pediatric studies. Study 3006 is a study in treatment-experienced children less than 13 years old and with less than 100,000 copies. Patients will be randomized to receive abacavir or placebo plus AZT/3TC for forty-eight weeks. Patients who meet a protocol defined switch criteria at 8 weeks or thereafter will have the option or receiving abacavir in addition with any approved antiretroviral therapy. Neurodevelopment assessments will be collected on all patients.

ACTG 321 is single and multiple dosing pharmacokinetic dose-escalation study in HIV-infected infants. All patients will receive standard postnatal AZT therapy in addition to single or multiple doses of abacavir. Results from the single dosing phase will be used in the multiple dosing part of the study.

To qualify, infants must be 0-72 hours old or 21-28 days old depending on which part of the study they enter. The treatment duration for patients in the multiple dosing phase of the study will be six weeks.

Compassionate use & expanded access. The abacavir expanded access program is open to patients over the age of 13 who are failing or intolerant to standard therapy and who, in the judgement of their physician, are unable to construct a viable treatment regimen without abacavir. There are no CD4 or viral load entry criteria. Separate programs are ongoing for children and people with dementia. As the drug is expected to be approved and reach market in only a few months it is clear that this expanded access is merely a premarketing program. For information, patients or physicians can call 1.800.501.4672.

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IVA. NUCLEOSIDE & NUCLEOTIDE ANALOGUES

iii. Adefovir dipivoxil / Preveon™ (Gilead Sciences)

by Theo Smart

BACKGROUND

Adefovir dipivoxil (Preveon™) is a nucleotide analogue with *in vitro* activity against HIV, HBV, HSV-1 and -2, EBV, HHV-6 and CMV. Despite the drug's broad spectrum activity, it has fallen through the cracks, overlooked by most activists and clinicians. The half log (70%) reduction in viral load that this drug achieved in studies of mostly antiretroviral-experienced people last year was overshadowed by the potency of abacavir, the protease inhibitors and non-nucleoside reverse transcriptase inhibitors (NNRTIs). Adefovir's potency also has been overshadowed by another Gilead drug further back in the pipeline, PMPA. The newer agent does not share adefovir's broad spectrum activity, but its anti-HIV potency is impressive. Well-publicized reports that dramatic potency — for example a single dose of PMPA blocked SIV infection in primates when given up to 24 hours after exposure, a feat not matched by any other agent — have led many activists to call for Gilead, a small company with limited resources, to drop the adefovir in favor of rapid development of PMPA. However, the oral formulation of PMPA has only recently entered phase I/II studies.

Meanwhile, adefovir is now in phase III pivotal studies for HIV, as a CMV prophylaxis, and in phase II for HBV. Though not the most potent of agents, aside from abacavir and perhaps 3TC, the drug appears to be as potent as any of the nucleoside analogues in the treatment-experienced population. Few would dispute that agents such as AZT, d4T and ddI still have a role in combination regimens. And adefovir may be particularly useful in treatment experienced patients, since it has a unique resistance profile.

Structure and Mechanism of Action. PMEA (9-[2phosphonomethoxy)ethyl]adenine) is a nucleotide analogue, which differs from a nucleoside analogue by virtue of a phosphate — the drug is one step closer to activatation when it enters a cell. When fully phosphorylated, PMEA is an inhibitor of viral polymerases (and HIV's reverse transcriptases) with submicromolar K_i values versus HIV reverse transcriptase (K_i 0.01μM), HCMV, HSV, and HBV polymerase. The fully phosphorylated nucleotide competes with adenosine, one of the building blocks of DNA, for incorporation into DNA. In the case of HIV, once added to the growing chain of HIV proviral DNA, no additional nucleotides can be placed next to this defective adenosine, which disrupts viral DNA chain synthesis, and aborts infection of the cell.

Adefovir or bis(POM) PMEA (bis(pivaloyloxymethyl)-9-[2-(phosphonomethoxy)-ethyl]adenine) is actually the prodrug of PMEA with improved oral bioavailability, tolerability and antiviral activity (by virtue of improved intracellular metabolism).

ANTIRETROVIRAL POTENCY

In vitro studies. Adefovir potently inhibits HIV in a number of cell lines including monocytes and macrophages. The IC_{50} values are listed below.

***In vitro* Potency of PMEA, Adefovir & AZT: IC_{50} (μM), HIV-1**

Cell type	*PMEA*	*Adefovir*	*AZT*	*Reference*
MT-2	16	0.5	0.1	*Srinivas*
Lymphocyte C8166	3.5	--	--	*Perno*
M/M	0.025	--	--	*Perno*
PBMC				
activated	0.4	--	0.008	*Shirisaka*
resting	0.023	--	17.5	*Shirisaka*

In vitro, adefovir is synergistic with d4T, ddC, AZT, nelfinavir, ritonavir, and saquinavir.

CLINICAL DATA

Activity data have been reported from several studies of adefovir.

Study 402 was a double-blind placebo-controlled study that enrolled 36 patients with CD4 cell counts above 100, p24 Ag > 50 pg/ml. Concurrent antiretroviral therapy was forbidden. Participants were randomized to take adefovir 125, 250, 500 mg or placebo for fourteen days (9 active/3 placebo per dose group). Results are shown below.

Antiretroviral Potency of Adefovir: Median HIV RNA Change

		Median log		*Median log change from baseline*
Dose Group	*N*	*Baseline*	*N*	*Week 2*
Placebo	8	5.2	8	-0.1
125 mg	9	4.6	9	-0.4
250 mg	8	4.9	8	-0.6
500 mg	9	4.8	9	-0.6
All active	26	4.8	26	-0.5

(Deeks 1996)

p=0.03, comparison of dose groups

There was no significant difference between the antiviral activity of dose groups, although further study of the 500 mg dose was discontinued because of a higher rate of adverse events (see below).

Gilead conducted a further study to determine whether there was a difference between the doses over time. Study 403 randomized seventy patients (75% nucleoside experienced) with CD4 cell counts over 200 to twelve weeks on adefovir monotherapy 125 and 250 mg or placebo. There was no difference in viral load changes between the two doses (median reduction 0.5 log) during the initial phase of the study. There was a median increase of 45 CD4 cells over the twelve week period.

After four weeks of washout, patients were allowed to restart adefovir in combination with concomitant antiretrovirals for six months. Sixteen patients who continued adefovir as a monotherapy had a 0.6 log reduction at week 24 of the maintenance phase in viral load from their new baseline, as did eleven patients who took adefovir with other antiretrovirals. It is unclear why there was no additional increase in viral load among the patients on other drugs, although it is possible that they may have initiated the other agents during the washout period. (Deeks 96) Also, adefovir does not have synergistic activity with all other nucleoside analogues, such as 3TC, and coadministration may even be slightly antagonistic.

Study 408: Pivotal surrogate marker trial. Study 408 was a 400-patient study of adefovir versus placebo, both in combination with continued, stable, antiretroviral therapy, in 400 patients with CD4 cell counts between 200-500, and viral load over 2,500 copies. Participants randomized to adefovir 120 mg daily in combination with standard anti-HIV therapy experienced on average a 0.39log10 drop after 24 weeks of treatment, as compared to patients taking placebo in combination with continued stable antiretroviral therapy for 24 week, who experienced on average no change in viral load. CD4+ cell increases in both groups were negligible (Gilden 1998).

RESISTANCE & CROSS RESISTANCE

As noted above, adefovir's effect as a monotherapy appears to be fairly durable with antiviral activity sustained out to a least nine months in patients who have been in long-term follow-up for that long. Most importantly for people who have exhausted most of the other antiretrovirals options, viral isolates resistant to almost all of the other nucleoside analogues and NNRTIs remain susceptible to adefovir.

Gilead has conducted a number of *in vitro* studies to predict what mutations might be associated with decreasing susceptibility to adefovir. In one experiment, the virus (HIV_{IIIb}) was grown in MT2 cell cultures in the presence of drug. The drug concentrations were aggressively increased with each serial passage. After 8-12 passages a unique mutation at codon 70 occurs which causes a 9-fold decrease in susceptibility to adefovir. In another experiment in H9 cell lines with HIV_{LAI}, concentrations of drug were increased more gradually. After more than thirty serial passages, a mutation was detected at position 65, which decreased susceptibility to adefovir by sixteen-fold. This mutation has been observed in 10-15% of patients treated with ddC. Aside from this, adefovir retains its activity against isolates resistant to all other reverse transcriptase inhibitors, including those containing the 151 mutation with confers resistance to AZT, ddI, ddC, d4T, 3TC and abacavir (Herrington 1997).

Over long-term follow-up of the ongoing clinical studies, researchers from Gilead have had little success in finding mutations in the reverse transcriptase enzyme in patients treated with adefovir (Miller 1998). The mutation at position 70 was observed in some viral isolates from one patient on adefovir monotherapy. Even so, at this timepoint the patient had a 0.9 reduction of viral load from baseline. Virus containing this mutation is 3- to 4-fold less competent than wild type virus, which could explain the continued suppression of viral load. Alternatively, Gilead may simply have caught the beginning of resistance and drug failure. Only a few other conserved mutations were observed, but in patients on concurrent therapies, all of whom continued to sustain reductions in viral load.

ADVERSE EVENTS & DRUG INTERACTIONS

A total of 366 serious or Grade 3/4 adverse clinical or laboratory events have been reported in 216/2,200 patients treated with adefovir as of March 31, 1998. The majority (67%) of these events were minor laboratory abnormalities that either were treated through, or that were reversible with therapy discontinuation.

In Study 402, there were few adverse events in the two lower doses. Four out of 18 patients experienced nausea, one case of vomiting, and two cases of eructation. At the discontinued higher dose, nausea, anorexia, vomiting, and flatulence were all observed. More adverse events occurred during the longer follow-up of study 403 (below).

Adefovir Adverse Events: Gilead Study 403

Grade 2-4 events	*Placebo*	*125 mg*	*250 mg*
N	24	24	24
Nausea	0	3	6
Diarrhea	1	2	4
Asthenia	2	1	4
Headache	1	2	1
Pain	1	2	1
Sinusitis	2	3	0

(Deeks 1996)

But in Study 408, a troubling incidence of Proximal Renal Tubular Dysfunction (PRTD), a kideney disorder, was seen in patients treated with adefovir. PRTD was defined in this study as more than three of the following laboratory abnormalities within two study visits:

* Serum creatinine $\geq$0.5 mg/dL above baseline
* Serum phosphate <2.0 mg/dL
* Serum bicarbonate <17mEq/L
* Proteinuria $\geq$2+
* glucosuria $\geq$1+

Most cases seen were mild to moderate. Onset is slow, normally occurring after about 24 weeks of treatment. In study 408, 1% of patients taking adefovir had PRTD at week 24, by by 48 weeks, approximately 33% of patients were affected. Typically, the syndrome is handled clinically by [ither reducing the dose of adefovir from 120 mg to 60 mg a day, or by discontinuing treatment.

Adefovir Nephrotoxicity: Proximal Renal Tubular Dysfunction (PRTD) in Gilead 408	
N	400
N developing PTRD	83/400 (20.75%)
N whose PTRD resolved*	62/83 (74.7%)
Resolved on 120 mg	7
Resolved on 60 mg	8
Resolved with Drug Interruption	6
Resolved with Drug Discontinuation	41
N whose PTRD did not resolve	21/83 (25.3%)
<8 weeks follow-up	16
>8 weeks follow-up	5

(Barriere 1998)

A quick calculation reveals that, at least in this study, about 5.25% (one in twenty) of people developed irreversible kidney damage. The relationship between overall dose exposure and duration of exposure and degree or duration of kidney damage is not clear from these data. Clearly, adefovir is a drug to be handled with care -- and one which needs longer term studies before it can deemed truly safe.

Gilead states that potential risk factors for PRTD include baseline creatinine clearance, concomitant medications, diabetes mellitus and hypertension. The company recommends that adefovir be suspended fora serum creatinine level $\geq$0.5 mg/dL above baseline or a descrease in serum phosphate to <2.0 mg/dL. Gilead proposes that adefovir may be reinstituted at half dose if serum creatinine returns to <0.3 mg/dL or pre-treatment value, and serum phosphate returns to >2.5 mg/dL and all other relevant laboratory values have returned to pre-treatment levels. Adefovir metabolism also depletes levels of L-carnitine, necessitating supplementation with oral L-carnitine 500 mg per day.

PHARMACOKINETICS, FOOD & DRUG INTERACTIONS

Adefovir has a long intracellular halflife that allows for once-daily dosing. Adefovir has a terminal serum half-life of approximately 5 hours. Its Cmax is dose proportional at the doses tested. It is renally excreted in unchanged form. There appears to be no drug accumulation over time.

Food effects. In a fasted state, adefovir is approximately 30% orally bioavailable. Food increases the oral bioavailability to 40%.

Dosing requirements. Based upon the dose evaluation studies, Gilead chose to use 120 mg dose in a number of its major pivotal. However, no dose-response was noted in the dose-ranging studies, and the recent discovery of a high rate of Proximal Renal Tubular Dysfunction (PTRD) in patients treated with 120 mg/day has piqued the manufacturer's interest in experimenting with lower doses. Unfortunately, because the efficacy margin of adefovir is so low, it is very difficult to determine whether a lower dose will also be effective. Gilead's ongoing study comparing 60 mg vs. 120 mg is too small to detect a clinically meaningful difference in antiviral response, and so although it will give us useful information about the drug's safety at 60 mg/day, we are unlikely ever to know whether this drug "works" at the lower dose.

ONGOING & PLANNED TRIALS

Gilead has outlined an ambitious clinical development plan for adefovir. Most of these studies are currently underway, except where noted.

Study 407, the CPCRA clinical endpoint study. This study is particularly ambitious, seeking to randomized 2,160 patients, with AIDS or with 100 cells or less, to adefovir (120 mg daily) or placebo both in combination with concommitant antiretroviral therapy (with change of concurrent therapy permitted). Unfortunately, accrual has been slow. The current plan is for the study to last around two and a half years (12 months after the last patient is enrolled). Given the slow accrual, the potency of current antiretroviral therapies, and the ability to switch or add drugs in this study, it is unclear when or whether the study will be able to show a statistically significant clinical result.

Gilead may be able to prove a statistically signicant benefit with a meta-analysis, by combining the CPCRA study results with a virtually identical trial being conducted in Europe and Australia. Study 410, or the ADHOC trial will also randomize ~2,000 patients with 100 CD4 cells or less to standard therapy plus adefovir or placebo. The study's endpoints will be survival and end-organ disease.

Lest adefovir come to market with no specific information about how to use it with other antiretrovirals, Gilead is initiating a number of smaller surrogate marker studies of specific combinations in early, intermediate, and advanced disease, including studies in protease failures.

Study 415 (ADHAART) will evaluate whether adefovir can extend the durability of highly active antiretroviral therapy (HAART). The trial will include around 60-120 patients on 3 months of stable HAART, CD4 cell counts over 200, and viral loads below 500 copies. Participants will be randomized to continue HAART alone, or add adefovir, and will continue for 48 weeks. Study endpoints are time to return of viral load to detectable, and time to eradication of tissue burden.

Study 417 will be a blinded dose comparison and triple combination 48-week study in at least 120 patients (Gilead is considering increasing the sample size to 200). The study will randomize subjects with CD4 cell counts over 100 and no prior protease inhibitor therapy to receive adefovir (60 or 120 mg daily) plus 1) nelfinavir/saquinavir (soft gel), 2) nelfinavir plus a nucleoside analogue, or 3) saquinavir (soft gel) plus a nucleoside analogue. The study will compare changes in viral load and CD4 cell counts between both doses, and between the three treatment arms.

ACTG 359 is a blinded triple/quadruple combination study in 400 indinavir failures, with viral loads over 5,000 copies. Changes in HIV RNA and safety will be monitored for 24 weeks, with a possible 24 week extension. The study arms are 1) saquinavir/ritonavir + adefovir, 2) saquinavir/ritonavir + adefovir + delavirdine, 3) saquinavir/ritonavir + adefovir + DMP 266, 4) saquinavir/ritonavir + DMP 266, and 5) saquinavir/ritonavir + delavirdine.

Study 419, at Stanford, will be in thirty patients with over 100 CD4 cells, viral loads over 5,000 and with more than six months prior protease and nucleoside analogue therapy. Participants in the 48 week study will be randomized to adefovir + nelfinavir + saquinavir + nevirapine, or nelfinavir + saquinavir + d4T + ddI.

Study 418, the pediatric study. In this 24-week trial, 30-40 protease inhibitor-naive HIV positive children over 20 kg in weight and able to swallow pills will be randomized to adefovir (1.5 or 3.0 mg/kg) for two weeks. Then patients will add nelfinavir and another nucleoside analogue. The study will monitor adefovir pharmacokinetics, and multiple dose safety, and changes in HIV RNA and CD4 cells.

Expanded access & compassionate use. Gilead's expanded access program began in mid-December 1997. Eligibility requirements include:

- Prior failure of a regimen including at least two commercially available nucleoside analogues and one commercially available protease inhibitor;
- Inability to construct a viable therapeutic regimen from currently marketed antretroviral drugs;
- Failure to qualify for other adefovir studies.

Participants will be advised to begin at least one new antiviral along with adefovir. This new agent could be another experimental drug. This program will include a randomization between 60 mg of adefovir a day versus 120 mg a day, along with L-carnitine (500 mg), a nutritional supplement as the body's reserves may be depleted by adefovir (although the clinical meaning of this depletion is not known). The company is using the same contract research organization (CRO) to administer the expanded access program that DuPont is using for its efavirinz expanded access program (see below), which should facilitate patient referral across the two programs. Gilead has been in discussions with Dupont Pharmaceuticals, as well as Glaxo-Wellcome, in an effort to coordinate cooperation and data collection on patients who are simultaneously enrolled in multiple expanded access programs. Physicians interested in obtaining adefovir via this program may call Gilead at 1.800.GILEAD.5.

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