

MEMORANDUM

DATE: September 6, 2001

TO: Advisory Committee Members and Guests

FROM: Tenofovir Review Team

THROUGH: Debra Birnkrant, M.D.
Acting Director
Division of Antiviral Drug Products

SUBJECT: Background Package for NDA 21-356: VIREAD (tenofovir disoproxil fumarate)

I. Summary of Regulatory Issues and Purpose of Meeting

This document provides background and perspective on the October 3, 2001, Antiviral Drug Products' Advisory Committee meeting. On this day, the committee will be asked to consider efficacy and safety data submitted to support the accelerated approval of tenofovir disoproxil fumarate 300 mg tablets for the treatment of HIV infection.

FDA's analysis of the safety and efficacy data submitted in the NDA support the applicant's results and conclusions. This NDA provides clear evidence of tenofovir's antiviral activity (over 24 weeks) when added to a stable background regimen. In two pivotal studies statistically significant reductions in HIV RNA were observed in antiretroviral-experienced patients. These results are noteworthy given that the majority of approvals have been based primarily on studies in treatment-naïve patients. For many drugs, there is a need for more data characterizing their activity in treatment-experienced patients with prior exposure to the same drug class.

Although tenofovir's risk benefit ratio was favorable among those patients included in the principal studies, the Division is convening this meeting to solicit the committee's comments on four issues: 1) the breadth of the treatment indication, 2) the non-clinical and clinical assessment of bone toxicities, 3) the clinical implications and limitations of the resistance data and 4) the design of a second confirmatory study for traditional approval. Given the diversity of these issues, we have invited several committee guests, some with expertise in bone metabolism and others with expertise in HIV resistance. The text and appendices of this background document provide detailed information related to these issues.

The applicant is proposing a broad indication for the treatment of HIV infection based on the results of two principal trials. However, in both of these studies the antiviral activity of tenofovir when added to stable background therapy was assessed in antiretroviral experienced patients with relatively low plasma HIV RNA levels and high CD4 cell counts (mean baseline values were 3.4 log₁₀ copies/mL and 410 cells, respectively). In both studies treatment with tenofovir produced sustained mean decreases in HIV RNA levels approximating 0.5 log₁₀. As discussed in section III, CD4 changes in these studies were modest. However, small changes in CD4 cell counts may be expected in treatment experienced patients with relatively stable CD4 cell counts at baseline. Given that the study population in this NDA was quite select, we would like to hear the committee's discussion regarding the most appropriate indication for tenofovir. Please refer to section III for a summary of the clinical efficacy data.

The second issue relates to the potential for tenofovir-induced bone toxicity in humans as observed in the animal toxicology data. Reductions in bone mineral density (BMD) were observed in three animal species following tenofovir treatment. Bone mineral loss occurred in juvenile rhesus monkeys following ten months of subcutaneous administration of tenofovir 30 mg/kg/day. Decreased bone mineral content and density was observed in rats at oral doses of 300 and 1000 mg/kg and in dogs at doses of 30 mg/kg given for 13 or 42 weeks. Mechanisms for changes in bone mineral density have not been defined but may have been a consequence of renal tubular reabsorption defects and/or a direct drug-related decrease in intestinal absorption of phosphate in some species. Review of the clinical trial data suggests no clinically significant changes in phosphate, calcium, PTH or BMD over time; however, PTH and BMD were only available for a small subset of patients. In addition, no obvious increases in fracture rates were observed over time in studies 902 and 907. Please refer to Section VB for further details on bone abnormalities.

After reviewing the entire animal toxicity data, along with human data, it is not likely that tenofovir related clinical fractures will occur over 48 weeks assuming the mechanism for bone abnormalities is mediated by renal phosphate wasting or decreases in intestinal absorption of phosphate. However, the exact mechanism of bone toxicity is unknown. Controlled safety data in more patients receiving tenofovir for longer periods of time are needed to address definitively the potential for bone toxicity in the clinical setting. This may be accomplished in an ongoing phase 3 study (#903) to be used for traditional approval (See section VI).

During the meeting we will be seeking your assessment of both nonclinical and clinical data with regard to bone abnormalities. We would like your general impression of the clinical implications of these data and your recommendations for additional preclinical and/or clinical studies to address the potential for bone toxicity. Additionally, we would like to hear your recommendations for long term monitoring for this potential toxicity

The third issue relates to the clinical virology data. Please refer to section IV for further details. This NDA included more clinical resistance data than any previous NDA for an antiretroviral drug. Prospective and exploratory analyses were conducted to evaluate HIV RNA response according to the presence of baseline mutations and baseline susceptibility. The genotypic and phenotypic data suggest potential for some cross resistance between tenofovir and abacavir, didanosine, stavudine, zalcitabine and zidovudine. Cross resistance was not observed between lamivudine and tenofovir. For some primary NRTI mutations or multi-drug resistance NRTI mutations, there were too few patients expressing these mutations at baseline to determine the clinical significance. It appears that the number and type of thymidine analogue mutations (TAMs, defined as M41L, D67N, K70R, L210W, T215Y/F and K219Q) present at baseline affect tenofovir efficacy.

The phenotypic data showed reduced response rates at > 4 fold reduced tenofovir susceptibility. Also decreased susceptibility to zidovudine at baseline affected virologic response. It does not appear that treatment with tenofovir altered the susceptibility of other NRTIs over 24 weeks compared to placebo.

We would like the committee's comments on the clinical resistance analyses conducted during the development of tenofovir. We would also like the committee to comment on the type of resistance data warranting inclusion into package inserts, including tenofovir and other drugs in the pipeline.

Finally, we will be asking the committee to comment on the applicant's proposed second confirmatory study for traditional approval and suggestions for alternative study designs or patient populations that should be studied in phase 4. A summary of the proposed confirmatory study can be found in section VI.

II. Clinical Development Summary

This NDA contains clinical data from four trials conducted with tenofovir tablets, including 2 pivotal studies (GS-98-902 and GS-99-907) and two supportive studies. Both pivotal studies were similar in design and evaluated the safety and efficacy of tenofovir versus placebo when added to a stable antiretroviral regimen in patients with prior nucleoside analogue experience. In addition, activity against nucleoside-resistant HIV was assessed in prospectively defined resistance subgroups.

Two supportive studies were also submitted. Study 901 investigated 5 doses of tenofovir vs. placebo in treatment naïve and experienced patients for 35 days. Study 908 was a compassionate use safety study in patients with limited therapeutic options. Summaries of these trials are provided in Table IIA.

Table IIA Summary of Clinical Trials

Study	Design	Regimens	# Enrolled	Pt Population	Entry CD4 criteria	Entry RNA criteria	Duration of Treatment	Endpoints
901	Randomized Double-Blind Placebo Controlled Dose Escalation	75 mg 150 mg 300 mg 600 mg 75 mg +HU Placebo	Blinded: N=59 Extended dosing N=7	HIV+	≥ 200	≥10,000	Blinded: 35 days Extended dosing: 12 months	Safety, PK, RNA and CD4
902	Randomized Double-Blind Placebo Controlled	75 mg 150 mg 300 mg placebo	Blinded: N=189 Extended dosing N=135	Tx experienced Must be on stable antiretroviral regimen	None	≥ 400 - ≤ 100,000	48 weeks 24 weeks randomized controlled Extended dosing: 12 months	RNA, CD4, safety, bone density
907	Randomized Double-Blind Placebo Controlled	300 mg Placebo	552	Tx experienced Must be on stable antiretroviral regimen	None	≥ 400 - ≤ 10,000	48 weeks 24 weeks randomized controlled	RNA, CD4, safety
908	Open label Safety Study	300 mg	291	Tx experienced Must be on stable antiretroviral regimen	≤ 50 or >50 and < 200 with OI	≥ 10,000	96 weeks	Safety

III. Summary of Efficacy

A. Dose Selection:

The choice of the 300 mg dose was based on results from studies 901 and 902. In studies 901 and 902, decreases in HIV RNA were greater in the 300 mg dose group compared to 75 mg and 150 mg dose groups. No further HIV RNA reductions were observed for the 600 mg dose in study 901. Based on pharmacokinetic, safety and activity data from studies 901 and 902, Gilead chose the 300 mg dose for phase 3 studies and marketing.

B. Study Design and Baseline Demographics:

Studies 902 and 907 were similar in design and enrolled patients with similar baseline characteristics. As shown in Table IIIA, both studies predominately enrolled Caucasian men with a mean age of 41 years and approximately 4 years of prior antiretroviral treatment. Study 902 enrolled patients with baseline HIV RNA levels between 400 and 100,000 copies/mL, whereas study 907 restricted baseline HIV RNA levels to values between 400 and 10,000 copies/mL. Consequently, the mean baseline HIV RNA level in study 902 was slightly higher than that of 907, whereas the mean baseline CD4 count was slightly higher in study 907.

Table IIIA: Baseline Characteristics: Studies 902 and 907

	Study 902	Study 907
Number of Patients Randomized	189	552
Number of Patients Receiving at Least 1 Dose of Study Drug	186	550
Age (Years)		
Mean	41.9	41.6
Median	41	40
Range	27 - 62	22 - 70
Sex		
Male	171 (92%)	469 (85%)
Female	15 (8%)	81 (15%)
Race		
Caucasian	138 (74%)	379 (69%)
Black	24 (13%)	92 (17%)
Hispanic	21 (11%)	68 (12%)
Other	3 (2%)	11 (2%)
CD4 Cell Count (cells/mm³)		
Mean	374	427
Median (Range)	331 (9 - 1240)	386 (23 - 1385)
N < 200	43	66
N ≥ 200	140	483
HIV RNA (copies/mL)		
Mean	15422	4457
Median (Range)	5057 (52-580193)	2431 (50-75000)
N < 5,000	92	407
N ≥ 5,000	94	142
Prior Antiretroviral Experience		
Median Time on Prior Antiretroviral Therapy	4 Years	4.9 Years
N < 4 prior agents	112	95
N ≥ 4 prior agents	74	454

C. Primary Efficacy Endpoint - HIV RNA:

The primary efficacy endpoint in the two pivotal studies was the time-weighted change in \log_{10} HIV RNA over 24 weeks (DAVG₂₄). Such analyses are useful for assessing antiviral activity in patients in whom plasma HIV-RNA reductions below the assay limit may not be frequently achieved. Therefore DAVG₂₄ was deemed an acceptable endpoint for evaluating virologic responses in treatment-experienced patients, such as those enrolled in studies 902 and 907. Secondary endpoints in these studies included the proportion of patients achieving HIV RNA levels below 400 and 50 copies/mL.

1. HIV RNA Results:

Table IIIB summarizes the efficacy results for studies 902 and 907. In both studies statistically significant differences of approximately 0.5-0.6 \log_{10} were observed for the primary efficacy endpoint (DAVG₂₄) favoring the tenofovir 300 mg over placebo. In study 902, patients receiving 300 mg had sustained HIV RNA reductions over 48 weeks that were larger than those receiving either 150 mg or 75 mg.

In study 902 there were no numerical differences between the 4 treatment groups with respect to the proportion of patients achieving HIV RNA levels < 400 or < 50 copies/mL. However, for patients with extensive prior antiretroviral therapy and/or higher baseline viral loads (> 5,000), the addition of one drug to a background regimen may not be expected to decrease HIV RNA to levels below assay limits. Of note, study 902 may not have been of sufficient size to detect treatment differences for the secondary endpoint of proportion below an assay limit.

In study 907, a much larger study than 902, a greater proportion of patients in the tenofovir group achieved HIV RNA < 400 copies/mL compared to placebo. In addition, baseline HIV RNA levels in this study were lower than that of study 902, making it more feasible to achieve HIV RNA levels below an assay limit.

Table IIIB. Summary of Efficacy (ITT)

	Study 902				Study 907	
	Placebo (N=28)	Tenofovir			Placebo (N=182)	Tenofovir (N=368)
		75 mg (N=53)	150 mg (n=51)	300 mg (N=54)		
Mean DAVG ₂₄	+0.02	-0.26	-0.34	-0.58	-0.03	-0.61
Mean DAVG ₄₈	NA	-0.33	-0.34	-0.62	NA	NA
<400 copies/mL (week 24)	6/28 (21%)	12/53 (23%)	14/51 (27%)	14/54 (26%)	21 (12%)	154 (42%)
<50 copies/mL (week 24)	3/28 (11%)	7/53 (13%)	6/51 (12%)	7/54 (13%)	2 (1%)	76 (21%)

In study 902 the protocol permitted changes to background therapy after week four. Changes to background regimens were discouraged in study 907; however, some patients did change their regimen. FDA conducted exploratory analyses to evaluate the impact of the addition of a new agent on the overall study results. Overall, treatment changes did not appear to affect the HIV RNA response. FDA concluded that the decline in HIV RNA in the tenofovir 300 mg groups observed in both studies was not attributed to the addition of another drug.

HIV RNA Results According to Demographic/Baseline Characteristics

Subgroup analyses are presented below for study 907. The applicant conducted several subgroup analyses based on age, sex, and race. Statistically significant differences in HIV RNA decreases favoring tenofovir over placebo were observed for each of the age, race and sex subgroups.

Randomization for study 907 was stratified according to baseline HIV RNA (<5000 , ≥ 5000 copies/mL), baseline CD4 (<200 , ≥ 200 cells) and prior antiretroviral therapy (< 4 agents, ≥ 4 agents). Treatment differences between the 300 mg group and placebo were significant favoring tenofovir for all randomization strata.

In addition, FDA conducted statistical tests to examine interactions between the randomization subgroups for viral load, CD4 cell counts and prior antiretroviral use. These tests explore whether the size of the treatment difference between tenofovir 300 mg and placebo is similar between subgroups. A statistically significant treatment interaction was observed for the subgroup of patients with $< 5,000$ vs $\geq 5,000$ copies/mL (p-value=0.077 significant at $\alpha=0.15$ level). This treatment interaction suggests that reduction in viral load is likely to be lower in patients with $\geq 5,000$ copies/mL at baseline (net treatment effect -0.62 vs -0.45 log reduction for $< 5,000$ vs $\geq 5,000$, respectively). Given the smaller reduction in viral load observed in patients with baseline HIV RNA $\geq 5,000$ copies/mL, it does raise some question regarding the activity of tenofovir in patients with higher baseline viral load. This issue could not be fully answered in study 907 because the entry criteria were restricted to patients with viral loads between 400 and 10,000 copies/mL. Additional data from patients with higher baseline viral loads, particularly those with HIV RNA levels $> 10,000$ copies/mL, are needed.

Similarly, a statistically significant interaction was also observed for the subgroup of patients with prior use of < 4 prior antiretroviral agents vs ≥ 4 prior antiretroviral agents (p-value=0.009 significant at $\alpha=0.15$ level). This treatment interaction suggests that reduction in viral load is likely to be lower in patients who received ≥ 4 prior antiretroviral agents (net treatment effect -0.80 vs -0.54 for < 4 prior agents vs ≥ 4 prior agents, respectively).

No significant interaction was seen for the subgroups based on baseline CD4 cell counts. The net benefit in reduction of viral load due to tenofovir was similar in patients who had either < 200 or ≥ 200 cell/mm³ at baseline.

2. CD4 Cell Count Responses

CD4 cell count changes are presented in table IIIC. In study 902, the mean $DAVG_{24}$ for CD4 cell counts was -10.5 cells/mm³ for the 300 mg group compared to -3.6 cells/mm³ for the placebo group. The difference between the 300 mg tenofovir dose compared to placebo was not statistically significant.

For study 907, there was a statistically significant difference in the $DAVG_{24}$ for CD4 favoring tenofovir. The mean $DAVG_{24}$ was $+12$ cells/mm³ in the tenofovir group compared to -10 cells/mm³ in the placebo group, resulting in a net treatment difference of approximately 20 cells/mm³.

Table IIIC: FDA Analyses: CD4 Cell Count Response

	Study 902		Study 907	
	Placebo (N=28)	Tenofovir 300 mg (n=54)	Placebo (N=182)	Tenofovir 300 mg (n=368)
Mean DAVG₂₄ (\pm SD)	-3.6 (80)	-10.5 (81)	-10.6 (88.4)	12.6 (78.4)

To further investigate the modest changes in CD4 cell counts observed in these studies, FDA conducted subgroup analyses to evaluate CD4 responses by baseline CD4 cell count based on pooled data. Patients with baseline CD4 cell counts < 200 had similar CD4 responses compared to patients with baseline CD4 counts \geq 200 (See Table IID). This finding is encouraging because CD4 increases in patients with lower cell counts are important for minimizing the risk of opportunistic infections.

It is also important to note that the study populations in studies 902 and 907 may not have been optimal for observing large increases in CD4 cell counts, given the fact that only one new drug was added to a stable background regimen. Similar results were reported for the abacavir CNA2003 study. CNA2003 was similar in design to tenofovir studies 902 and 907. The activity of abacavir versus placebo when added to stable antiretroviral regimens was evaluated in treatment experienced patients. No statistically significant differences for change in CD4 from baseline was noted between abacavir and placebo; however there was a numerical difference favoring the abacavir group (+30 vs +1 cells/mm³). Results from studies CNA2003, 902 and 907 suggest that small changes in CD4 cell counts may be expected in treatment experienced patients with relatively stable CD4 cell counts at baseline. For these patients, the addition of one new agent to a stable background antiretroviral regimen did not produce substantial increases in CD4 cell counts over time. Further evaluations of CD4 cell counts in studies with different designs are needed.

Table IIID: CD4 Cell Count Response by baseline CD4: Studies 902 and 907

Baseline CD4 group	Placebo (N=210)	Tenofovir 300 mg (N=422)	Net Treatment Effect	Difference 8.6 P=0.65
< 200 cells/mm ³	-6.5	20	26.5	
\geq 200 cells/mm ³	-10	7.9	17.9	

IV. Summary of Clinical Virology:

A. Baseline Genotype:

Studies 902 and 907 included analysis plans to prospectively evaluate HIV RNA response according to the presence of baseline mutations. Primary analysis plans specified the evaluation of thymidine analogue mutations (TAMs) and the M184V mutation. The six mutations commonly defined as TAMs are M41L, D67N, K70R, L210W, T215Y/F and K219Q. Secondary analyses evaluated virologic response by other primary NRTI, NNRTI and PI associated mutations.

Table IVA summarizes FDA analyses of virologic response for the protocol specified baseline mutation groups. These analyses incorporated pooled data from studies 902 and 907. As shown in Table IVA, decreases in HIV RNA were observed in tenofovir treated patients with baseline TAMs or M184V. The data suggest that tenofovir's activity was not affected by the M184V mutation, indicating absence of cross resistance between tenofovir and lamivudine

The applicant also performed analyses to determine if the presence of the M184V mutation at baseline resulted in better responses to tenofovir compared to those without this mutation. These results did not show “enhanced” activity when the M184V mutation was present despite in vitro evidence of increased tenofovir susceptibility in the presence of this mutation. Tenofovir treated patients with M184V and TAMs had slightly greater HIV RNA decreases compared to patients with TAMs and no M184V (DAVG₂₄ -0.52 vs -0.45 log₁₀). However, when considering the effect of placebo, the net treatment effects in these groups were similar (-0.60 vs -0.58 log₁₀).

Table IVA RNA Responses by Baseline Resistance Mutations (ITT)^A

Baseline Mutation Group	Mean DAVG ₂₄ (n)		Net Treatment Effect
	Placebo	Tenofovir	
All Patients	-0.03 (110)	-0.59 (222)	-0.56
No M184V	+0.08 (40)	-0.42 (73)	-0.50
M184V	-0.08 (70)	-0.67 (149)	-0.59
M184V / No TAM ¹	-0.12 (20)	-0.96 (51)	-0.84
No TAM ¹	-0.11 (29)	-0.80 (68)	-0.69
TAM ¹	0.00 (81)	-0.50 (154)	-0.50
TAM ¹ / No M184V	+0.13 (3)	-0.45 (56)	-0.58
TAM ¹ + M184V	-0.08 (50)	-0.52 (98)	-0.60
T215Y/F	+0.03 (53)	-0.35 (106)	-0.38
T215Y/F / No M184V	+0.14 (24)	-0.37 (44)	-0.51
T215Y/F + M184V	-0.06 (29)	-0.34 (62)	-0.28
T69D/N	-0.08 (19)	-0.48 (25)	-0.56
L74V/I	+0.11 (16)	-0.17 (18)	-0.28
K65R	0	-0.01 (6)	-0.01
Q151M	+0.05 (2)	+0.38 (2)	+0.33
T69S Insertions	0	+0.29 (2)	+0.29
NNRTI-R ²	+0.06 (53)	-0.50 (97)	-0.56
PI-R ³	0 (70)	-0.52 (129)	-0.52

^APatients included in these subgroups may have other TAMs or mutations in addition to the baseline TAM listed

¹TAM=M41L, D67N, K70R, L210W, T215Y/F or K219Q/E/N (Also known as TAMs)

²NNRTI-R = K103N, V106A, V108I, Y181C/I, Y188C/L/H, G190A/S/E or P263L

³PI-R = D30N, V32I, G48V, I50V, V82A/F/T/S, I84V or L90M

In addition to the protocol-specified analyses evaluating the impact of baseline mutations on virologic response, FDA conducted several additional analyses to evaluate the effect of specific mutations and mutational patterns on virologic outcome. Results of these analyses are presented in Table IVB. Because of the large number of potential comparisons, statistical testing was not conducted. Descriptions of numerical differences are presented below.

Table IVB: HIV RNA Response by Baseline TAMs

Baseline TAMs ¹	Mean DAVG ₂₄ (N)			
	MUTATION PRESENT		MUTATION ABSENT	
	Tenofovir	Placebo	Tenofovir	Placebo
Any TAM	-0.50 (154)	0 (81)	-0.80 (68)	-0.11 (29)
M41L	-0.26 (81)	+0.06 (40)	-0.78 (141)	-0.07 (70)
D67N	-0.53 (79)	-0.03 (43)	-0.62 (143)	-0.02 (67)
K70R	-0.71 (67)	-0.03 (40)	-0.54 (155)	-0.02 (70)
L210W	-0.17 (46)	+0.06 (22)	-0.70 (176)	-0.05 (88)
T215Y/F	-0.35 (106)	+0.03 (53)	-0.80 (116)	-0.07 (57)
K219Q/E/N	-0.60 (57)	+0.11 (27)	-0.58 (165)	-0.07 (83)

¹Patients included in these subgroups may have other TAMs or mutations in addition to the baseline TAM listed

First, these analyses show that mean virologic responses were slightly greater (first row) among patients without any TAMs compared to those with at least one TAM. Second, the activity of tenofovir appeared to be diminished in patients expressing the M41L, L210W or T215Y/F mutation compared to patients who did not have these mutations at baseline. Tenofovir associated decreases in HIV RNA were approximately 0.5 log₁₀ less when these mutations were present compared to when they were absent at baseline.

Several additional exploratory analyses were then conducted to further investigate the impact of these mutations on virologic response. Table IVC summarizes the results from these analyses. Although the above analysis showed a diminished response in tenofovir treated patients expressing the T215Y/F mutation at baseline it was found that this mutation might not have directly affected the activity of tenofovir. The diminished response noted in patients expressing the T215Y/F mutation at baseline appears to be due to the presence of the M41L or L210W mutation and not the T215Y/F mutation. Patients expressing the T215Y/F mutation at baseline without the M41L or L210W mutation had a -0.70 log₁₀ decrease through week 24 compared to a -0.25 log₁₀ decrease if the T215Y/F mutation was present with the M41L or L210W mutation. Additional supportive evidence that the T215Y/F mutation does not affect HIV RNA response is seen in patients with the T215Y/F mutation alone and in patients with T215Y/F mutation in addition to the D67D, K70R and K219Q/E/N. HIV RNA responses for these patients appeared to be unaffected by the T215Y/F mutation, in fact responses were similar to the overall group.

Table IVC: Impact of T215Y/F Mutation on Virologic Response

Baseline Mutations ¹	Mean DAVG ₂₄ (N)		Net Treatment Effect
	Placebo	Tenofovir	
T215Y/F	+0.03 (53)	-0.35 (106)	-0.38
No T215Y/F	-0.07 (57)	-0.80 (116)	-0.73
T215Y/F / No 41 or 210	-0.01 (13)	-0.70 (25)	-0.69
T215Y/F + 41 or 210	+0.04 (40)	-0.25 (82)	-0.29
D67N+K70R+T215Y/F+K219Q/E/N	+0.37 (3)	-0.60 (12)	-0.97
T215Y/F alone	+0.18 (2)	-1.02 (8)	-1.20

¹Patients included in these subgroups may have other TAMs or mutations in addition to the baseline TAM listed

It also appears that the number and type of TAMs present at baseline also affected tenofovir efficacy. Tenofovir efficacy was diminished in patients with ≥ 3 TAMs in the presence of the M41L or L210W mutation compared to patients with ≥ 3 TAMs in the absence of the M41L or L210W mutation. HIV RNA responses by number and type of baseline TAMs are summarized in Table IVD.

Table IVD: HIV RNA Response by Number of Baseline TAMs

# baseline TAMs	Mean DAVG ₂₄ (N)	
	Tenofovir	Placebo
No TAMs	-0.80 (68)	-0.11 (29)
Any TAMs	-0.50 (154)	0 (81)
1 –2 TAMs	-0.66 (55)	-0.4 (33)
3 TAMs	-0.44 (59)	+0.04 (29)
≥ 4 TAMs	-0.35 (40)	+0.03 (19)
≥ 3 TAMs + M41L or L210W	-0.21 (57)	+0.01 (29)
≥ 3 TAMs / No M41L or L210W	-0.67 (42)	+0.7 (19)

The affect of other primary NRTI mutations on tenofovir efficacy was also assessed. However for some primary NRTI mutations or multi-drug resistance NRTI mutations, there were too few patients expressing these mutations at baseline to determine the clinical significance.

The L74V/I mutation, a primary mutation conferring resistance to abacavir, didanosine and zalcitabine, also affected tenofovir efficacy. Patients with the L74V/I mutation did not appear to respond to tenofovir treatment. The mean DAVG₂₄ for this group was $-0.17 \log_{10}$. Patients expressing the L74V/I mutation at baseline were further evaluated to determine if the diminished response was attributed to the presence of other NRTI mutations, specifically TAMs. Response rates were similar (-0.12 to $-0.19 \log_{10}$) regardless if the M41L or L210W mutation was present or absent. This finding suggests the potential for cross resistance between tenofovir and didanosine; however, more data from patients with this mutation are needed to make any definitive conclusions.

Viruses expressing the K65R mutation have been shown to reduce susceptibility to tenofovir in vitro. Other NRTIs can also select for this mutation; therefore an analysis was conducted to determine if K65R mutation at baseline affected tenofovir activity. Six patients in the tenofovir group had the K65R mutation present at baseline. No placebo patients expressed this mutation at baseline. Patients with the K65R mutation did not appear to respond to tenofovir treatment (mean DAVG₂₄ =0).

B. Development of HIV mutations by Weeks 24- 48:

Since tenofovir can select for the K65R mutation in vitro, the development of this mutation over time was assessed. Overall in studies 902 and 907, 7 patients developed the K65R mutation and the mean DAVG₂₄ for this group was $-0.22 \log_{10}$ copies/mL. More patients are needed in order to assess the clinical relevance of the development of this mutation.

In study 907, the proportion of patients who developed NRTI, NNRTI or PI associated mutations by week 24 was less in the tenofovir group than in the placebo group. Similar results were also seen in study 902. The development of NNRTI or PI associated mutations was infrequent in this trial (8-9%). Twenty-two percent and 39% of patients in the 300 mg group developed a NRTI associated mutation by weeks 24 and 48, respectively. With the exception of the K65R mutation, the development of HIV-associated mutations did not appear to adversely affect tenofovir efficacy.

C. Phenotypic Analyses:

Responses to tenofovir treatment by baseline ZDV and tenofovir susceptibility are summarized in Table IVE below. Patients with baseline tenofovir susceptibility within 3 fold of wild type virus experienced a -0.55 to $-0.74 \log_{10}$ decrease in HIV RNA through week 24. Nine patients had a greater than 4 fold reduced susceptibility to tenofovir at baseline. These patients did not respond to tenofovir therapy, (mean DAVG₂₄ $-0.12 \log_{10}$). Although there are too few patients to determine a susceptibility breakpoint for tenofovir at this time, this dataset shows that tenofovir efficacy is

diminished in patients with reduced susceptibility to tenofovir and zidovudine at baseline. This finding is compatible with the genotypic analyses, in that tenofovir efficacy is reduced in the presence of certain TAMs, ≥ 3 TAMs or the K65R mutation.

Table IV E: HIV RNA Response by Baseline Tenofovir and ZDV Susceptibility

	Tenofovir Mean DAVG ₂₄ (n)
Baseline Tenofovir Susceptibility	
≤ 1	-0.74 (35)
> 1 and ≤ 2	-0.57 (36)
> 2 and ≤ 3	-0.55 (13)
> 3 and ≤ 4	-0.3 (7)
≤ 4	-0.61 (91)
> 4	-0.12 (9)
Baseline ZDV Susceptibility	
≤ 1	-0.84 (18)
> 1 and ≤ 4	-0.73 (39)
> 4 and ≤ 10	-0.37 (21)
> 10	-0.23 (21)

Changes in tenofovir and other NRTI susceptibility during treatment were also assessed over 24 weeks in study 907. The results from these analyses are presented in Table IV F. Overall it appears that changes in susceptibility to tenofovir or other NRTI were minimal over 24 weeks. It does not appear that treatment with tenofovir is altering the susceptibility of other NRTIs over 24 weeks compared to placebo. Of note, the largest change in susceptibility was for ZDV and was most prominent in patients who developed a ZDV associated mutations during the study. It is unclear if tenofovir directly or indirectly contributed to changes in zidovudine susceptibility.

Table IV F: Changes in Susceptibility From Baseline (Study 907)

Pts Developing New NRTI Mutations	N	Mean fold change in Susceptibility from Baseline					
		Tenofovir	ZDV	D4T	DDI	3TC	ABC
Tenofovir Group:	35	2.2	3.6	2.2	1.7	1.4	1.7
Placebo Group	24	1.5	2.6	2.5	1.3	1.8	1.5
All Patients Analyzed	59	1.9	3.2	2.3	1.5	1.6	1.6

D. Summary of Resistance Data

Although this NDA included more clinical resistance data than any previous NDA for an antiretroviral drug, the large amount of potential comparisons in evaluating the impact of resistance limits the ability to conduct tests for statistical significance. Much of the analyses presented above were exploratory and described numerical differences. Given these caveats, the genotypic and phenotypic data suggest potential for some cross resistance between tenofovir and abacavir, didanosine, stavudine, zalcitabine and zidovudine. However for some primary NRTI mutations or multi-drug resistance NRTI mutations, there were too few patients expressing these mutations at baseline to determine the clinical significance. Cross resistance was not observed between lamivudine and tenofovir. It appears that the number and type of TAMs present at baseline affect

tenofovir efficacy. Tenofovir efficacy is reduced in patients with ≥ 3 TAMs which include the M41L or L210W mutation compared to patients with ≥ 3 TAMs without the M41L or L210W mutation.

Viruses expressing the K65R mutation show reduced susceptibility to tenofovir in vitro. Overall 6 patients expressed this mutation at baseline; all 6 patients were in the tenofovir group. These patients did not respond to tenofovir treatment. It appears that the K65R mutation affects tenofovir activity, however, more patients expressing this mutation at baseline are needed in order to make any definitive conclusions regarding the clinical significance of this mutation at this time.

Phenotypic analyses show reduced response rates at > 4 fold reduced tenofovir susceptibility. Also decreased susceptibility to zidovudine at baseline affected response. It does not appear that treatment with tenofovir altered the susceptibility of other NRTIS over 24 weeks compared to placebo.

We look forward to your comments on the types of clinical virology analyses conducted and suggestions for displaying resistance data in package inserts.

V. Safety Summary

Overall treatment with tenofovir appears to be well tolerated and similar to placebo over 24 weeks. The most common events reported were asthenia (19%), headache (14%), diarrhea (22%), nausea (20%) and pharyngitis (18%). More patients randomized to tenofovir compared to placebo experienced GI events, including diarrhea (22% vs 17%), flatulence (6% vs 2%), nausea (20% vs 15%) and vomiting (12% vs 6%). The types of laboratory abnormalities noted during the trials did not differ between groups and were consistent with what is expected in patients receiving multiple antiretroviral agents.

Results from preclinical studies guided a more detailed review of renal and bone abnormalities. These events are summarized below.

A. Renal Toxicity

Preclinical studies showed evidence of renal toxicity in 4 animal species, mouse, rat, dog and monkey. Kidney changes were associated directly with exposure to tenofovir. Renal tubular toxicity occurred after 56 days to 42 weeks of tenofovir treatment in the mouse, rat, dog and monkey. Interstitial nephritis was noted after chronic dosing in dogs. Increases in serum creatinine, BUN, glycosuria, proteinuria, phosphaturia and/or calciuria and decreases in serum phosphate were observed in varying degrees in these animals. These toxicities were noted at exposure levels 2-20 times higher than the human clinical exposures following administration of tenofovir at 300 mg/day. In addition, decreased renal clearance of tenofovir and a Fanconi-like syndrome with glucosuria and hypophosphatemia occurred in monkeys following a dose of 30 mg/kg (subcutaneously) for 11-24 months.

Given the preclinical evidence for renal toxicity, renal parameters were monitored closely during clinical trials. FDA analyses of renal parameters focused on changes in creatinine, phosphate, bicarbonate, proteinuria and glycosuria. With the available data there does not appear to be any significant renal toxicity associated with tenofovir use. It is important to note that another antiretroviral from the class of nucleotide analogues, adefovir, was associated with delayed nephrotoxicity. It will be important to monitor long term changes in renal parameters.

B. Bone abnormalities

Reductions in bone mineral density occurred in three animal species following tenofovir administration. Bone mineral loss was noted in juvenile rhesus monkeys following ten months of subcutaneous administration of tenofovir 30 mg/kg/day. Decreased bone mineral content and density was observed in rats at oral doses of 300 and 1000 mg/kg and in dogs at doses of 30 mg/kg given for 13 or 42 weeks. Two possible mechanisms for bone abnormalities have been suggested. First, changes in bone mineral density are thought to be secondary to renal tubular reabsorption defects. Hypophosphatemia was observed in monkeys and hypercalciuria was seen in rats and dogs. Secondly, findings in rat and monkey studies suggest a direct drug-related decrease in intestinal absorption of phosphate with potential secondary reduction in bone mineral density. Preclinical studies show that renal tubular dysfunction and bone mineral losses are dose-related and generally reversible. Long term administration (up to 10 months) of low dose tenofovir or short term administration with high doses of tenofovir was not associated with bone abnormalities. Bone abnormalities were observed following chronic administration of intermediate and high doses of tenofovir. It is not known if chronic administration (> 42 weeks) of low dose tenofovir will cause bone abnormalities. A summary of the observations for each animal species is summarized below.

1. Bone Abnormalities: Monkeys

Bone lesions were observed in monkeys after 10 months of daily subcutaneous administration of tenofovir 30 mg/kg/day. Bone toxicities were also noted at 2 and 7.5 months of age in neonates following tenofovir exposure in utero during the second trimester. These animals received both pre and postnatal chronic tenofovir treatment.

Bone toxicity characterized by abnormal growth plates and trabeculae, bone deformities and displacements, bone fractures, joint swelling and decreased bone density in the spine or pelvis were noted following tenofovir 30 mg/kg/day SQ. Also moderate to marked decreases in serum phosphorus, increases in alkaline phosphatase, non-hyperglycemic glycosuria and proteinuria were seen in conjunction with the bone changes. A tenofovir dose of 10 mg/kg SQ administered to newborn monkeys daily for 2 years was not associated with skeletal changes or changes in alkaline phosphatase, phosphorus, non-hyperglycemic glucosuria or proteinuria.

A separate study was conducted as part of a master's thesis to evaluate the effects of tenofovir on bone metabolism and cortical bone strength in fetal and juvenile monkeys. Microradiographs showed completely unmineralized secondary osteons consistent with osteomalacia. As a result of these findings, assessment of osteoid accumulation, bone porosity, determination of mineral content, and evaluation of bone fragility were conducted on all bone specimens. Wide osteoid seams were noted in 50% of the tenofovir treated juvenile monkeys. Normal bone remodeling was noted in all untreated juvenile specimens. The applicant concluded that tenofovir 30 mg/kg/day SQ was associated with defective mineralization of osteoids. When the dose was reduced to 10 mg/kg/day or when tenofovir dosing was discontinued osteoid width normalized or showed improvement. It is important to note that the most severely affected monkeys received tenofovir for four months or longer. The author of the master's thesis also concluded, "Although these data clearly show a significant effect of high dose tenofovir administration on bone remodeling, the data presented are largely inconclusive with regard to the relationship between osteoid seam width, microdamage accumulation and bone strength."

2. Bone Abnormalities: Rats and Dogs

Renal toxicity, including increases in creatinine and marked hypercalciuria, was observed in rats following 42 weeks with tenofovir. No evidence of renal phosphate wasting was observed. There was evidence for increased bone turnover, with loss of mineral from both cortical and trabecular bone. In dogs, hypercalciuria, glycosuria, and proteinuria associated with increased bone turnover (bone markers) and loss of bone mineral were observed. In addition to loss of bone mineral and changes in bone turnover markers in rats and dogs, there were increases in PTH levels and decreases in levels of 1,25-dihydroxy-vitamin D₃.

The Division is currently reviewing data to assess if tenofovir has direct toxicity to bone. At this time a definitive conclusion cannot be made until additional data has been submitted and reviewed. Further nonclinical studies may be necessary to determine if tenofovir treatment has any direct effects on bone metabolism. However, there is supportive evidence that tenofovir may not be toxic to bone. No bone abnormalities were observed in monkey fetuses exposed to tenofovir, at AUCs ten times greater than the human exposure, in utero for final two-thirds of gestation. In addition no abnormal bone development in newborn rats or rabbits exposed in utero in reproductive toxicology studies were noted.

3. Bone Abnormalities: Relationship of Animal Exposure to Human Dose

Review of exposure data and bone abnormalities noted in animal studies gives some reassurance that there is a margin of safety for the 300 mg daily dose. AUCs of 18 : g*hr/ml, 30 : g*hr/ml and 97.9 to 240 : g*hr/ml in rats, dogs, and monkeys, respectively were noted in conjunction with bone abnormalities. The average AUC in humans receiving tenofovir 300 mg is 3.34 : g*hr/ml.

Reductions in bone mineral density in rats and dogs occurred at exposure levels 6-10 times higher than the human clinical exposures following administration of tenofovir at 300 mg/day. Bone abnormalities in monkeys were noted at exposure levels 29-80 times higher than the human clinical exposures with tenofovir 300 mg. However, safety margins were lower at doses of tenofovir that elicited no effects in animal toxicology studies.

The variability of drug concentrations in humans were minimal over time, thus providing assurances that individual patient exposures were not approaching those associated with bone abnormalities in the animal studies.

4. Bone Abnormalities: Clinical Data from Studies 902 and 907

Because of the findings of bone toxicity in three animal species, the Division asked the sponsor to monitor markers of bone metabolism using measurements such as bone mineral density (BMD), serum calcium and phosphate, alkaline phosphatase, vitamin D and PTH. However for some measurements, data were only collected for a subset of patients and vitamin D levels were not submitted for review. In addition, the sponsor calculated fractional urinary excretion of calcium and phosphate. These data are discussed below.

a. Clinical Assessment of Bone Abnormalities: BMD

BMD changes of the spine, as measured by DEXA, were evaluated in studies 902 and 907 and were available for 21 patients receiving placebo and 40 patients receiving tenofovir 300 mg. The "All Tenofovir" group includes patients randomized to 75 and 150 mg and patients who were initially randomized to placebo but later crossed over to tenofovir 300 mg. Table VA summarizes mean percent changes from baseline for BMD of the spine. Overall small changes were observed. These changes were not thought to be clinically significant. For study 902 alone there did not appear to be any dose effect.

Table VA: BMD Results: Percent Change From Baseline* – Studies 902 and 907

Time Point	Placebo (0-24 weeks)	Tenofovir 300 mg (0-24 weeks)	All Tenofovir
Week 24	N=18	N=33	N=58
Mean	0.6 +/- 3.46	-1.3 +/- 2.41	-0.8 +/- 2.42
Median	0.9	-0.7	-0.7
Range	-5.0 to 6.6	-5.9 to 3.8	-6.2 to 4.5
Week 48			N=46
Mean	---	---	-0.8 +/- 3.44
Median			-0.7
Range			-10.8 to 6.5
Week 72			N=35
Mean	---	---	-1.4 +/- 3.87
Median			-1.3
Range			-11.2 to 9.9
Week 96			N=17
Mean	---	---	-0.1 +/- 4.48
Median			-1.7
Range			-6.7 to 10.2

*BMD was measured as g/cm²

The applicant assessed "marked changes" in BMD (marked defined as >5% decrease from baseline at the lumbar spine) during the 24 week placebo controlled period. The incidence of marked changes was roughly similar in both groups, 6% for placebo vs. 9% tenofovir.

BMD changes of the hip were evaluated in study 907. No differences were noted between groups. Mean loss of BMD at week 24 was 0.1% and 0.8% in the tenofovir and placebo groups, respectively. No spontaneous fractures of the hip or other weight-bearing bones were reported.

b. Clinical Assessment of Bone Abnormalities: PTH (Study #902)

PTH was evaluated for those patients participating in the BMD substudy of study 902. Samples were collected at baseline, weeks 24, 48, 72 and 96. The applicant reported no marked changes from baseline in median or mean PTH levels in any treatment group through week 24 or in the three tenofovir dose groups through week 96. There did not appear to be a dose relationship for PTH changes. Mean changes in PTH levels from baseline are summarized in Table VB.

Table VB: PTH Results – Substudy 902

Time Point	Placebo (0-24 weeks)	75 mg / 300 mg	150 mg / 300 mg	300 mg / 300 mg
Baseline	21.9 (+/- 9.2) N=8	28.1 (+/- 10.8) N=18	21.6 (+/-7) N=11	24.6 (+/- 12) N=16
Mean change at Week 24	-4.8 (+/- 4.5) N=6	1.2 (+/-10.5) N=17	2.3 (+/-8.8) N=11	-0.9 (+/-11.7) 13
Mean change at Week 48	NA	3.4 (+/-20.3) N=14	10.1 (+/-16.7) N=7	3.4 (+/-11.2) N=10
Mean change at Week 72	NA	10.4 (+/-15.5) N=14	4.3 (+/-11.2) N=8	-0.1 (+/-11.3) N=10

Source: serial 034 dated July 5, 2001

c. Clinical Assessment of Bone Abnormalities: Serum Phosphate and Fractional Excretion of Phosphorus

Serum phosphate changes were assessed over 24 weeks for the 300 mg and placebo groups in studies 902 and 907. Additionally, phosphate abnormalities are summarized below for these groups and the "All Tenofovir" group. The "All Tenofovir" group includes patients randomized to 75 and 150 mg and patients who were initially randomized to placebo but later crossed over to tenofovir 300 mg. Results of these analyses are presented in Table VC below. Through week 24, there was a slightly higher incidence of grade 2 hypophosphatemia in the tenofovir group compared to placebo. Phosphorus abnormalities occurred sporadically throughout the course of treatment in studies 902 and 907, and generally resolved without treatment interruption. Of the 62 patients with a grade 2+ phosphate abnormality, 51 (82%) patients had an abnormal value at only one visit. The abnormality resolved by the subsequent visit. Ten patients had a sustained (≥ 2 consecutive values) grade 2 abnormality. Additionally FDA pooled all available phosphorus data in studies 902 and 907. Based on this analysis, the incidence and severity of phosphate abnormalities did not worsen with increasing duration of tenofovir treatment. This finding is important because the applicant contends that the mechanism for bone abnormalities may be mediated via renal phosphate wasting or decreases in intestinal absorption of phosphate.

Table VC: Serum Phosphate Changes: Studies 902 and 907

	Placebo (N=210) (weeks 0-24)	Tenofovir 300 mg (N=443) (weeks 0-24)	All Tenofovir (N=687)
Mean change from baseline	0.66 mg/dL	0.04 mg/dL	ND

Grade 1 (2-2.2 mg/dL)	10 (5%)	27 (6%)	51 (7%)
Grade 2 (1.5-1.9 mg/dL)	5 (2%)	28 (6%)	58 (8%)
Grade 3 (1-1.4 mg/dL)	1 (<1%)	0	3 (<1%)
Grade 4 (< 1 mg/dL)	0	1 (<1%)	1 (<1%)

Fractional excretion of phosphorus was also calculated for studies 902 and 907. In study 902, patients originally randomized to receive tenofovir had an increase in excretion of urinary phosphorus at each study visit through week 48. The mean change from baseline ranged from 0.2% to 2.3%. At week 24 there was a statistically significant difference between placebo and the tenofovir 300 mg group; -2.3% placebo vs 2.3% tenofovir.

In study 907, no significant differences were noted between groups. Mean change from baseline in fraction excretion of phosphorus at week 24 was 0.4% and -0.9% for the tenofovir and placebo groups, respectively.

d. Clinical Assessment of Bone Abnormalities: Calcium and Fractional Excretion of Calcium

In study 902, the mean change in calcium from baseline through week 96 ranged from -0.2 mg/dL to +0.4 mg/dL. There did not appear to be a dose effect for reductions in serum calcium. Changes from baseline in fractional excretion of calcium were small and were similar for all dose groups and placebo. At week 96, the mean change ranged from -0.4% to +0.2%.

In study 907, mean change from baseline in calcium was similar at every time point through week 24 for both treatment groups. The mean change from baseline was +0.19 mg/dL and 0.17 mg/dL for the tenofovir and placebo groups, respectively.

Overall the incidence of hypocalcemia was similar between both groups. Calcium abnormalities were not clinically significant. These results are summarized in Table VD .

Table VD: Calcium Abnormalities (weeks 0-24): Studies 902 and 907

Toxicity Grade	Placebo	Tenofovir
Grade 1 (3-2.4 mg/dL)	10%	8%
Grade 2 (2.5-2.9 mg/dL)	<1%	1%
Grade 3 (2-2.4 mg/dL)	0	<1%
Grade 4 (< 2 mg/dL)	0	0

e. Clinical Assessment of Bone Abnormalities: Alkaline Phosphatase

The incidence of elevations in alkaline phosphatase was similar between groups and not thought to be clinically significant. No patients developed grade 3+ elevations in alkaline phosphatase.

f. Clinical Assessment of Bone Abnormalities: Incidence of Clinical Fractures:

The incidence of fractures was evaluated in all studies. A total of ten patients (5.5%) developed a fracture in study 902. All fractures occurred in patients receiving tenofovir and were observed with all three tenofovir doses (75, 150 and 300 mg). All fractures occurred as a result of trauma/accidental injury and no spontaneous fractures or vertebral compression fractures were documented in study 902. Time to fracture ranged from 8 to 135 weeks. Five patients developed a

fracture during the first 48 weeks of study and five patients developed a fracture after 48 weeks of tenofovir treatment. In study 907 the percentage of patients who sustained a fracture was lower, 1% (5/488), but duration of treatment was shorter in 907 compared to study 902.

The applicant cited prolonged protease inhibitor (PI) use as a possible risk factor for fractures; however, to date it is not known if PI use is related to an increased risk for fractures. Reports in the literature have suggested that HIV + patients receiving PI regimens may develop osteopenia and osteoporosis at higher rates than HIV – controls or HIV + patients receiving no treatment or non- PI regimens. However, the clinical significance of the reported loss of bone mineral in HIV + patients is unknown. To better understand these issues and to explore the frequency of fractures in other studies of antiretrovirals, FDA conducted a retrospective analysis of 13 trials to evaluate fracture rates in patients receiving PI vs Non PI containing regimens. All 13 trials in this analysis enrolled antiretroviral naï ve patients or patients with limited nRTI experience. The studies included in the analysis had an approximate mean follow up of 48 weeks. All of the approved antiretroviral agents were represented in this sample. Studies chosen for this analyses included commercial phase 3 studies that were submitted to the Division during 1999-2001 in support of accelerated or traditional approval or a dosing change. Data collected included: number of patients with clinical fractures in each treatment group; time to fracture; CD4, HIV RNA level and weight at baseline and at time of fracture; steroid use; and other risk factors. Overall in this meta-analysis of 13 studies, 2% of patients (202/10166) developed a clinical fracture. The proportion of patients who developed a fracture in the PI and Non PI group was 1.7% (97/5565) and 2.3% (105/4601), respectively. The mean time to fracture event was 311 days. The majority of fractures were a result of trauma/accidental injury.

The proportion of patients who experienced a fracture in study 902 was higher than that in seen in the meta-analysis. Due to the small sample size of study 902, this observation may in fact be an anomaly. However, further investigation of this potential safety signal is warranted.

To evaluate the potential for fractures following tenofovir treatment, overall fracture rates were calculated for study 902, study 907 and studies 902 and 907 combined. No reports of vertebral compression fractures were noted in studies 902 and 907 through 116 weeks Results for overall rates are summarized below in Table VF.

Table VF Overall Fracture Rate in Studies 902, 907 and Both Studies Combined

	Study 902		Study 907		Pooled	
	TNF n=179	Placebo n=28	TNF n=538	Placebo n=182	TNF n=717	Placebo n=210
Fractures (#)	10	0	5	3	15	3
Exposure (person-years)	311	13	488	86	799	99
Fracture Rate (95% C.I.)	3.2 (1.5 – 5.9)	0 (0 – 28.4)	1 (0.3 – 2.4)	3.5 (0.7 – 10.2)	1.9 (1.1 – 3.1)	3.0 (0.6 – 8.9)

After reviewing the entire animal toxicity and pharmacokinetic data, along with human pharmacokinetics and fracture data, it is unlikely that tenofovir-related clinical fractures will occur over 48 weeks assuming the mechanism for bone abnormalities is mediated by renal phosphate wasting or decreases in intestinal absorption of phosphate. As discussed above no significant changes in renal parameters, in particular phosphate, were observed. However, to evaluate whether risk of fractures were increasing with longer term treatment, fracture rates were calculated according to 6 month time intervals as shown in Table VG below

Table VG Number of Fractures and Fracture Rates in 6 month Intervals

Study	Study 902		Study 907		Pooled	
Treatment	TNF	Placebo	TNF	Placebo	TNF	Placebo
0-6 Months						
Fractures/patients(#)	3/179	0/28	3/538	3/182	6/717	3/210
Rate (person yr.) (95% C.I.)	3.5 (0.7 – 10.2)	0 (0 – 30.7)	1.1 (0.2 – 3.3)	3.6 (0.7 – 10.6)	1.7 (0.6 – 3.8)	3.2 (0.7 – 9.2)
6-12 Months						
Fractures/patients(#)	2/161	0/2	0/474	0/9	2/635	0/11
Rate (person yr.) (95% C.I.)	2.6 (0.3 – 9.5)	0 nd	0 (0 – 2)	0 (0 – 92.2)	0.8 (0.1 – 2.7)	0 (0 – 92.2)
12-18 Months						
Fractures/patients(#)	3/136	0/2	2/216	0/3	5/352	0/4
Rate (person yr.) (95% C.I.)	4.8 (1.0 – 14.1)	0 (0 – na)	5.3 (0.6 – 19)	0 (0 – na)	5 (1.6 – 11.7)	0 (0 – na)
18-24 Months						
Fractures/patients(#)	1/115	--	0/4	--	1/119	--
Rate (person yr.) (95% C.I.)	1.9 (0.1 – 10.7)	--	0 (0 – na)	--	1.9 (0.1 – 10.7)	--
>24 Months						
Fractures/patients(#)	1/92	--	--	--	1/92	--
Rate (person yr.) (95% C.I.)	2.9 (0.1 – 15.9)	--	--	--	2.9 (0.1 – 15.9)	--

Insufficient numbers of patients receiving prolonged tenofovir treatment and lack of a control group past 24 weeks make it difficult to conclude whether or not tenofovir use will cause clinical fractures. More patients are needed to determine if the risk of fracture increases over time. However, with the available data, no obvious increases in fracture rates were observed over time.

The division is looking forward to hearing your assessment of the preclinical and clinical data with regard to bone effects. For completeness, a summary of all fractures from studies 901, 902, 903, 907, 908 and 910 are presented in Appendix A. To date, a total of 30 bone fractures have been reported. The majority of fractures were related to trauma/accidental injury. Fractures occurred over a range of 7-135 weeks. Calcium and renal laboratory parameters were within normal limits unless otherwise stated. The majority of patients did not have any known history of hypogonadism, hyperparathyroidism, thyroid disease, corticosteroid use, malnutrition, recent immobilization or other osteoporosis risk factors. Bone mineral densities are noted for those patients participating in the 902 or 907 BMD substudies in who developed a fracture. All fractures were confirmed by radiograph unless otherwise noted. Several patients had follow up X-rays that demonstrated healing. No patient developed a subsequent fracture while continuing on tenofovir treatment.

VI. Traditional Approval: Second Confirmatory Study

In general, the Division has required two studies assessing HIV RNA for a minimum of 48 weeks duration to support traditional approval. Gilead currently is conducting one phase 3 trial (study 903) to support traditional approval. This fully enrolled study is comparing tenofovir + lamivudine + efavirenz to stavudine + lamivudine + efavirenz in treatment naïve patients. For a second confirmatory trial Gilead has proposed a study in treatment experienced children. The study design is summarized below.

Before discussing the proposed second confirmatory study which will be conducted in pediatric patients it is important to comment on the reasons for the delay in the pediatric development plan. Reports of bone loss and osteomalacia observed in animal studies were brought to the Division's attention early in tenofovir's development. As a consequence the Division and the applicant agreed that studies in pediatric patients, in which manifestations of bone toxicity could be more severe, would be delayed until these safety issues could be addressed in adults. Because of the fracture rate in study 902, it was unclear as to the amount and duration of safety data needed to support an NDA filing. During a pre NDA meeting in March of this year, the applicant provided the number of patients and duration of follow up that would be available at the time of the NDA and during the review. At this time there appeared to be a sufficient amount of safety information to submit an NDA application. It was not until the initial review of the NDA, that all the non-clinical and clinical data were presented to assess potential bone toxicity. Preliminary data suggested that fractures noted in the first 48 weeks were not likely due to tenofovir use. Therefore at this time it was thought that studies in pediatric patients with limited therapeutic options should be initiated. Intensive safety monitoring for bone abnormalities will be incorporated in all studies.

The applicant is working diligently to collect pharmacokinetic data in pediatrics. Two single center studies to assess single and multiple dose pharmacokinetics in pediatrics patients are scheduled to begin in September and October 2001. The applicant is also committed to collecting pharmacokinetic data in children who receive tenofovir on a compassionate use basis. In sum, the delay in the pediatric development is not due to the applicant's unwillingness to study tenofovir in this patient population but rather to waiting for long term follow up in adults with respect to the frequency of bone toxicity as requested by the Division.

A Phase 3, Randomized, Double-Blind, Placebo-Controlled Study of the Safety and Efficacy of Tenofovir Plus An optimized Background Regimen (OBR) Versus OBR Alone in HIV-1 infected, Antiretroviral Treatment-Experienced Children

Study Design: Two – part hybrid design (N=100 patients)

Part A: (2 weeks in duration)

Two-arm, randomized (1:1), double-blind, placebo-controlled study:

- Arm 1: Existing ART + blinded tenofovir
- Arm 2: Existing ART + tenofovir placebo

Part B: (46 weeks in duration)

Two arms remain the same, but the background ART changes to an optimized regimen while patients stay on their randomized, blinded study drug

- Arm 1: OBR + blinded tenofovir
- Arm 2: OBR + tenofovir placebo

OBR is defined as at least 3 and no more than 4 antiretroviral agents

Genotyping and virtual phenotyping at prebaseline will be done to optimize the antiretroviral background regimen in Part B of the study

Patients receiving placebo and do not have a > 0.5 log decrease from baseline by week 16 may add blinded tenofovir to their regimen. This will be managed by a third party unblinded to treatment assignment

Patient Population:

- 6 months to 17 years of age
- HIV RNA > 30,000 copies/mL
- CD4% < 20% or CD4 < 30% with documented evidence of an AIDS-defining active OI within 90 days prior to screening
- Treatment-experienced with at least one member of each class of approved agents (PI, NNRTI, NRTI)
- Stable antiretroviral treatment at least 8 weeks prior to baseline.

Statistical Methods:

- Time-weighted change from baseline in \log_{10} HIV RNA at week 2 (DAVG 2) and 48 (DAVG 48) will be compared between the two treatments using a two-sided t-test
- A sample size of 45 patients in each group will provide 86% power to detect a treatment difference in DAVG 48 of 0.4 \log_{10} copies/mL under a significance level of 0.05 and a standard deviation in DAVG 48 of 0.61

VII Draft Questions for the Committee

Listed below are the draft questions for the discussion period. Please note that these questions may change prior to the meeting.

1. In what patient population has tenofovir demonstrated efficacy and safety?
2. Please provide your assessment of the preclinical and clinical data with regard to bone effects. Please provide recommendations for additional preclinical and/or clinical studies to address the potential for bone abnormalities. Also provide recommendations for long term monitoring for this potential toxicity.
3. In follow up to the November 2-3, 1999 Advisory Committee meeting in which issues relating to the role of resistance data in antiretroviral drug development were discussed, please provide comments on the clinical resistance analyses conducted during the development of tenofovir. Also please provide recommendations for the types of clinical virology analyses that should be conducted for future antiretroviral drug development and suggestions for type of resistance data/analyses warranting display in package inserts.
4. Please provide comments on the applicant's proposed second confirmatory study for traditional approval. Please also provide comments for other study designs or patient populations that should be studied as phase 4 commitments.

Appendix A: Summary of Fractures

Study/Pt#	Sex Age Race	Fracture Site/History	Tenofovir Dose	Weeks on Tenofovir	Relevant Labs or Medical history/Comments
902-427-1568	39 Female Black	Right distal radius Fall on outstretched hand while rollerblading	150 mg	8	PI use 2+ years Hydrocortisone use 1 month Discontinued at week 28 for noncompliance
902-110-1283	37 Male White	Right clavicle, right and left tibias Motorcycle accident	150 mg	24	Episodic alcohol abuse Continue in study through 136 weeks with no additional bone fractures
902-362-1533	38 Female White	Right distal radius Fall while attempting to disembark from a moving boat	300 mg	16	PI use 2.5+ years Continued on study through 116 weeks with no additional bone fractures
902-255-1004	41 Male White	Right index finger Work related traumatic injury Fracture was not confirmed by radiograph	75 mg	39	Tobacco use (duration and amount unknown) Methylprednisolone X 5 days PI use 3 years Discontinued from study after 92 weeks due to lack of virologic response
902-427-1569	35 Male White	Left Thumb Bicycle Accident	150 mg	37	BMD: not performed Continued on tenofovir through 100 weeks with no additional bone fractures Discontinued 5/2001 due to structured treatment interruption
902-354-1723	44 Male Black	Right Femur Fall down stairs post hip replacement for avascular necrosis	300 mg	64	HIV related AVN (1998) PI use 4+ years BMD + 3.5% from baseline (spine) 2+ year use androstanolone Decreased mobility post operatively but no other risk factors* Continued on tenofovir through 127 weeks with no additional bone fractures
902-441-1485	54 Male White	Left Femur Loss of balance with fall in hospital bathroom	300 mg	56 weeks (20 weeks QOD)	Tobacco use (duration and amount unknown) Bedridden x 18 days prior to event for pneumonia and CMV encephalitis PI use 4+ years Hypogonadism BMD Jan 2000: -11.9% spine and -3.9% hip BMD April 2, 2000: -13.5% spine Discontinued when moved out

					of state in June 2000
902-427-1562	46 Male White	Left Lateral malleolus Jumped 4 ft from delivery truck onto uneven pavement	300 mg	60	Tobacco use (duration and amount unknown) Hypogonadism but no other risk factors* Continued on tenofovir through 92 weeks with no additional fractures Discontinued 8/31/00: investigators choice
902-362-1526	56 Male White	Left 10 th rib Fall in bathtub	300 mg	87	Crohn's disease (1992) Chronic diarrhea (1992) PI use 4+ years Continued on tenofovir through 132 weeks with no additional fractures
902-407-1761 (rolled over into study 910)	41 Male White	Tibia, fibula, right hip, pelvis and ribs Hit by a car on the freeway while riding a motorcycle	300 mg	87 (plus an additiona l 48 weeks on 150 mg)	Wasting syndrome Hypogonadism PI use 3+ years
907-407-3081	66 Male White	Right toe, fifth digit Toe crushed under chair leg while sitting in chair	300 mg	24	Prior traumatic fractures to same toe C4 and C5 disc herniation Hypogonadism but no other risk factors* Testosterone use 2+ years Clinically demonstrated interval healing with no follow up X-ray taken, but reinjured same toe six weeks later
907-646-3772	38 Male White	Right tibia 8 foot fall from ladder	300 mg	19	Tobacco use (1 pack/day for > 30 years) PI use 4+ years Decreased mobility due to case BMD: week 24 +2.9% change from baseline (spine) and +2.1% (hip) Week 48: -7.2% change from week 24 (spine) and -5.2% (hip) Continued on tenofovir through 30 weeks
907-692-5128	49 Male White	Left arm Fall from ladder	300 mg	10	Ex-smoker Continued on tenofovir through 64 weeks
907-645-2410 (rolled over into study 910)	30 Male White	C1 and C2 vertebrae Flipped over handlebars while mountain biking	300 mg	68	PI use Fluticasone inhaler 2 + years
907-302-2108 (rolled over into study 910)	32 Male Black	Hairline fracture of right distal tibia Fell and twisted ankle	300 mg	53	PI use 3+ years Continued on tenofovir through 65 weeks

		during contact sport activity			
907-654-2823 (rolled over into study 910)	36 Male White	Multiple left-sided rib fractures Coughing	300 mg	68	PI use 4+ years History of multiple rib fractures due to coughing Previous use of high dose IV and PO corticosteroids and continued use of inhaled corticosteroids
907-685-5335	51 Male White	Left calcaneus Fell down a flight of stairs	Placebo	NA	PI Use: 7 months
907-599-2176	48 Female Black	Left small toe Accidental injury Not confirmed radiographically	Placebo	NA	PI use 3+ years
907-881-5699	70 Male White	Right femur Fall Not confirmed radiographically	Placebo	NA	PI use 3+ years
908-774-4236	38 Male White	Left Lateral Maleolus Slipped off Curb	300 mg	30	Wasting syndrome Chronic diarrhea Malnutrition (TPN use) Hypogonadism PI use (unknown duration) Nandrolone use 1+ years Discontinued after 32 weeks of study due to progression of HIV
908-835-4350	48 Male White	Left 5 th finger Injury during basketball game	300 mg	18	Hypogonadism (5+ years) Wasting syndrome Prior broken wrist from basketball game (1981) History PI use (5+ years) History testosterone use (5+ years) Continued on tenofovir through 64 weeks
908-730-4025	66 Male White	Left femur Slipped and fell in driveway	300 mg	58	Osteonecrosis of left hip and recently underwent core hip decompression procedure to promote blood flow to left hip Hypogonadism (5+ years) PI use (5+ years) Continued on tenofovir through week 70
908-356-4240	46 Male White	Right Wrist Fall from a motorcycle accident	300 mg	54	Hypogonadism Wasting syndrome PI use 1+ year Inhaled steroids 2+ years
908-1031-4340	49 Male White	Compression Fracture L3 Fell at home secondary to orthostatic hypotension	300 mg	60	PI use 5+ years Tobacco use 20+ years Hypogonadism Wasting syndrome Malnutrition Chronic oral and inhaled

					corticosteroid use for adrenal insufficiency
908-730-4389	48 Female Hispanic	Compression fracture (T6, T8 & T10) Patient complained of scapula pain during hospitalization for pancreatitis Discharge summary notes compression fractures – MRI revealed compression fractures, which were secondary to old, not acute, trauma	300 mg	19	Long standing osteoporosis. At screening pt was wheelchair dependent due to right ankle fracture and severe debilitation Hypogonadism Wasting Syndrome Chronic Diarrhea Discontinued from study after 32 weeks due to a change in antiretroviral regimen
908-121-4409	46 Male White	Right radius Loss of balance and fell while riding moped Not confirmed radiographically	300 mg	25	Hypogonadism Wasting syndrome Hypotestosteronemia
903-731-8776	37 Male White	Right femur Fell 3-6 feet from the sidewalk while under the influence of alcohol	300 mg or d4T	24	ETOH abuse Malnutrition Tobacco use (20 cigarettes/day) Patient remains on study
903-987-8129	35 Male White	Right great toe Fell down a flight of stairs	300 mg or d4T	7	Tobacco use (1/2 pack/day for 14 years) Continues on study through 36 weeks
903-1021-8428	41 Male White	Right clavicle Hit by automobile while riding bike	300 mg or d4T	20	None Continues on tenofovir through 48 weeks
901-117-0126 (rolled over into study 910)	50 Male White	Right distal fibula Twisted right foot in a pavement hole while crossing street	300 mg	133 (includes 13 months in cohort 6 75 mg + HU)	PI use (4+ years) BMD: z scores of –1 (spine); -1.2 (hip) Continues on study through 145 weeks