7.2.5. Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog

Test Article: RPV

Species/strain: dog/Beagle	**Administration route/method:** Oral/Gavage	
Age at first dose: 8 – 8.5 months	**Doses:** 0 (vehicle), 5, 10, 40 mg/kg/day TMC278 base	**Study no.:** TMC278-NC115
Duration of dosing: 182-183 days	**Test article batch:** [redacted]	**GLP compliance:** Yes
Duration of postdose: /	**Vehicle/formulation:** PEG400 + CA	**Date of First Dose:** [redacted]

No Observed Adverse Effect Level: /

Toxicokinetics:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
No. of animals	**M:0**	**F:0**	**M:3**	**F:3**	**M:3**	**F:3**	**M:3**	**F:3**
Plasma concentration								
Day 1 (Day of 1st administration)								
C_{max} (µg/mL)			1.0	0.70	1.3	1.2	1.5	0.82
$AUC_{0\text{-}24h}$ (µg·h/mL)			11	9.2	22	20	23	10
Day 86								
C_{max} (µg/mL)			1.4	1.1	2.0	2.2	2.5	3.1
$AUC_{0\text{-}24h}$ (µg·h/mL)			21	18	28	27	41	52
Day 177								
C_{max} (µg/mL)			1.5	1.4	2.0	1.9	4.0	2.9
$AUC_{0\text{-}24h}$ (µg·h/mL)			21	17	26	32	68	43
Tissue concentration (ng/g)								
Liver								
Day 92			10885	7795	22450	15950	50650	40500
Day 183 or 184			10127	7427	13897	12247	25157	25167
Adrenal								
Day 92			5460	3250	18550	8080	35300	30150
Day 183 or 184			5950	2360	6127	4273	17673	15427

/ = not determined; F = female; M = male. PEG400: Polyethylene glycol 400; CA: citric acid.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
No. of animals	**M:6**	**F:6**	**M:6**	**F:6**	**M:6**	**F:6**	**M:6**	**F:6**
Dead or Sacrified Moribund	0	0	0	0	0	0	0	0
Clinical Observations								
Feces, soft	6	6	6	5	6	6	6	6
Salivation	-	4	-	-	-	-	-	6
Vomiting	6	6	6	6	6	6	6	6
Feces, mucous	4	5	-	-	-	-	5	4
Feces, pale	1	0	-	-	-	-	4	4
Body weight	-	-	-	-	-	-	-	-
Food consumption	-	-	-	-	-	-	-	-
Ophthalmoscopy	-	-	-	-	-	-	-	-
ECG	-	-	-	-	-	-	-	-
Hematology	-	-	-	-	-	-	-	-
Serum Chemistry[a]								
3m Interim kill (n = 6)								
Cholesterol (mg/dL)	121	158	-	-	-	-	1.397	1.228
Total bilirubin (mg/dL)	0.13	0.13	-	-	-	1.462	1.308	1.769
Alk. Phosphatase (U/L)	148	101	-	1.842	1.284	1.851	1.723	3.436
6m Terminal kill (n = 4)								
Cholesterol (mg/dL)	123	161	-	-	-	-	1.699	1.335
Total bilirubin (mg/dL)	0.16	0.12	-	1.333	-	1.667	-	1.667
Alk. Phosphatase (U/L)	224	144	-	1.576	-	1.667	2.304	5.438
Endocrinology[a]								
ACTH (pg/mL.h)								
Day 85 (n = 6)	1159	1582	-	1.139	1.601	1.881	2.323	1.409
Day 176 (n = 4)	583	1051	1.796	-	1.904	-	2.967	-
Cortisol (nmol/L.h)								
Day 85 (n = 6)	1085	1294	0..660	-	0.618	0.488	0.451	0.402
Day 176 (n = 4)	822	966	0.703	-	0.743	0.697	0.485	0.429

a For controls, group means are shown. For treated groups, multiples of control/baseline are shown; - = No noteworthy findings; F = female; M = male.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
Endocrinology (cont'd)								
Progesterone								
Day 85 **(n = 6)**	0.76	57	-	0.965	2.763	0.930	9.474	0.246
Day 176 **(n = 4)**	1.2	1.4	-	1.214	1.917	2.000	4.333	5.929
17 α-OH-progesterone								
Day 85 **(n = 6)**	0.56	-	4.643	-	10.357	-	50.000	-
Day 176 **(n = 4)**	0.51	-	4.314	-	5.294	-	21.569	-
Organ Weights[a]								
3m Interim kill **(n = 2)**								
Ovaries		1.608		1.297		1.492		1.396
6m Terminal kill **(n = 4)**								
Ovaries		1.185		-		1.322		2.030
Gross Pathology								
6m Terminal kill								
No. of animals		4		4		4		4
Ovaries: swollen		0		0		0		3
Uterus: swollen		0		0		0		2
Vagina: swollen		0		0		0		2

a = For controls, group means are shown. For treated groups, multiples of control/baseline are shown; - = No noteworthy findings; F = female; M = male.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings

Dose (mg/kg/day)	0 (vehicle)		5		10		40	
Histopathology-3m Interim Kill								
Adrenal Glands (n° examined)	2	2	2	2	2	2	2	2
Swollen cells/dense staining cytoplasm (zona reticularis)					1	1	2	2
Grade 1					1	1	1	2
Grade 2							1	
Swollen cells/dense staining cytoplasm (zona fasciculata)					1	1	1	2
Grade 1					1	1		2
Grade 2							1	
Adrenal glands (Oil red O-stain) (n° examined)	2	2	2	2	2	2	2	2
Oil red O-positive droplets (zona fasciculata)	2	2	2	2	2	2	2	2
Grade 2							1	1
Grade 3								1
Grade 4		1		2	1	1		
Grade 5	2	1	2		1	1	1	
Oil red O-positive droplets (zona reticularis)	2	2	2	2	2	2	2	2
Grade 2							1	2
Grade 3		1		2	1	1		
Grade 4	1					1		
Grade 5	1	1	2		1		1	

F = female; M = male.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	0 (vehicle)		5		10		40	
Histopathology-3m Interim Kill (cont'd)								
Epididymides (n° examined)	2		2		2		2	
Cellular debris					1		2	
Grade 1					1		2	
Liver (n° examined)	2	2	2	2	2	2	2	2
Brown pigmented macrophages							1	
Grade 1							1	
Testes (n° examined)	2		2		2		2	
Atrophic tubuli, focal							2	
Grade 1							2	
Leydig cell hyperplasia/hypertrophy					1		2	
Grade 1					1		1	
Grade 2							1	
Thymus (n° examined)	2	2	2	2	2	2	2	2
Involution	1	1	1	1	2		2	2
Grade 1	1	1	1		1		1	1
Grade 2				1	1			1
Grade 3							1	
Ovaries (n° examined)		2		2		2		2
Atretic follicles		2		2		2		2
Grade 2				1				
Grade 4		2						
Grade 5				1		2		2
Corpora lutea (active)		1		1		1		1
Grade 1						1		1
Grade 2		1		1				

F = female; M = male.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
Histopathology-3m Interim Kill (cont'd)								
Ovaries (cont'd)								
Corpora lutea (regressive)								2
Grade 2								1
Grade 5								1
Tertiary follicles		1		2		2		2
Grade 1		1		1		1		
Grade 2				1				
Grade 3						1		2
Histopathology-6m Terminal Kill								
Adrenal Gland (n° examined)	4	4	4	4	4	4	4	4
Swollen cells/dense staining cytoplasm (zona reticularis)			1	1	2	2	3	3
Grade 1			1	1	2	2	3	3
Swollen cells/dense staining cytoplasm (zona fasciculata)			1	1	2	2	3	3
Grade 1			1	1	2	2	3	3

F = female; M = male.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
Histopathology-6m Terminal Kill (cont'd)								
Adrenal glands (Oil red O-stain) (n° examined)	4	4	4	4	4	4	4	4
Oil red O-positive droplets (zona fasciculata)	4	4	4	4	4	4	4	4
Grade 2								1
Grade 3				1	2	2	3	2
Grade 4	3	1	3	2	2	2	1	1
Grade 5	1	3	1	1				
Oil red O-positive droplets (zona reticularis)	4	4	4	4	4	4	4	4
Grade 1								1
Grade 2			2	1	2	4	3	2
Grade 3	1	4	1	3	1		1	1
Grade 4	3		1		1			
Epididymides (n° examined)	4		4		4		4	
Cellular debris					2		3	
Grade 1					1		2	
Grade 2					1		1	
Reduced amount of spermatozoa					1		1	
Grade 1					1		1	
Gall bladder (n° examined)	4	4	4	4	4	4	4	4
Prominent bile/brown pigment (epithelial cells)					1		3	3
Grade 1					1		3	3
Liver (n° examined)	4	4	4	4	4	4	4	4
Brown pigmented macrophages					2		2	2
Grade 1					2		2	2

F = female; M = male.

Test Article: RPV

Report Title: TMC278-NC115: Six-Month Repeated Dose Toxicity Study with 3-Month Interim Kill in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	0 (vehicle)		5		10		40	
Histopathology 6m Terminal Kill (cont'd)								
Testes (n° examined)	4		4		4		4	
Atrophic tubuli, focal			1		2		4	
Grade 1			1		2		4	
Leydig cell hyperplasia/hypertrophy							2	
Grade 1							2	
Ovaries (n° examined)		4		4		4		4
Atretic follicles		4		4		4		4
Grade 2		1		1				
Grade 3		2		3		1		
Grade 4		1				1		2
Grade 5						2		2
Corpora lutea (regressive)		3		4		3		4
Grade 1		1		2		1		
Grade 2		2		2		2		2
Grade 3								2
Grade 5								
Tertiary follicles				2		3		2
Grade 1				2		3		2

F = female; M = male.

7.2.6. Twelve-Month Repeated Dose Oral Toxicity Study in the Dog

Species/strain: Dog/Beagle	**Administration route/method:** Oral/gavage	
Age/weight at first dose: 5 or 6 months/ 6.7 – 9.3 kg for males and 6.3-8.5 kg for females	**Doses:** 0, 5, 10 and 40 mg/kg/day TMC278 base	**Study No.:** TMC278-NC107
	Test article batch: [redacted]	**GLP compliance:** Yes
Duration of dosing: 52 weeks	**Vehicle/formulation:** PEG400 + CA	
Duration of postdose: None		

No Observed Adverse Effect Level: /

Toxicokinetics:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
No. of animals	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**
Day 1								
C_{max} (µg/mL)	-	-	0.70	0.75	0.90	1.2	2.4	2.5
$AUC_{0\text{-}24h}$ (µg·h/mL)	-	-	11	9.7	15	15	37	41
Day 90								
C_{max} (µg/mL)	-	-	1.2	1.4	2.0	2.3 [a]	3.6	5.5 [a]
$AUC_{0\text{-}24h}$ (µg·h/mL)	-	-	18	21	29	29 [a]	60	88 [a]
Day 273								
C_{max} (µg/mL)	-	-	1.1	1.1	1.3 [a]	2.3	3.2	3.5 [a]
$AUC_{0\text{-}24h}$ (µg·h/mL)	-	-	16	18	19[a]	31	60	51 [a]
Day 363								
C_{max} (µg/mL)	-	-	1.1	1.5	1.3 [b]	2.2	4.1	5.5 [a]
$AUC_{0\text{-}24h}$ (µg·h/mL)	-	-	17	19	24 [b]	36	65	61 [a]

a : n = 3; b : n = 2; / = not determined;.

Test Article: RPV

Report Title: TMC278-NC107: Twelve-Month Repeated Dose Oral Toxicity Study in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
No. of animals	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**
Died or killed prematurely	0	0	0	0	0	0	0	0
Clinical observations								
Salivation	0	0	1	0	0	1	2	0
Feces liquid	3	4	4	4	4	4	4	4
Body weight gain (kg)[c]	2.2	3.6	-	-	-	-	0.591	0.444**
Food consumption (g)	-	-	-	-	-	-	-	-
Ophthalmoscopy	-	-	-	-	-	-	-	-
ECG	-	-	-	-	-	-	-	-

c For controls, group means are shown. For treated groups, multiples of control/baseline are shown. * - $p<0.05$, ** - $p<0.01$, *** - $p<0.001$. ECG: electrocardiogram; F: female; M: male; - no noteworthy changes. PEG400: polyethylene glycol; CA: citric acid.

Test Article: RPV

Report Title: TMC278-NC107: Twelve-Month Repeated Dose Oral Toxicity Study in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
No. of animals	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**
Hematology (Week 52)[a]								
Red blood cells (M/mm^3)	6.96	-	-	-	-	-	0.889*	-
Hemoglobin (g/dL)	16.1	-	-	-	-	-	0.869*	-
Hematocrit (%)	47.3	-	-	-	-	-	0.886*	-
Serum chemistry (Week 52)[a]								
Calcium (mg/L)	101	106	-	-	-	-	0.901*	0.934*
Inorganic phosphorus (mg/L)	29	-	-	-	-	-	1.172	-
Creatinine (mg/L)	-	7.2	-	-	-	-	-	1.181
Alkaline phosphatase (IU/L)	220	288	-	-	-	-	2.877	1.549
Urinalysis (Week 52)[a]								
Volume (mL)	65	-	-	-	-	-	3.51	-
Endocrinology [b] (Week 52)								
Progesterone (nmol/L)	0.00	-	1.8	-	3.7	-	7.5	-
17α-OH-progesterone (nmol/L)	1.40	-	6.142	-	8.429	-	14.43	-
Estradiol (pmol/L)	-	-	-	-	-	-	-	-
Cortisol (nmol/L)	-	27	-	-	-	0.259	-	0.111
ACTH (pg/mL/h)	-	-	-	-	-	-	-	-
Luteinized hormone (ng/mL)	-	-	-	-	-	-	-	-
Testosterone (nmol/L)	-	-	-	-	-	-	-	-
Organ weights								
Adrenal glands (g)	1.24	1.29	-	-	--	1.233	1.234	1.233
Ovaries (g)	-	1.92	-	-	-	1.396	-	2.193
Gross pathology	-	-	-	-	-	-	-	-

a = For controls, group means are shown. For treated groups, multiples of control/baseline are shown.; b Actual group means are shown as division by zero gives infinity;
* - p<0.05, ** - p<0.01, ***. F: female; M: male; - no noteworthy changes

Test Article: RPV

Report Title: TMC278-NC107: Twelve-Month Repeated Dose Oral Toxicity Study in the Dog (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**		**5**		**10**		**40**	
No. of animals	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**	**M:4**	**F:4**
Histopathology								
Adrenal glands								
Cytoplasmic density (zona fasciculate/reticularis)	0	0	0	4	1	4	3	4
Pigment deposit in the cortex	0	0	0	0	0	0	2	2
Ovaries								
Antral follicles	-	4	-	4	-	4	-	4
Prominent early luteinized follicles	-	0	-	0	-	0	-	2
Prominent corpora lutea	-	2	-	3	-	3	-	3
Liver								
Yellow pigmentation	0	0	0	0	0	1	1	3
Kidney								
Acute interstitial nephritis	0	0	0	0	0	0	2	0
Corticomedullary mineralization	0	0	0	0	0	0	0	3
Testes								
Hypertrophy of the Leydig cells	0	-	0	-	0	-	2	-

F: female; M: male; - no noteworthy changes

7.2.7. Endocrinological Oral (Gavage) 8-week Study in the Female Sexually Immature Cynomolgus Monkey

Test Article: RPV

Species/Strain: Monkey/cynomolgus	**Administration route/method:** Oral/Gavage	
Initial Age/weight: 1-2 years/1.5-2.0 kg	**Doses:** 0 (vehicle), 100 , 250 mg base eq./kg/day TMC278.HCl twice daily	**Study No.:** TMC278-NC248
Duration of dosing: 8 weeks	**Test article batch:** [redacted]	**GLP compliance:** Yes
	Vehicle/formulation: 1% HPMC + 0.5% Tween 20	**Date start of study:** [redacted]
Special features: Hormone analysis		
No Observed Adverse Effect Level: /		
Toxicokinetics:		

Dose (mg base eq./kg/day)	**0 (vehicle)** [a]	**200**	**500**
No. of animals	**F:8**	**F:8**	**F:8**
Day 55			
$C_{max\ 1}$ (µg/mL)	/	0.14	0.31
$C_{max\ 2}$ (µg/mL)		0.18	0.31
$AUC_{0\text{-}24h}$ (µg·h/mL)	/	2.7	4.6

Test Article: RPV

Report Title: TMC278-NC248: Endocrinological Oral (Gavage) 8-week Study in the Female Sexually Immature Cynomolgus Monkey (continued)

Noteworthy Findings			
Dose (mg base eq./kg/day)	**0**	**200**	**500**
N° of animals	**F:8**	**F:8**	**F:8**
Died or killed prematurely	**0**	**0**	**0**
Clinical observations	**-**	**-**	**-**
Body weights (kg)	**-**	**-**	**-**
Menses	**-**	**-**	**-**
ACTH (pmol/L)	-	-	-
Cortisol (nmol/L)	-	-	-
17-hydroxyprogesterone (nmol/L) D 15	7.57	11.7	13.0
17-hydroxyprogesterone (nmol/L) D 29	7.97	17.0	18.2
17-hydroxyprogesterone (nmol/L) D 43	6.49	16.1	14.9
Progesterone (nmol/L) D15	10.6	11.3	14.5
Progesterone (nmol/L) D29	9.65	14.3	16.2
Progesterone (nmol/L) D43	8.12	13.4**	14.3**

[a] In the control-dosed group, plasma concentrations (26 out of 80) were above the limit of quantification (0.001 µg/mL) and ranged from 0.001 to 0.003 µg/mL.

** $p \leq 0.01$ (Analysis of variance); D: day; / = not determined; - = no noteworthy findings; HPMC: hydroxypropylmethylcellulose; F: female

Test Article: RPV

Report Title: TMC278-NC248: Endocrinological Oral (Gavage) 8-week Study in the Female Sexually Immature Cynomolgus Monkey (continued)

Noteworthy Findings:

Dose (mg base eq./kg/day)	**0**	**200**	**500**
N° of animals	**F:8**	**F:8**	**F:8**
Androstenedione (nmol/L) D15	2.64	1.94*	1.81*
Androstenedione (nmol/L) D29	2.10	1.66	1.53
Androstenedione (nmol/L) D43	2.01	1.44	1.27
DHEA (nmol/L) D15	245	264	196
DHEA (nmol/L) D29	289	296	225
DHEA (nmol/L) D43	257	270	199
Estradiol	-	-	-
LH	-	-	-
Hematology	-	-	-
Clinical chemistry	-	-	-
Urine analysis	-	-	-
Organ weights	-	-	-
Macroscopic	-	-	-
Histopathology			
Thyroid			
Follicular cell hypertrophy	1	3	4

* $p \leq 0.05$ (Analysis of variance); D: day; - no noteworthy findings; F: female

7.3. TAF

7.3.1. 13-Week Oral Gavage Toxicity and Toxicokinetic Study in Mice

Test Article: TAF

Report Title: 13-Week Oral Gavage Toxicity and Toxicokinetic Study with GS-7340-02 in Mice		**Gilead Study No.** TX-120-2007 [redacted] **Study No. 8241074**
Species / Strain: Mouse/Crl:CD1(ICR)	**Duration of Dosing:** 13 weeks	
Initial Age: 5 to 6 weeks old	**Duration of Postdose:** NA	**GLP Compliance:** yes
Date of First Dose: [redacted]	**Method of Administration:** Oral Gavage	**Lot Number:** [redacted]
Vehicle: 0.1% (v/v) Tween 20 and 0.1% (w/v) HPMC K100LV prepared in reverse osmosis water		
Special Features: Insufficient GS-7340 data for TK analysis; Tenofovir TK analyses presented		

No Observed Adverse Effect Level: Not determined

Daily Dose (mg/kg/day)	**0 (Control)**		**10**		**30**		**100**	
Gender: Number of Animals (TK)	**M: 6**	**F: 6**	**M: 42**	**F: 42**	**M: 42**	**F: 42**	**M: 42**	**F: 42**
Toxicokinetics (Tenofovir): $AUC_{(0-t)}$ (ng·hr/mL)								
Day 1	NA[a]	NA[a]	148	193	1140	1424	5829	7239
Week 13	NA[a]	NA[a]	191	239	1561	1453	7797	6545
Toxicokinetics (Tenofovir): C_{max} (ng/mL)								
Day 1	NA[a]	NA[a]	62.3	57.6	248	337	1024	999
Week 13	NA[a]	NA[a]	58.8	99.6	422	280	958	767

Test Article: TAF

Report Title: 13-Week Oral Gavage Toxicity and Toxicokinetic Study with GS-7340-02 in Mice	**Gilead Study No. TX-120-2007** [redacted] **Study No. 8241074**

Daily Dose (mg/kg/day)	**0 (Control)**		**10**		**30**		**100**	
Gender: Number of Animals (Main)	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**
Noteworthy Findings								
Died or sacrificed moribund	-	-	-	-	-	-	1[b]	-
Body Weight Change								
Dosing Days 1 – 92 (g)	9.3	8.0	9.4	7.1	9.3	6.1	7.9	7.3
Food Consumption (%)[c]								
Dosing Week 1	41.6 g	36.7 g	+3.4	-0.8	-13.9*	-12.8	-10.8*	-12.5*
Dosing Week 2	31.1 g	28.9 g	+2.9	+0.3	+11.6*	+8.3	+14.1*	+13.1*
Clinical Observations	-	-	-	-	-	-	-	-
Ophthalmoscopy	-	-	-	-	-	-	-	-
Hematology	-	-	-	-	-	-	-	-
Serum Chemistry	-	-	-	-	-	-	-	-
Dosing Phase – Final Phase Sacrifice								
Number Evaluated	15	15	15	15	15	15	14	15
Organ Weights	-	-	-	-	-	-	-	-

Test Article: TAF

Report Title: 13-Week Oral Gavage Toxicity and Toxicokinetic Study with GS-7340-02 in Mice	**Gilead Study No. TX-120-2007** [redacted] **Study No. 8241074**

Daily Dose (mg/kg/day)	**0 (Control)**		**10**		**30**		**100**	
Gender: Number of Animals (Main)	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**
Dosing Phase – Final Phase Sacrifice (cont.)								
Number Evaluated	15	15	15	15	15	15	15	15
Macroscopic Observations	-	-	-	-	-	-	-	-
Histopathology								
Nasal turbinates								
Infiltrate, neutrophil, respiratory mucosa								
Minimal	1	1	3	3	4	6	6	10
Slight	0	0	2	0	2	0	5	4
Infiltrate, neutrophil, olfactory mucosa								
Minimal	1	1	7	4	7	4	7	11
Slight	0	0	2	0	1	0	7	2
Degeneration, olfactory epithelium								
Minimal	2	2	5	4	5	7	3	8
Slight	0	0	3	0	2	0	8	4
Moderate	0	0	0	0	1	0	2	1

Test Article: TAF

Report Title: 13-Week Oral Gavage Toxicity and Toxicokinetic Study with GS-7340-02 in Mice	**Gilead Study No. TX-120-2007** [redacted] **Study No. 8241074**

Daily Dose (mg/kg/day)	**0 (Control)**		**10**		**30**		**100**	
Gender: Number of Animals (Main)	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**
Dosing Phase – Final Phase Sacrifice (cont.)								
Exudate, lumen								
Minimal	0	0	0	0	2	2	2	2
Rectum								
Apoptosis, increased								
Minimal	0	0	0	0	0	0	5	2

a Values below the lower limit of quantitation (with the exception of Animal No. A54618 (Group 5 female) for tenofovir on Day 1 of the dosing phase)

b On Day 54 of the dosing phase Animal No. A54419 (Group 4 male) was sacrificed in moribund condition due to gavage-related injury. Respectively on Days 15 and 45 of the dosing phase, Animal Nos. A54647 (Group 6 TK female) and A54746 (Group 8 TK female) were found dead. These deaths were not considered test article-related.

c For controls, group means are shown. For treated groups, percent differences or fold change from controls are shown. Statistical significance is based on actual data not on the percent.

M = Male; F = Female; TK = toxicokinetic; NA = Not Applicable

− Comparable to control animals or no treatment-related findings; * = $p \leq 0.05$ (ANOVA and Dunnett's t-test where applicable)

7.3.2. A 28-Day Oral Toxicity Study in the Albino Rat

Test Article: TAF

Report Title: A 28-Day Oral Toxicity Study of GS-7340-02 in the Albino Rat		**Gilead Study No.** R990182 [redacted] **Study No. 89176**
Species / Strain: Sprague-Dawley rat	**Duration of Dosing:** 28 days	
Initial Age: 10 weeks old	**Duration of Postdose:** None	**GLP Compliance:** yes
Date of First Dose: [redacted] 19[redacted]	**Method of Administration:** Oral gavage	**Lot Number:** [redacted]
Vehicle: 50 mM Citric Acid		
Special Features: Biochemical markers of bone turnover , Peripheral Quantitative Computed Tomography (pQCT)		
No Observed Adverse Effect Level: 6.25 mg/kg/day		

Daily Dose (mg/kg/day)	**0 (Control)**		**1.5**		**6.25**		**25**		**100**		**400**	
Gender: Number of Animals (Main)	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**
Toxicokinetics (Tenofovir): AUC (hr·µg/mL)												
Day 1	NA[a]	NA[a]	45.5	49.2	288	296	2430	2770	12700	13600	61800	78400
Day 28	NA[a]	NA[a]	52.7	55.1	358	321	2450	2820	12200	14600	62700	75100
Toxicokinetics (Tenofovir): C_{max} (µg/mL)												
Day 1	NA[a]	NA[a]	35.5	35.6	167	229	1340	1970	6970	6990	12900	20100
Day 28	NA[a]	NA[a]	49.7	52.8	169	211	1300	1690	4780	6160	10300	14400

Test Article: TAF

Report Title: A 28-Day Oral Toxicity Study of GS-7340-02 in the Albino Rat	**Gilead Study No.** R990182 [redacted] **Study No. 89176**

Daily Dose (mg/kg/day)	**0 (Control)**		**1.5**		**6.25**		**25**		**100**		**400**	
Gender: Number of Animals (Main)	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**
Noteworthy Findings												
Died or sacrificed moribund	-	-	-	-	1	-	-	-	-	-	2	-
Body Weight Gain (%)[b]												
Week 1 to 2	49.9	30.9	-	-	-	-	-29% A	-35% B	-37% B	-24% A	-67% B	-56% B
Week 2 to 3	20.4	9.8	-	-	-	-	-	-	-	-	-68% B	-
Week 3 to 4	5.5	1.8	-	+49 % B	-	-	-	-	-	-	-	-
Food Consumption (%)[b]												
Week 1 to 2	209.2	162.0	-	-	-	-	-	-	-	-	-17% B	-14% B
Week 2 to 3	203.4	159.5	-	-	-	-	-	-	-	-	-18% B	-14% A
Week 3 to 4	195.3	154.6	-	-	-	-	-	-	-	-	-14% B	-11% A

Test Article: TAF

Report Title: A 28-Day Oral Toxicity Study of GS-7340-02 in the Albino Rat	**Gilead Study No.** R990182 [redacted] **Study No. 89176**

Daily Dose (mg/kg/day)	**0 (Control)**		**1.5**		**6.25**		**25**		**100**		**400**	
Gender: Number of Animals (Main)	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**
Clinical Observations												
Fur, staining, red, muzzle	1	1	1	-	1	-	1	-	2	-	4	2
Hematology[b]												
White blood cell ($x10^3/mm^3$)	7.9	-	-	-	-	-	-	-	-	-	-57% B	-
Monocytes (n/mm^3)	28 6.9	-	- 53% A	-	- 100 %	-	-98% B	-	-91% B	-	-93% A	-
Lymphocytes (n/mm^3)	65 90. 3	-	-	-	-	-	-	-	-	-	-57% B	-
Red blood cell ($x10^6/mm^3$)	7.7 1	7.06	-	-	-	-	-	-	-	-	-11% B	- 12% B
Hemoglobin (g/dL)	14. 9	14.1	-	-	-	-	-	-	-	-	-6% B	-7% B
Hematocrit (%)	42. 4	39.7	-	-	-	-	-	-	-	-	-8% B	-8% B
Mean Corpuscular hemoglobin (pg)	19. 3	20.0	-	-	-	-	-	-	-	-	+6% B	+6% A
Red blood cell distribution width (%)	12. 2	11.7	-	-	-	-	-	-	-	-	+20% B	+15 % B
Clinical Chemistry[b]												
Urea nitrogen (mg/dL)	16. 3		-	-	-	-	-	-	-	-	+28% B	-
Alkaline phosphatase (U/L)	11 0.3	71.0	-	-	-	-	-	-	-	-	-37% B	- 44% B
Cholesterol (mg/dL)	54. 8	70.3	-	-	-	-	-	-	-	-	+71% B	+30 % A

Test Article: TAF

Report Title: A 28-Day Oral Toxicity Study of GS-7340-02 in the Albino Rat	**Gilead Study No.** R990182 [redacted] **Study No. 89176**

Daily Dose (mg/kg/day)	**0 (Control)**		**1.5**		**6.25**		**25**		**100**		**400**	
Gender: Number of Animals (Main)	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**
Organ Weights[b]												
Relative liver weight (%)	-	2.630	-	-	-	-	-	-	-	-	-	+11% B
Relative thymus weight (%)	0.071	-	-	-	-	-	-	-	-	-	- 61% B	-
Gross Pathology												
Thymus - Number Evaluated	10	10	10	10	10	10	10	10	10	10	10	10
Discoloration dark	-	-	-	-	-	-	-	-	-	-	1	-
Small	-	-	-	-	-	-	-	-	-	-	7	1
Histopathology												
Femur - Number Evaluated	10	10	-	-	-	-	-	-	-	-	10	10
Atrophy – cancellous bone	-	-	-	-	-	-	-	-	-	-	4	9
Kidney – Number evaluated	10	10	10	10	10	10	10	10	10	10	10	10
Tubular basophilia	1	-	1	-	1	-	-	-	-		8	3
Tubular karyomegaly	-	-	-	-	-	-	-	-	-	-	7	5
Hyaline droplets	1	-	-	-	2	-	3	-	6	-	2	-
Thymus – Number examined	10	10	10	10	10	10	10	10	10	10	10	10
Atrophy	-	-	-	-	1	-	-	-	-	-	9	6

Test Article: TAF

Report Title: A 28-Day Oral Toxicity Study of GS-7340-02 in the Albino Rat	**Gilead Study No.** R990182 [redacted] **Study No. 89176**

Daily Dose (mg/kg/day)	**0 (Control)**		**1.5**		**6.25**		**25**		**100**		**400**	
Gender: Number of Animals (Main)	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**	**M: 10**	**F: 10**
Bone parameters[b]												
Periosteal circumference (mm)	16.09	13.44	-	-	-	-	-	-	-	-	-4%	-1.5%
Total mean slice area – distal femur (mm^2)	20.65	14.41	-	-	-	-	-	-	-	-	-7%	-3%
Bone mineral density (mg/cm^3)	576.5	752.3	-	-	-	-	-	-	-	-	-7%	-14% B
1.25-Dihydroxy Vitamin D (pg/mL)	51.67		-		-		-		-51% A		-74% A	

a Values below the lower limit of quantitation
b For controls, group means are shown. For treated groups, percent differences from controls are shown. Statistical significance is based on actual data not on the percent.
M = Male; F = Female, NA = Not applicable,
− Comparable to control animals or no treatment-related findings, Significantly different from control (group 1) value: A - $p<0.05$, B - $p<0.01$ (Dunnett's)

7.3.3. A 26-Week Oral Gavage Toxicity Study of GS-7340-02 in the Albino Rat

Test Article: TAF

Report Title: A 26-Week Oral Gavage Toxicity Study of GS-7340-02 in the Albino Rat		**Gilead Study No.** TOX-120-001 [redacted] **Study No. 89686**
Species / Strain: Sprague-Dawley rats (Crl: CD® (SD) BR) / *Rattus norvegicus*	**Duration of Dosing:** 26 weeks	
Initial Age: 6 weeks at start of dosing	**Duration of Postdose:** N/A	**GLP Compliance:** yes
Date of First Dose: [redacted]	**Method of Administration:** Oral gavage	**Lot Number:** [redacted]
Vehicle: 0.5% polysorbate 20, NF; 0.5% carboxymethylcellulose; 0.9% Benzyl Alcohol		
Special Features: Bone parameters		
No Observed Adverse Effect Level: 25 mg/kg/day		

Daily Dose (mg/kg/day)	**0 (Control)**		**5**		**25**		**100**	
Gender: Number of Animals (TK)	**M: 0**	**F: 0**	**M: 8**	**F: 8**	**M: 8**	**F: 8**	**M: 8**	**F: 8**
Toxicokinetics (Tenofovir): AUC (hr·µg/mL)								
Day 1	NA[a]	NA[a]	0.625	0.584	3.024	3.704	12.555	12.275
Week 13	NA[a]	NA[a]	0.798	0.656	3.639	3.809	15.844	14.245
Week 26	NA[a]	NA[a]	0.739	0.601	3.526	3.991	15.931	15.138
Toxicokinetics (Tenofovir): C_{max} (µg/mL)								
Day 1	NA[a]	NA[a]	0.326	0.292	1.214	1.354	5.071	4.817
Week 13	NA[a]	NA[a]	0.337	0.351	1.335	1.593	4.766	6.262
Week 26	NA[a]	NA[a]	0.282	0.252	1.249	1.798	4.499	5.323

Test Article: TAF

Report Title: A 26-Week Oral Gavage Toxicity Study of GS-7340-02 in the Albino Rat					**Gilead Study No.** TOX-120-001 [redacted] **Study No. 89686**			
Daily Dose: GS-7340-02 (mg/kg/day)	**0 (Control)**		**5**		**25**		**100**	
Gender: Number of Animals (Main)	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**
Died or sacrificed moribund[d]	-	1	-	2	-	-	-	3
Body Weights	-	-	-	-	-	-	-	-
Food Consumption	-	-	-	-	-	-	-	-
Clinical Observations	-	-	-	-	-	-	-	-
Ophthalmology	-	-	-	-	-	-	-	-
Hematology	-	-	-	-	-	-	-	-
Clinical Chemistry	-	-	-	-	-	-	-	-
Urinalysis	-	-	-	-	-	-	-	-
Organ Weights	-	-	-	-	-	-	-	-
Macroscopic Observations	-	-	-	-	-	-	-	-
Noteworthy Findings								
Histopathology								
Bone – Tibia								
Number Evaluated	15	14	-	13	-	15	15	13
Atrophy: cancellous bone	-	-	-	-	-	-	-	8
Kidney								
Number Evaluated	15	15	15	15	15	15	15	15
Basophilia: tubular	6	4	4	4	6	3	7	3
Karyomegaly: tubular	-	-	-	-	-	-	11	2

Test Article: TAF

Report Title: A 26-Week Oral Gavage Toxicity Study of GS-7340-02 in the Albino Rat					**Gilead Study No.** TOX-120-001 [redacted] **Study No. 89686**			
Daily Dose: GS-7340-02 (mg/kg/day)	**0 (Control)**		**5**		**25**		**100**	
Gender: Number of Animals (Main)	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**
Degeneration: tubular	-	-	-	-	1	-	-	-
Hyaline droplet: tubular	8	-	3	-	4	-	11	-
Pyelitis/pyelonephritis	-	1	-	4	-	1	1	1
Dilatation: pelvis	-	1	1	1	1	1	-	-
Cyst	-	1	-	1	1	1	-	-
Fibrosis	-	-	-	-	1	-	-	1
Bone Densitometry (pQCT)[b]								
Right proximal tibia - metaphysis								
Trabecular bone mineral density (BMD), Week 26 (mg/cm^3)	221.9	280.79	-2.1%	-9.3%	-2.3%	-2.3%	-18.4%	-21% A
Trabecular bone mineral content (BMC), Week 26 (mg/mm)	2.26	0.83	-6.2%	-12%	-4.4%	-2.4%	-22.6%	-33.7%
Right distal femur - metaphysis								
Trabecular (BMD), ex-vivo (mg/cm^3)	259.8	301.2	c	c	c	c	-15.4%	-5.3%
Trabecular (BMC), ex-vivo (mg/mm)	3.5	1.9	c	c	c	c	-17.1%	-5.3%
Urine Biochemical markers of bone turnover, Week 13[b]								
C-Telopeptide (µg/mmol creatinine)	3.673	-	-		+78% D	-	+135% F	-

Test Article: TAF

Report Title: A 26-Week Oral Gavage Toxicity Study of GS-7340-02 in the Albino Rat					**Gilead Study No.** TOX-120-001 [redacted] **Study No. 89686**			
Daily Dose: GS-7340-02 (mg/kg/day)	**0 (Control)**		**5**		**25**		**100**	
Gender: Number of Animals (Main)	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**	**M: 15**	**F: 15**
Deoxypyridinoline (nM/mM creatinine)	80.603	-	-	-	-	-	+25% A	-
C-Telopeptide (µg/mmol creatinine)	3.673	-	+55%	-	+78% D	-	+135% F	-
Serum Biochemical markers of bone turnover and hormones, Week 26 [b]								
1,25-Dihydroxy Vitamin D (pmol/L)	-	105.021	-	-10%	-	-45%	-	-59% F
25-Hydroxy Vitamin D (ng/mL)	-	22.358	-	+14%	-	-15%	-	-22% B

a Values below the lower limit of quantitation
b For controls, group means are shown. For treated groups, percent differences from controls are shown. Statistical significance is based on actual data not on the percent.
c Bone densitometry (pQCT) was only evaluated in the right distal femur (metaphysis) of animals in the control and 100 mg/kg/day groups.
d There were no unscheduled deaths recorded during the 26-week treatment period which were considered to treatment related. Two high dose female TK animals were euthanized on days 732 and 164; in both cases the deterioration of their condition could not be attributed to an accident during dose administration.
e $AUC_{0-\infty}$ on Day 1 and $AUC_{0-\tau}$ in Weeks 13 and 26
M = Male; F = Female; TK = toxicokinetic; NA = Not Applicable; − Comparable to control animals or no treatment-related findings; pQCT = peripheral quantitative computed tomography
A = $p \leq 0.05$; B = $p \leq 0.01$; C = $p \leq 0.001$ (Dunnett); D = $p \leq 0.05$; E = $p \leq 0.01$; F = $p \leq 0.001$ (Dunn's)

7.3.4. A 28-Day Oral Gavage Toxicity Study in the Beagle Dog

Test Article: TAF

Report Title: A 28-Day Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog		**Gilead Study No.** D990175 [redacted] **Study No. 89128**
Species / Strain: Beagle dog	**Duration of Dosing:** 28 days	
Initial Age: 7 to 8 months at dosing	**Duration of Postdose:** N/A	**GLP Compliance:** yes
Date of First Dose: [redacted] 19[redacted]	**Method of Administration:** Oral gavage	**Lot Number:** [redacted]
Vehicle: Citric Acid (formulated in Sterile Water for Irrigation, USP)		
Special Features: bone parameters		
No Observed Effect Level: 1 mg/kg/day		

Daily Dose (mg/kg/day)	**0 (Control)**		**0.1**		**0.3**		**1**		**3**		**10**	
Gender: Number of Animals	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**
Toxicokinetics (Tenofovir): AUC (hr·µg/mL)												
Day 1	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	NC	
Day 28	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	5.26 $_{(0-\tau)}$	
Toxicokinetics (Tenofovir): C_{max} (µg/mL)												
Day 1	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	0.385	
Day 28	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	0.444	
Toxicokinetics (GS-7340) : AUC (hr·µg/mL)												
Day 1	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	NC	
Day 28	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	0.985 $_{(0-\tau)}$	

Test Article: TAF

Report Title: A 28-Day Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog									**Gilead Study No.** D990175 [redacted] **Study No. 89128**			
Daily Dose (mg/kg/day)	**0 (Control)**		**0.1**		**0.3**		**1**		**3**		**10**	
Gender: Number of Animals	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**
Toxicokinetics (GS-7340): C_{max} (µg/mL)												
Day 1	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	0.582	
Day 28	NA[a]	NA[a]	NC	NC	NC	NC	NC	NC	NC	NC	1.28	
Noteworthy Findings												
Died or sacrificed moribund, adverse findings etc.	There were no unscheduled deaths or adverse findings for body weight, food consumption, clinical observations, ophthalmology examinations or hematology parameters.											
Serum Chemistry												
AST (U/L)	38.3	39.8	37.0	37.5	43.0	34.5	34.0	34.3	38.3	46.0	42.8	46.8
Day 28	34.8	35.3	33.8	36.0	39.8	33.8	34.5	35.8	37.5	36.0	47.0	52.5*
Urinalysis	-	-	-	-	-	-	-	-	-	-	-	-
Organ Weights	-	-	-	-	-	-	-	-	-	-	-	-
Gross Pathology												
Number Evaluated	4	4	4	4	4	4	4	4	4	4	4	4
Cecum, Area dark	1	1	-	1	-	-	1	1	2	-	2	-
Colon, Area dark	-	-	-	-	-	-	-	1	-	-	2	1
Duodenum, Area dark	-	-	-	-	-	-	-	-	-	-	1	-
Rectum , Area dark	-	-	-	-	-	-	-	-	-	-	-	2
Lung , Area depressed	-	-	-	-	1	-	-	-	-	1	-	-

Test Article: TAF

Report Title: A 28-Day Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog											**Gilead Study No.** D990175 [redacted] **Study No. 89128**	
Daily Dose (mg/kg/day)	**0 (Control)**		**0.1**		**0.3**		**1**		**3**		**10**	
Gender: Number of Animals	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**	**M: 4**	**F: 4**
Area dark	-	-	-	-	-	-	-	1	-	-	-	-
Area pale	-	-	-	-	-	-	1	-	1	1	-	-
Area raised	2	1	1	2	-	1	1	-	1	1	-	1
Discoloration pale	-	-	-	-	-	-	-	-	1	-	-	-
Spleen, Area dark	-	-	-	-	-	-	1	-	-	-	-	-
Area depressed	1	1	1	-	1	1	-	-	-	-	-	1
Area pale	1	-	1	1	-	-	-	-	-	-	1	-
Area raised	-	-	-	-	-	-	-	-	-	-	1	-
Adhesion	-	-	-	-	-	-	-	-	-	-	-	1
Histopathology												
Kidney												
Karyomegaly: tubular	-	-	-	-	-	-	-	-	1	1	4	4
Basophilia: tubular	-	-	-	-	-	-	-	-	-	-	2	3
Bone Densitometry (pQCT)	-	-	-	-	-	-	-	-	-	-	-	-

M = Male; F = Female; TK = toxicokinetic; NA = Not Applicable; NC = Not calculated (insufficient data); − Comparable to control animals or no treatment-related findings,
* = $p \leq 0.01$ (Dunnett); pQCT = peripheral quantitative computed tomography
a not assayed

7.3.5. A 9 Month Oral Gavage Toxicity Study in the Beagle Dog with a 3 Month Recovery Period for Selected Animals

Test Item: TAF

Report Title: A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals		**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**
Species / Strain: beagle dog	**Duration of Dosing:** 9 months	
Initial Age: 5-6 months	**Duration of Postdose:** 3 months	**GLP Compliance:** yes
Date of First Dose: [redacted]	**Method of Administration:** oral gavage	**Lot Number:** [redacted]
Vehicle: 0.5% polysorbate 20, NF; 0.5% carboxymethylcellulose; 0.9% Benzyl Alcohol		
Special Features: Bone densitometry, biochemical markers of bone turnover and associated parameters		
No Observed Adverse Effect Level: 2 mg/kg/day		

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12** [a]	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 8**	**F: 8**
Toxicokinetics (GS-7340): AUC_{0-t} (hr·µg/mL)								
Day 1	NA [b]		0.03		0.37		2.07	
Week 13	NA [b]		0.07		0.66		2.23	
Week 39/40	NA [b]		0.08		1.03		1.95	
Toxicokinetics (GS-7340): C_{max} (µg/mL)								
Day 1	NA [b]		0.08		0.72		3.60	
Week 13	NA [b]		0.13		1.10		3.24	
Week 39/40	NA [b]		0.14		1.42		2.62	

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals				**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**			
Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12** [a]	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 8**	**F: 8**
Toxicokinetics (Tenofovir): AUC_{0-24} (hr·µg/mL)								
Day 1	NA [b]		0.40		1.31		3.80	
Week 13	NA [b]		1.21		4.10		12.48	
Week 39/40	NA [b]		1.18		4.45		13.73	
Toxicokinetics (Tenofovir): C_{max} (µg/mL)								
Day 1	NA [b]		0.08		0.25		0.80	
Week 13	NA [b]		0.16		0.56		1.25	
Week 39/40	NA [b]		0.18		0.54		1.32	
Noteworthy Findings								
Died or sacrificed moribund[d]	-	-	-	-	-	-	2	-
Body Weight (%) [c]								
Week 13	9.95 kg	-	-	-	-	-	-22.4 [B]	-
Week 26	10.95 kg	-	-	-	-	-	-23.1 [B]	-
Week 39	11.50 kg	8.88	-6.7	-	-13.2	-	-25.7 [C]	-11.9

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals				**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**			
Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12 [a]**	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 8**	**F: 8**
Food Consumption (%) [c]								
Week 13	256.72	-	-	-	-	-	-23.8	-
Week 26	319.40	-	-	-	-	-	-21.7	-
Week 39	329.48	-	-	-	-	-	-19.8	-
Clinical Observations	An increased incidence of salivation was recorded both pre-dose and following dose administration for animals from all treated groups including the controls. In addition, a red/brown fur staining was also observed on the paws of the treated animals with a higher incidence observed in animals administered 18/12 mg/kg/day. This change was most often distributed on the lower extremities, the hind paws and the forepaws, and was still apparent to a lesser degree during the three month recovery period.							
Ophthalmoscopy	-	-	-	-	-	-	-	-

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals				**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**			
Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12 [a]**	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 8**	**F: 8**
Electrocardiography [c]								
Heart Rate (beats/min)								
Week 13	117.0	106.0	-	-	-	-	-32.5% [B]	-29.2% [B]
Week 39	111.0	-	-	-	-	-	-49.5% [C]	-
PR (msec)								
Week 13	-	86.7	-	-	-	-	-	+26.2 % [B]
Week 39	83.8	87.5	-	-	+28.3 % [B]	+25.7 %	+31.3% [B]	+41.9 % [D]
QT								
Week 13	189.2	200.8	-	-	-	-	+17.2% [B]	+10.2 % [B]
Week 39	200.0	217.5	-	-	-	-	+26.3% [C]	+10.7 % [A]
Hematology [c]								
Eosinophil (n)								
Week 13	0.175	0.257	-	-	-	-	-53.1%	-64.6% [E]
Week 26	0.273	0.218	-	-	-	-	-76.2% [A]	-48.6% [D]
Week 39	0.323	0.190	-	-	-	-	-82.3%	-60.5%

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals	**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12 [a]**	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**
Hemoglobin (g/dL)								
Week 13	15.52	-	-	-	-	-	-12.5% [B]	-
Week 26	16.23	-	-	-	-	-	-11.5% [B]	-
Week 39	16.28	-	-	-	-	-	-14.6% [B]	-
Biochemistry[c]								
AST (U/L)								
Week 13	26.3	27.0	-	-	+48% [D]	-	+124% [F]	+96% [F]
Week 26	30.0	31.8	-	-	+42% [A]	-	+103% [C]	+48% [A]
Week 39	23.0	20.3	-	-	-	-	+162% [E]	+170% [B]
ALP (U/L)								
Week 13	-	80.2	-	-	-	-	-	+29% [A]
Week 26	65.5	-	-	-	-	-	+103% [A]	-
Week 39	56.3	-	-	-	-	-	+146% [E]	-
Total bilirubin (g/dL)								
Week 13	-	0.128	-	-	-	-	-	+39% [B]
Week 39	-	0.138	-	-	-	-	-	+74% [D]

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals	**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12 [a]**	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**
Creatinine (mg/dL)								
Week 13	0.80	-	-	-	-	-	+19% [B]	-
Week 26	0.85	-	-	-	-	-	+27% [A]	-
Week 39	0.83	0.70	-	-	-	-	+17% [A]	+31% [A]
Triglycerides (mg/dL)								
Week 26	32.0	-	-	-	-	-	+34% [A]	-
Week 39	39.3	50.3	-	-	-	+53% [A]	+64% [B]	+67% [B]
A/G-P (ratio)								
Week 13	1.465	1.468	-	-	-	-	-30% [B]	-21% [B]
Week 26	1.325	-	-	-	-	-	-37% [B]	-
Week 39	1.565	1.578	-	-	+15% [A]	-	-37% C	+27% [A]
T3 (ng/dL)								
Week 39	108.8	100.2	-	-	-	-	-32% [B]	-30% [D]
Phosphorus (mg/dL)								
Week 13	5.298	-	-	-	-	-	+18% [B]	-
Creatinine Kinase (U/L)								
Week 39	91.5	87.5	-	-	+76% [A]	-	+207% [C]	+127% [E]

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals	**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12** [a]	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**
Albumin (g/dL)								
Week 13	3.55	3.37	-	+6% A	-	+8% B	-10% B	-7% A
Week 26	3.55	-	-	-	-	-	-11% A	-
Week 39	4.03	3.80					-15% C	-8% A
Bone Parameters[c]								
1.25-dihydroxy Vitamin D (pmol/L)								
Week 13	539.15	-	-	-	-	-	-27% A	-
Week 39	440.06	-	-	-	-	-	-51% A	-
25-hydroxy Vitamin D (ng/mL)								
Week 13	-	28.87	-	-	-	-	-	+185% E
Week 39	56.00	-	-	-	-	-	-41% A	-
N-telopeptide; NTx (nM BCE/mM Creatinine)								
Week 39	253.7	229.3	-	-	-	-	-153% E	+60% A

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals	**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12 [a]**	
Gender: Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**
Body mineral density (g/cm^2)								
Whole body								
Week 13	0.336	-	-	-	-	-	-13% [C]	-
Week 39	0.372	-	-	-	-	-	-19% [F]	-
Lumbar vertebrae								
Week 13	0.554	0.533	-	-	-	-	-15% [C]	-9% [A]
Week 39	0.588	0.596	-	-	-	-	-21% [C]	-13% [B]
Organ weight relative to body weight (%)								
Lung								
Week 39	0.78400	0.83031	-	-	-	-	+58% [B]	+35% [B]
Spleen								
Week 39	0.66659	-	-	-	-	-	+119% [C]	-
Gross pathology								
No. examined Week 13	(2)	(2)	(2)	(2)	(2)	(2)	(2)	(2)
Kidney: discoloration pale	-	-	-	-	-	-	-	2
No. examined Week 39	(4)	(4)	(4)	(4)	(4)	(4)	(4)	(4)

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals	**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12 [a]**	
Gender:Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**
Kidney: discoloration pale	-	-	-	-	1	-	4	4
Lung: area raised	1	-	-	-	1	2	3	2
Histopathology								
No. examined Week 39	(4)	(4)	(4)	(4)	(4)	(4)	(4)	(4)
Eye Infiltration: mononuclear cell	-	-	-	-	-	-	3	1
No. examined Week 39	(4)	(4)	(4)	(4)	(4)	(4)	(4)	(4)
Adrenal								
Deposits: pigment	-		-	-	-	-	1	1
Kidney								
Degeneration/regeneration: tubular	-	-	1	-	4	2	4	4
Karyomegaly: tubular	-	-	2	-	4	3	4	4
Infiltration: mononuclear cell	1	-	-	-	-	-	2	2
Liver								
Deposits: pigment	-	-	-	-	2	1	4	4
Hepatocellular cytoplasmic acidophilic inclusion	-	-	-	-	-	-	3	2

Test Item: TAF

Report Title:	A 9 Month Oral Gavage Toxicity Study of GS-7340-02 in the Beagle Dog with a 3 Month Recovery Period For Selected Animals	**Gilead Study No. TOX-120-002** [redacted] **Study No. 89647**

Daily Dose (mg/kg/day)	**0 (Control)**		**2**		**6**		**18/12** [a]	
Gender:Number of Animals	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**	**M: 6**	**F: 6**
Lung								
Infiltration: macrophage	-	-	2	1	1	1	4	4
Histiocytosis	-	-	-	-	-	1	3	2
Inflammation: interstitial	-	-	2	-	-	2	4	-
Spleen								
Infiltration: macrophage	-	-	-	-	-	-	4	3
Recovery Phase Gender:Number of Animals	M: 0	F: 0	M: 0	F: 0	M: 0	F: 0	M: 2	F: 2
Recovery phase	NA	NA	NA	NA	NA	NA	-	-

a Due to severe clinical signs and reduced body weights the dose level was decreased on Day 45 and 51 for males and females, respectively.
b Values below the lower limit of quantitation
c For controls, group means are shown. For treated groups, percent differences from controls are shown. Statistical significance is based on actual data not on the percent.
d One death was related to gavage trauma Significantly different from control group (group 1) value: A - $p \leq 0.05$ B - $p \leq 0.01$ C - $p \leq 0.001$ (Dunnett); D - $p \leq 0.05$ E - $P \leq 0.01$ F - $p \leq 0.001$ (Dunn)

M = Male; F = Female; TK = toxicokinetic; NA = Not Applicable; − Comparable to control animals or no treatment-related findings

7.3.6. A 28-Day Toxicity Study of GS-7340-02 and Tenofovir (GS-1278) Administered Orally to Rhesus Monkeys

Test Article: TAF

Report Title: A 28-Day Toxicity Study of GS-7340-02 and Tenofovir (GS-1278) Administered Orally to Rhesus Monkeys		**Gilead Study No. P2000114** [redacted] **Study No. 1050-11**
Species / Strain: Rhesus monkey	**Duration of Dosing:** 4 weeks	
Initial Age: 2.5 -5.2 years old	**Duration of Postdose:** None	**GLP Compliance:** yes
Date of First Dose: [redacted]	**Method of Administration:** Nasogastric gavage	**Lot Number:** [redacted]
Vehicle: 50mM Citric Acid		
Special Features: Biochemical markers of bone marrow turnover, parathyroid serum levels, and ionized calcium and lactate levels in plasma		
No Observed Adverse Effect Level: 30 mg/kg/day TAF (TFV data not shown)		

Daily Dose (mg/kg/day)	**0 (Control)**		**3**		**30**	
Gender: Number of Animals (TK)	**M: 3**	**F: 3**	**M: 3**	**F: 3**	**M: 3**	**F: 3**
Toxicokinetics (Tenofovir): AUC (hr·µg/mL)[d]						
Day 1	NA[a]		0.472		6.67	
Day 28	NA[a]		0.352		5.87	
Toxicokinetics: (Tenofovir) C_{max} (µg/mL)						
Day 1	NA[a]		0.090		0.904	
Day 28	NA[a]		0.050		0.963	
Noteworthy Findings						
Died or sacrificed moribund	-	-	-	-	1[c]	-
Body Weight (%)[b]	4.1 kg		- 9.7%		+2.4%	

Test Article: TAF

Report Title: A 28-Day Toxicity Study of GS-7340-02 and Tenofovir (GS-1278) Administered Orally to Rhesus Monkeys	**Gilead Study No. P2000114 [redacted] Study No. 1050-11**

Daily Dose (mg/kg/day)	**0 (Control)**		**3**		**30**	
Gender: Number of Animals (Main)	**M: 3**	**F: 3**	**M: 3**	**F: 3**	**M: 3**	**F: 3**
Low Food Consumption (Number of animals effected)	3	2	3	3	2	3
Clinical Observations						
Unformed or watery stools (Number of animals effected)	3	2	3	2	3	3
Hematology[b]						
White blood cell count	15.1		- 43.7%*		- 22.5%	
Serum Chemistry						
Alkaline phosphatase (U/mL)	261		+ 43.3%		+ 54.4%*	
Urinalysis[b]						
Specific gravity [Day 7]	1.001		0%		-0.1%	
Specific gravity [Day 14]	1.007		- 0.3%*		-0.1%	
Specific gravity [Day 28]	1.011		+ 0.2%		-0.3%	
Creatine (mg/dL) [Day 7]	36.3		+ 25.3%		+ 4.4%	
Creatine (mg/dL) [Day 14]	28		- 35.4%*		-36.4%	
Creatine (mg/dL) [Day 28]	32.8		+ 48.2%		+58.8%	

Test Article: TAF

Report Title: A 28-Day Toxicity Study of GS-7340-02 and Tenofovir (GS-1278) Administered Orally to Rhesus Monkeys	**Gilead Study No. P2000114 [redacted] Study No. [redacted] 1050-11**

Daily Dose (mg/kg/day)	**0 (Control)**		**3**		**30**	
Gender: Number of Animals (Main)	**M: 3**	**F: 3**	**M: 3**	**F: 3**	**M: 3**	**F: 3**
Histopathology	No test article histopathological changes were observed					

a Values below the lower limit of quantitation
b For controls, group means are shown. For treated groups, percent differences or fold change from controls are shown. Statistical significance is based on actual data not on the percent.
c Euthanized on Day 18 – this animal had been dosed previously with GS-7340-02 in a single dose PK study ([redacted] 1006-11). Although it had been determined healthy prior to assignment to that study, it developed chronic diarrhea during the 5-day period prior to dosing in and this continued throughout that study, and also into this study. The animal was administered Pepto-Bismol and additional fluids, but still developed signs of dehydration by Day 5. The animal also exhibited signs of malaise. By Day 14 body weight had decreased 17% from prestudy weight. Microscopic evaluation of the GI tract was suggestive of bacterial infection.
d AUC0-∞ on Day 1 and AUC0-τ on Day 28
M = Male; F = Female; TK = toxicokinetic; NA = Not Applicable; + Mild, ++ Moderate, +++ Marked
− Comparable to control animals or no treatment-related findings; * = $p \leq 0.05$ (Dunnett's)

7.4. FTC/TDF

7.4.1. 4-Week Oral Toxicity Study in Dogs

Test Article: FTC + TDF

Report Title: 4-Week Oral Gavage Toxicity and Toxicokinetic Study in Male Dogs with a 4-Week Recovery Period		**Gilead Study No. TX-164-2004** ([redacted] 6511-155)
Species/Strain: Dog/Beagle	**Duration of Dosing:** 4 Weeks (29 days)	
Initial Age: Approximately 4.5 to 5 Months Old	**Duration of Postdose:** 4 Weeks	**GLP Compliance:** Yes
Date of First Dose: [redacted]	**Method of Administration:** Oral gavage	**Vehicle/Formulation:** 50mM citrate buffer
Special Features: Immunophenotyping and Natural Killer Cell Assay		**Lot No.:** FTC = [redacted] TDF = [redacted]
No observable Effect Level: 20 mg/kg/day (FTC), 2/3 mg/kg/day (FTC/TDF)		

Dose (mg/kg)	**0 (Vehicle)**	**20 FTC**	**30 TDF**	**2 FTC/3 TDF**	**20 FTC/ 30 TDF**
Number of Animals	**M: 4**	**M: 4**	**M: 4**	**M: 4**	**M: 6[a]**
Toxicokinetics:					
C_{max} (Day 27) (µg/mL for FTC) (ng/mL for TFV)	-	6.9	2996 TFV	0.9 FTC 154 TFV	7.3 FTC 3439 TFV
AUC_{tau} (Day 27)(µg·h/mL for FTC) (ng·h/mL for TFV)	-	25.3	18413 TFV	2.7 FTC 909 TFV	32.3 FTC 20872 TFV
Noteworthy Findings					
Died or Sacrificed Moribund	0	0	0	0	0
Clinical Observations	No treatment-related findings.				
Body Weight (%)	No treatment-related findings.				
Food Consumption (%)	No treatment-related findings.				
Ophthalmic Examinations	No treatment-related findings.				
Hematology					
Increase APTT	-	-	+	-	+

Test Article: FTC + TDF

Report Title: 4-Week Oral Gavage Toxicity and Toxicokinetic Study in Male Dogs with a 4-Week Recovery Period				**Gilead Study No. TX-164-2004** ([redacted] 6511-155)	
Dose (mg/kg)	**0 (Vehicle)**	**20 FTC**	**30 TDF**	**2 FTC/3 TDF**	**20 FTC/ 30 TDF**
Number of Animals	**M: 4**	**M: 4**	**M: 4**	**M: 4**	**M: 6a**
Increase MCV	-	+	-	-	+
Serum Chemistry					
Creatinine	-	-	I	-	I
Urinalysis	No treatment-related findings.				
Organ Weights (%)	No treatment-related findings.				
Gross Pathology	No treatment-related findings.				
Histopathology					
+ Tubular epithelial necrosis	-	-	2/4		3/4
+ Tubular regeneration	1/4	1/4	4/4	-	4/4
Additional Examinations					
Immunophenotyping	No treatment-related findings.				
NK cell analysis	No treatment-related findings.				
Postdose Evaluation					
Number Evaluated	-	-	-	-	2
Findings	No treatment-related findings.				

a Two males designated for recovery. The hematological and kidney microscopic effects were reversed following recovery.
- = no noteworthy findings; I = increased; + = mild; ++ = moderate; +++ = marked

8. GENOTOXICITY: IN VITRO

8.1. FTC: Ames

Test Article: FTC

Report Title: Mutagenicity Test with FTC in the Salmonella-Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmatory Assay		**Gilead Study No. 18637-0-409R**
Test for Induction of: Reverse mutations at the histidine locus in *S. typhimurium* & Reverse mutations at the tryptophan locus in *E. coli*		
Strains: *Salmonella typhimurium* (TA98, TA100, TA1535, TA1537) & *Escherichia coli* (WP2uvrA)	**No. of Independent Assays:** - S9: 4 experiments; + S9: 4 experiments	**GLP Compliance:** Yes
Metabolizing System: Aroclor™-induced rat liver (S9 mix)	**No. of Cells Analyzed/Culture:** 0.5×10^9 bacteria/mL	**Lot No.:** [redacted]
Vehicles: DMSO **For Test Articles:** FTC- lot [redacted] (± S9)	**For Positive Controls:** DMSO (± S9) 2-aminoanthracene or 2-nitrofluorene or sodium azide or ICR-191 or 4-nitroquinoline-N-oxide	**No. of Replicate Cultures:** [a] Rangefinding study a) single b) single Mutagenicity assays a) triplicate b) triplicate
Treatment[b]: After the overlay had solidified, the plates were inverted and incubated for 48 ± 8 hr at 37 ± 2°C.		**Date of Treatment:** [redacted], 19[redacted] – [redacted], 19[redacted]
Cytotoxic Effects:	No cytotoxicity was observed in the presence or absence of S9 mix as evidenced by a normal background lawn and no test article precipitation.	
Genotoxic Effects:	Range-finding study – No decrease in the number of revertants per plate. Mutagenicity assays – No positive increases in the number of revertants per plate were observed In the confirmatory assay - No positive increases in the number of revertants per plate were observed. The 2.1-fold, non-dose responsive increase was considered artifactual due to a lower than routinely observed vehicle control value. **Conclusion:** FTC did not cause a positive increase in the number of revertants per plate of any of the tester strains in either the presence or absence of microsomal enzymes prepared from Arochlor® -induced rat liver (S9).	

a Dose Range-finding study: a) - S9: 6.67, 10.0, 33.3, 66.7, 100, 333, 667, 1000, 3330, 5000 µg/plate. Vehicle control. b) + S9: (same)
Mutagenicity assays (initial, confirmatory, additional): a) - S9: 100, 333, 1000, 3330, 5000 µg/plate. Positive and vehicle controls. b) + S9: (same)

b When all the required components have been added (tester strain, vehicle or test article, ± S9), the mixture was vortexed and overlaid onto the surface of 25 mL of minimal bottom agar contained in a 15 × 100 mm petri dish.

Assay 1: Experiment ID 18637-B1 **Test Article: FTC**

Report Title: Mutagenicity Test with FTC in the Salmonella-Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmatory Assay	**Gilead Study No. 18637-0-409R**

Metabolic Activation	Dose Level (µg/plate)	Mean revertants per plate with standard deviation									
		TA98		TA100		TA1535		TA1537		WP2uvrA	
		MEAN	S.D.	MEAN	S.D.	MEAN	S.D.	MEAN	S.D.	MEAN	S.D.
Microsomes: Rat											
Vehicle Control		28	7	114	11	13	4	12	2	13	5
Test Article	100	27	5	116	10	10	2	7	3	15	1
	333	28	11	83	7	10	6	8	2	8	2
	1000	26	6	100	15	10	3	8	2	9	2
	3330	23	5	93	12	15	3	12	2	11	4
	5000	21	7	85	9	12	2	7	2	12	1
Positive Control**		1116	109	1103	128	133	8	150	21	160	28
Microsome: None											
Vehicle Control		10	4	87	8	11	5	6	1	8	4
Test Article	100	14	4	93	8	13	2	9	4	14	5
	333	14	3	88	9	12	1	6	1	9	2
	1000	12	2	90	5	13	3	7	2	11	4
	3330	11	1	73	9	12	2	3	3	9	1
	5000	12	4	74	16	11	3	5	2	7	3
Positive Control***		142	30	542	14	527	82	289	30	253	10

Assay 2: Experiment ID 18637-C1 **Test Article: FTC**

Report Title: Mutagenicity Test with FTC in the Salmonella-Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmatory Assay	**Gilead Study No. 18637-0-409R**

Metabolic Activation	**Dose Level (µg/plate)**	**Mean Revertants Per Plate With Standard Deviation**									
		TA98		**TA100**		**TA1535**		**TA1537**		**WP2uvrA**	
		MEAN	**S.D.**	**MEAN**	**S.D.**	**MEAN**	**S.D.**	**MEAN**	**S.D.**	**MEAN**	**S.D.**
Microsomes: rat liver Vehicle Control		16	5	97	7	11	5	9	1	7	4
Test Article	100	17	5	94	8	10	1	7	4	12	4
	333	10	4	93	18	10	1	8	3	10	4
	1000	12	3	67	5	11	3	5	0	10	1
	3330	15	6	70	16	11	3	7	2	15	4
	5000	11	2	52	9	13	5	6	4	12	3
Positive Control**		1045	44	1051	37	109	6	165	27	268	27
Microsome: none Vehicle Control		6*	2	69	9	5	1	6	1	11	3
Test Article	100	NC	-	66	7	10	1	6	1	10	6
	333	NC	-	50	5	8	5	5	1	12	5
	1000	NC	-	79	13	8	2	4	2	12	3
	3330	NC	-	57	7	5	3	2	2	9	4
	5000	NC	-	56	7	2	3	3	1	14	9
Positive Control***		NC	-	411	46	431	38	236	34	203	35

Assay 3: Experiment ID 18637-D1 **Test Article: FTC**

Report Title: Mutagenicity Test with FTC in the Salmonella-Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmatory Assay	**Gilead Study No. 18637-0-409R**

Metabolic Activation	**Dose Level (µg/plate)**	**Mean Revertants Per Plate With Standard Deviation**				
		WP2uvrA			**WP2uvrA**	
		1	**2**	**3**	**MEAN**	**S.D.**
Microsomes: rat liver Vehicle Control		18	24	7	16	9
Test Article	100	11	14	13	13	2
	333	15	12	15	14	2
	1000	9	12	17	13	4
	3330	13	8	12	11	3
	5000	13	10	9	11	2
Positive Control**		289	227	182	233	54

Metabolic Activation	**Dose Level (µg/plate)**	**TA98**			**TA98**	
		1	**2**	**3**	**MEAN**	**S.D.**
Microsomes: rat liver Vehicle Control		14	15	15	15	1
Test Article	100	10	12	11	11	1
	333	17	22	22	20	3
	1000	6	17	9	11	6
	3330	9	7	12	9	3
	5000	6	7	11	8	3
Positive Control***		137	129	141	136	6

* The mean vehicle control value for tester strain TA98 in the absence of S9 was not within the acceptable range. See retest data (experiment ID 18637-D1)

** TA98 2-aminoanthracene: 2.5 µg/plate
TA100 2-aminoanthracene: 2.5 µg/plate
TA1535 2-aminoanthracene: 2.5 µg/plate
TA1537 2-aminoanthracene: 2.5 µg/plate
WP2uvrA 2-aminoanthracene: 25.0 µg/plate

*** TA98 2-nitrofluorene: 1.0 µg/plate
TA100 sodium azide: 2.0 µg/plate
TA1535 sodium azide: 2.0 µg/plate
TA1537 ICR-191: 2.0 µg/plate
WP2uvrA 4-nitroquinoline-N-oxide: 1.0 µg/plate

8.2. FTC: Ames

Test Article: FTC

Report Title: Salmonella/Mammalian-Microsome Assays with 524W91		**Gilead Study No.** MUT203
Test for Induction of: Reverse mutations at histidine locus in *S. typhimurium*	**No. of Independent Assays:** - S9: 3 experiments; + S9: 3 experiments	
Date of Treatment: [redacted] 19[redacted] – [redacted] 19[redacted]	**No. of Replicate Cultures:** [a] Range-finding study a) single b) single Mutagenicity assays a) triplicate b) triplicate	**GLP Compliance:** Yes **Lot No.:** [redacted]
Metabolizing System: Aroclor™-induced rat liver (S9 mix)	**No. of Cells Analyzed/Culture:** 0.5 x 10^9 bacteria/mL	
Vehicles For Test Articles: DMSO 524W91UO reference number; 92/0277-049-C and (± S9)	**Vehicles For Positive Controls:** DMSO + 2-aminoanthracene or 2-nitrofluorene or Sodium azide or ICR-191 or 2-nitrofluorene and (± S9)	
Treatment:[b] Incubation was at 37 ± 2°C for 48 ± 8 hours.	**Strains:** *Salmonella typhimurium* (TA98, TA100, TA1535, TA1537, TA1538)	
Cytotoxic Effects:	No cytotoxicity was observed in presence or absence of S9 mix as evidenced by a normal background lawn and no test article precipitation.	
Genotoxic Effects:	FTC exerted no detectable mutagenic activity in the Ames *Salmonella* assay, either by plate incorporation or by suspension preincubation modification.	

a Dose Range finding study: a) - S9: 6.67, 10.0, 33.3, 66.7, 100, 333, 667, 1000, 3330, 5000 µg/plate. Vehicle control. b) + S9: (same)
Mutagenic, confirmatory, additional: a) - S9: 100, 333, 1000, 3330, 5000 µg/plate. Positive and vehicle controls. b) +S9: (same)

b The test article, the tester strain, and the S9 mix (where appropriate) were combined in molten agar which was overlaid into a minimal agar.

Assay 1: Experiment ID 15437-B1 **Test Article: FTC**

Report Title: Salmonella/Mammalian-Microsome Assays with 524W91 **Gilead Study No. MUT203**

Mean Revertants Per Plate With Standard Deviation

Metabolic Activation	Dose Level (µg/plate)	TA98		TA100		TA1535		TA1537		TA1538	
		MEAN	S.D.	MEAN	S.D.	MEAN	S.D.	MEAN	S.D.	MEAN	S.D.
Microsomes: rat liver Vehicle Control		35	5	251	9	13	1	8	1	20	7
	100	28	7	NC	-	14	5	9	2	20	3
	333	35	3	NC	-	13	2	8	3	20	7
	667	30	6	NC	-	13	3	6	3	22	10
	1000	23	7	NC	-	10	1	6	1	20	3
	3330	40	9	NC	-	8	3	5	2	24	3
Test Article	5000	28	11	NC	-	14	8	6	1	19	7
Positive Control**		976	56	1182	59	181	17	128	7	1189	58
Microsome: none											
Vehicle Control		19	8	159	26	12	4	5	4	14	3
Test article	100	16	3	167	13	11	3	3	1	9	1
	333	16	4	165	20	8	4	9	2	12	4
	667	14	5	164	29	12	4	5	2	15	3
	1000	16	8	176	9	9	3	7	4	13	1
	3330	19	3	187	25	10	3	5	1	10	2
	5000	16	5	188	7	8	0	7	2	10	1
Positive Control***		193	73	719	54	321	24	267	32	327	52

NC = No count due to unacceptable mean vehicle control value. See retest data experiment ID 15437-B2

** = TA98 2-aminoanthracene 2.5 µg/plate
TA100 2-aminoanthracene 2.5 µg/plate
TA1535 2-aminoanthracene 2.5 µg/plate

*** = TA98 2-nitrofluorene 1.0 µg/plate
TA100 sodium azide 2.0 µg/plate
TA1535 sodium azide 2.0 µg/plate

8.3. FTC: Ames

Test Article: FTC

Report Title: Mutagenicity Test of FTC Using Microorganisms		**Gilead Study No.** K01-3154
Test for Induction of: Reverse mutation in bacterial cells	**No. of Independent Assays:** 2	
Species/Strain: *S. typhimurium* (TA98, TA100, TA1535, TA1537), *E. coli*(WP2 *uvr A)*	**No. of Replicate Cultures:** 3 (all negative controls), 2 (#1), 2 (#2)	**GLP Compliance:** Yes **Lot No.:** [redacted]
Metabolizing System: Phenobarbital and 5, 6-benzoflavone induced rat liver S9	**Genotoxic Effects:** None	
Vehicles: Distilled water (FTC); DMSO, except distilled water for sodium azide (positive controls)		**Treatments:** Pre-incubation method
Cytotoxic Effects: No appreciable toxicity was observed	**Date of Treatments:** [redacted]	

Metabolic Activation	Test Article	Dose Level (µg/plate)	Assay No. 1 Initial Mutagenicity Assay Revertant Colony Counts (Mean)				
			TA100	TA1535	WP2 *uvr* A	TA98	TA1537
Without Activation	(Vehicle)	--	131	13	30	26	13
	Test Article	4.88	137	10	33	21	9
		19.5	127	9	35	24	14
		78.1	129	14	26	20	14
		313	118	8	32	23	15
		1250	123	10	35	23	11
		5000	125	12	33	19	13
	AF-2	0.01	523		284		
	Sodium azide	0.5		451			
	AF-2	0.1				509	
	ICR-191	0.5					1139
With Activation	(Vehicle)	--	149	8	34	27	31
	Test Article	4.88	148	7	34	40	21
		19.5	140	11	40	37	23
		78.1	135	6	37	31	26
		313	135	9	32	29	23
		1250	128	5	41	37	29
		5000	137	10	40	33	22
	2AA	1	772				
	2AA	2		203			213

Test Article: FTC

Report Title: Mutagenicity Test of FTC Using Microorganisms	**Gilead Study No. K01-3154**

Metabolic Activation	**Test Article**	**Dose Level (µg/plate)**	**Assay No. 1 Initial Mutagenicity Assay Revertant Colony Counts (Mean)**				
			TA100	**TA1535**	**WP2 *uvr* A**	**TA98**	**TA1537**
With Activation	2AA	10			565		
	2AA	0.5				312	
Metabolic Activation	**Test Article**	**Dose Level (µg/plate)**	**Assay No. 2 Confirmatory Mutagenicity Assay Revertant Colony Counts (Mean)**				
			TA100	**TA1535**	**WP2 *uvr* A**	**TA98**	**TA1537**
Without Activation	(Vehicle)	--	117	10	20	19	8
	Test Article	313	110	8	23	20	12
		625	118	9	26	25	10
		1250	112	12	15	17	12
		2500	115	7	22	16	12
		5000	111	9	26	18	10
	AF-2	0.01	471		216		
	Sodium azide	0.5		411			
	AF-2	0.1				484	
	ICR-191	0.5					1270
With Activation	(Vehicle)	--	121	8	23	25	19
	Test Article	313	132	9	24	32	20
		625	118	7	21	35	28
		1250	137	10	26	25	22
		2500	123	14	29	33	14
		5000	127	9	27	28	16
	2AA	1	650				
	2AA	2		178			172
	2AA	10			538		
	2AA	0.5				381	

AF2: 2-(2-Furyl)-3-(5-nitro-2-furyl) acrylamide
ICR-191: 2-Methoxy-6-chloro-9[3-(2-chloroethyl)-aminopropylamino]acridine-2HCl
2AA: 2-Aminoanthracene

8.4. FTC: Mouse Lymphoma Assay

Test Article: FTC

Report Title: In Vitro Mammalian Cell Gene Mutation Test (Mouse Lymphoma Assay)	**Gilead Study No.** TOX012
Test for Induction of: Forward mutations at the thymidine kinase locus in cultured L5178Y cells. Mutants are unable to metabolize the non-toxic thymidine metabolite, trifluorothymidine (TFT) to its lethal metabolite and hence are resistant to its lethal effect.	**Lot No.:** [redacted]
No. of Independent Assays: Preliminary Toxicity assay: 1 experiment; Mutagenicity assay: 1 experiment	**GLP Compliance:** Yes
Strains: L5178Y mouse lymphoma cells TK+/-	**No. of Replicate Cultures:** [a] Preliminary Toxicity assay a) single b) single Mutagenicity assay a) duplicate b) duplicate
Metabolizing System: Aroclor 1254-induced rat liver (S9 mix)	**No. of Cells Analyzed/Culture:** Adjusted to 3 x 10^5/mL at 24-48 hours after treatment, Cultures less than 3 x 10^5/mL were not adjusted
Vehicles: Sterile distilled water **For Test Article:** FTC (lot [redacted] (± S9))	**For Positive Controls:** a) - S9: Methyl methanesulfonate (MMS) 10 and 20 µg/mL b) + S9: 7,12-Dimethyl-benz(a)anthracene (7,12-DMBA) 2.5 and 4.0 µg/mL
Treatment: Treatment period was for 4 hours at 37 ± 1°C. Recovery at approximately 24 and 48 hours after treatment (± S9).	**Date of Treatment:** [redacted], 19[redacted] – [redacted] 19[redacted]
Cytotoxic Effects:	Suspension growth relative to the solvent controls at 5000 µg/mL was 76% without activation and 99% with S9 activation.
Genotoxic Effects:	FTC was concluded to be negative in the L5178Y/TK+/- Mouse Lymphoma Mutagenesis Assay.

a Preliminary Toxicity assay: a) - S9: 0.5, 1.5, 5.0, 15, 50, 150, 500, 1500, 5000 µg/mL. Vehicle control. b) + S9: (same)
Mutagenicity assay: a) - S9: 1000, 2000, 3000, 4000, 5000 µg/mL. Positive and vehicle controls. b) +S9: (same)

Test Article: FTC

Report Title: In Vitro Mammalian Cell Gene Mutation Test (Mouse Lymphoma Assay)	**Gilead Study No. TOX012**

Assay 1: Study No. G97BG07.702

MUTATION ASSAY WITH ACTIVATION

Test Condition	**Daily Cell Counts (conc.10^6cells)**		**Mutant Colonies Avg.**	**Suspension Growth[a] Total[b]**	**Suspension Growth Control[c] (%)**	**Viable Colonies (VC) Avg.**	**Cloning Growth Avg. VC**	**Cloning Growth Control[d] (%)**	**Total Growth[e] (%)**	**Mutant Frequency[f] (10^6 cells)**
	Day 1	Day 2								
Vehicle Control 1	1.475	1.514	48	24.8		173	173	-		55
Vehicle Control 2	1.398	1.502	51	23.3		172	172	-		60
Positive Control 1										
Methyl Methanesulfonate (µg/mL)										
10	1.029	1.356	224	15.5	64	105	105	61	39	426
20	0.885	0.910	218	8.9	37	50	50	29	11	865
FTC (µg/mL)										
1000A	1.245	1.575	45	21.8	91	174	174	101	91	52
B	1.304	1.631	39	23.6	98	141	141	82	80	55
2000A	1.180	1.611	55	21.1	88	165	165	96	84	66
B	1.221	1.467	58	19.9	83	146	146	85	70	79
3000A	1.098	1.641	53	20.0	83	176	176	102	85	61
B	1.081	1.557	54	18.7	78	145	145	84	65	74
4000A	1.064	1.502	55	17.7	74	143	143	83	61	77
B	0.945	1.505	59	15.8	66	142	142	82	54	83
5000	0.856	1.410	54	13.4	56	166	166	96	54	66

Test Article: FTC

Report Title: In Vitro Mammalian Cell Gene Mutation Test (Mouse Lymphoma Assay)	**Gilead Study No. TOX012**

Assay 1: Study No. G97BG07.702

MUTATION ASSAY WITH ACTIVATION

Test Condition	**Daily Cell Counts (conc.10^6cells)**		**Mutant Colonies Avg.**	**Suspension Growth[a] Total[b]**	**Suspension Growth Control[c] (%)**	**Viable Colonies (VC) Avg.**	**Cloning Growth Avg. VC**	**Cloning Growth Control[d] (%)**	**Total Growth[e] (%)**	**Mutant Frequency[f] (10^6 cells)**
A										
B	0.874	1.387	44	13.5	56	161	161	93	52	55

Report Title: In Vitro Mammalian Cell Gene Mutation Test (Mouse Lymphoma Assay)										Gilead Study No. TOX012
Assay 1: Study No. G97BG07.702			MUTATION ASSAY WITHOUT ACTIVATION							
Test Condition	**Daily Cell Counts (conc.10^6cells)**		**Mutant Colonies Avg.**	**Suspension Growth[a] Total[b]**	**Suspension Growth Control[c] (%)**	**Viable Colonies (VC) Avg.**	**Cloning Growth Avg. VC**	**Cloning Growth Control[d] (%)**	**Total Growth[e] (%)**	**Mutant Frequency[f] (10^6 cells)**
	Day 1	Day 2								
Vehicle Control 1	1.124	1.510	51	18.9		130	130			79
Vehicle Control 2	1.052	1.517	40	17.7		171	171			47
Positive Control 1										
7,12-Dimethyl-benz(a)anthracene (µg/mL)										
2.5	0.816	1.467	158	13.3	73	112	112	74	54	282
4	0.640	0.993	231	7.1	39	87	87	57	22	534
FTC (µg/mL)										
1000 A	1.178	1.530	42	20.0	109	159	159	106	116	53
B	1.163	1.525	42	19.7	108	144	144	96	103	58
2000 A	1.038	1.568	33	18.1	99	132	132	88	87	50
B	1.100	1.513	43	18.5	101	117	117	78	78	74
3000 A	1.106	1.475	50	18.1	99	166	166	110	109	61
B	1.103	1.390	53	17.0	93	138	138	91	85	77
4000 A	1.055	1.450	46	17.0	93	153	153	101	94	60
B	1.058	1.529	46	18.0	98	170	170	112	111	54
5000 A	1.072	1.338	52	15.9	87	171	171	113	99	61
B	1.00	1.57	52	17.5	96	129	129	86	82	81

Report Title: In Vitro Mammalian Cell Gene Mutation Test (Mouse Lymphoma Assay)	Gilead Study No. TOX012

Assay 1: Study No. G97BG07.702

MUTATION ASSAY WITHOUT ACTIVATION

Test Condition	Daily Cell Counts (conc.10^6cells)		Mutant Colonies Avg.	Suspension Growth[a] Total[b]	Suspension Growth Control[c] (%)	Viable Colonies (VC) Avg.	Cloning Growth Avg. VC	Cloning Growth Control[d] (%)	Total Growth[e] (%)	Mutant Frequency[f] (10^6 cells)
	1	1								

a Cultures containing < 0.3×10^6 cells/mL on day 1 and 2 are considered as having 0% total suspension growth.
b Total suspension growth = (Day 1 cell conc./0.3×10^6 cells/mL) × (Day 2 cell conc./Day 1 adjusted cell conc.).
c % of control growth = (total treatment suspension growth / average solvent control total suspension growth) × 100.
d % control cloning growth = (average V.C. of treated culture/average V.C. of solvent control) × 100.
e % total growth = (% suspension growth × % cloning growth)/100.
f Mutant frequency (per 10^6 surviving cells) = (Average # TFT colonies / average # V.C. colonies) × 200.
Note: A and B or 1 and 2 are duplicate cultures

8.5. RPV: Ames

Test Article: RPV

Test: In vitro Ames test in *Salmonella typhimurium*
Species/strain: *Salmonella typhimurium* strain TA1535, TA1537, TA98, TA100, TA102
Metabolizing system: Induced rat (Aroclor 1254) liver metabolic system S9-mix
No. of independent assays: 2
No. of replicate cultures: 3

Treatment: 48 hours
Concentrations: 0, 7.81, 15.63, 31.25, 62.50, 125, 250 and 500 µg/plate TMC278 base
Test article batch: [redacted]
Vehicle test article: DMSO
Positive controls: 2-nitrofluorene, sodium azide, 9-aminoacridine, 2-aminoanthracene, 4-nitroquinoline-N-oxide

Testing facility: J&J PRD
Study No.: TMC278-Exp5540
GLP compliance: Yes

Cytotoxic effects: no - strong precipitation limited the dose

Genotoxic effects: no genotoxic effects

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/plate)	Assay # 1 Revertant Colony Counts (Mean ± SD)				
			TA98	TA1537	TA100	TA1535	TA102
Without S9	DMSO	0.00	16 ± 2.1	4 ± 1.2	112 ± 25.9	11 ± 2.1	229 ± 20.0
	TMC278	7.81	17 ± 2.0	9 ± 2.3	96 ± 3.8	12 ± 4.4	212 ± 18.8
		15.63	12 ± 2.1	8 ± 1.2	97 ± 11.5	14 ± 1.7	221 ± 16.5
		31.25	21 ± 1.4	6 ± 1.2 [a]	93 ± 8.1 [a]	10 ± 3.5 [a]	208 ± 31.2
		62.50	13 ± 4.0 [a]	5 ± 2.3 [a]	79 ± 9.6 [a]	14 ± 2.5 [a]	187 ± 17.0 [a]
		125.00	11 ± 5.3 [a]	6 ± 2.0 [a]	88 ± 10.1 [a]	11 ± 4.4 [a]	179 ± 13.5 [a]
		250.00	15 ± 4.6 [a]	4 ± 1.0 [a]	94 ± 8.5 [a]	12 ± 4.0 [a]	173 ± 31.4 [a]
		500.00	17 ± 1.7 [a]	6 ± 2.9 [a]	77 ± 7.5 [a]	8 ± 1.5 [a]	173 ± 10.1 [a]
	2- Nitrofluorene	5.00	640 ± 68.4				
	Sodium Azide	1.00			699 ± 28.1	344 ± 76.3	
	9-Amino-Acridine	50.00		365 ± 153.1			
	4-Nitroquinoline-N-Oxide	5.00					1224 ± 251.8

a Precipitation at this dose level and above; DMSO: dimethyl sulfoxide.

Test Article: RPV

Study: TMC278-Exp5540: In Vitro Bacterial Reverse Mutation Test with *Salmonella typhimurium* (continued)

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/plate)	Assay # 1 Revertant Colony Counts (Mean ± SD)				
			TA98	TA1537	TA100	TA1535	TA102
With S9 (20 µL)	DMSO	0.00	17 ± 3.2	9 ± 4.4	94 ± 4.4	12 ± 1.2	344 ± 42.1
	TMC278	7.81	26 ± 5.1	5 ± 4.0	90 ± 3.0	12 ± 1.5	293 ± 9.2
		15.63	27 ± 5.3	7 ± 3.5	115 ± 12.3	11 ± 0.6	288 ± 34.9
		31.25	20 ± 5.7 [a]	11 ± 0.6 [a]	97 ± 12.1 [a]	12 ± 1.5 [a]	393 ± 49.7 [a]
		62.50	17 ± 3.6 [a]	7 ± 4.4 [a]	84 ± 10.4 [a]	10 ± 4.4 [a]	356 ± 10.6 [a]
		125.00	17 ± 10.6 [a]	8 ± 4.0 [a]	89 ± 12.0 [a]	7 ± 4.0 [a]	251 ± 58.0 [a]
		250.00	22 ± 1.2 [a]	5 ± 1.5 [a]	87 ± 21.5 [a]	10 ± 0.6 [a]	248 ± 18.3 [a]
		500.00	18 ± 4.0 [a]	4 ± 1.2 [a]	67 ± 7.0 [a]	9 ± 6.0 [a]	207 ± 35.9 [a]
	2-Amino-Anthracene	1.00	837 ± 122.2	-	784 ± 71.1	144 ± 20.0	-
		2.50	-	276 ± 34.9	-	-	1000 ± 194.0

a precipitation at this dose level and above; DMSO: dimethyl sulfoxide.

Test Article: RPV

Study: TMC278-Exp5540: In Vitro Bacterial Reverse Mutation Test with *Salmonella typhimurium* (continued)

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/plate)	Assay # 2 Revertant Colony Counts (Mean ± SD)				
			TA98	TA1537	TA100	TA1535	TA102
Without S9	DMSO	0.00	18 ± 2.6	9 ± 0.6	95 ± 12.5	11 ± 2.0	248 ± 31.2
	TMC278	7.81	16 ± 9.3	8 ± 2.6	86 ± 11.1	9 ± 2.3	201 ± 16.2
		15.63	16 ± 6.6	9 ± 1.0	76 ± 16.8	10 ± 2.1	260 ± 69.4
		31.25	17 ± 5.1 [a]	9 ± 5.1	94 ± 15.6	13 ± 5.5	252 ± 14.4 [a]
		62.50	15 ± 3.6 [a]	9 ± 5.5 [a]	93 ± 8.4 [a]	14 ± 3.5 [a]	315 ± 36.1 [a]
		125.00	15 ± 1.2 [a]	7 ± 2.1 [a]	96 ± 10.2 [a]	8 ± 1.0 [a]	284 ± 39.4 [a]
		250.00	15 ± 4.2 [a]	8 ± 2.6 [a]	104 ± 12.0 [a]	13 ± 4.7 [a]	171 ± 14.0 [a]
		500.00	16 ± 1.5 [a]	8 ± 1.5 [a]	105 ± 15.9 [a]	11 ± 2.0 [a]	172 ± 20.0 [a]
	2- Nitrofluorene	5.00	661 ± 105.6	-	-	-	-
	Sodium Azide	1.00	-	-	837 ± 82.1	253 ± 41.1	-
	9-Amino-Acridine	50.00	-	144 ± 20.8	-	-	-
	4-Nitroquinoline-N-Oxide	5.00	-		-	-	1071 ± 208.7
With S9 (50 µL)	DMSO	0.00	19 ± 2.9	7 ± 1.2	95 ± 13.6	14 ± 3.1	337 ± 34.9
	TMC278	7.81	33 ± 6.2	5 ± 1.0	115 ± 11.5	9 ± 1.7	353 ± 26.0
		15.63	33 ± 4.6	7 ± 0.6	90 ± 20.2	8 ± 3.1	385 ± 26.6
		31.25	31 ± 1.2 [a]	7 ± 2.5	108 ± 12.1	10 ± 2.1	393 ± 34.9 [a]
		62.50	29 ± 3.6 [a]	10 ± 3.5 [a]	92 ± 5.6 [a]	8 ± 1.7 [a]	295 ± 58.3 [a]
		125.00	34 ± 4.7 [a]	6 ± 3.5 [a]	94 ± 4.2 [a]	11 ± 3.1 [a]	261 ± 38.9 [a]
		250.00	29 ± 9.6 [a]	6 ± 0.6 [a]	88 ± 0.6 [a]	5 ± 4.0 [a]	180 ± 4.0 [a]
		500.00	33 ± 2.5 [a]	5 ± 2.0 [a]	65 ± 11.0 [a]	8 ± 3.5 [a]	124 ± 21.2 [a]
	2-Amino-	1.00	215 ±	-	435 ±	64 ± 5.5	-

Test Article: RPV

	Anthracene		112.0		20.1		
		2.50	-	97 ± 6.4	-	-	1184 ± 284.1

a precipitation at this dose level and above; DMSO: dimethyl sulfoxide.

8.6. RPV: Ames

Test Article: RPV

Report Title: **Mutagenicity Test with RPV in the Salmonella typhimurium Reverse Mutation Assay with a Confirmatory Assay**

Study No.: TMC278-Exp5693

Test: *In Vitro* Mammalian Forward Mutation Test
Species/strain: *Salmonella typhimurium*, strains TA1535, TA1537, TA98, TA100, TA102
Metabolizing System: Induced rat (Aroclor 1254) liver metabolic system S9-mix
No of independent assays: 2
No of replicate cultures: 3

Treatment: 48 hours
Concentrations: 7.81, 15.63, 31.25, 62.50, 125, 250 and 500 µg/plate TMC278 base
Test article batch: [redacted]
Vehicle test article: DMSO
Positive controls: 2-nitrofluorene, sodium azide, 9-aminoacridine, 2-aminoanthracene, 4-nitroquinoline-N-oxide

GLP compliance: Yes

Cytotoxic effects: Weak decrease in revertants with some strains
Genotoxic effects: No genotoxic effects

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/plate)	Assay # 1 Revertant Colony Counts (Mean ± SD)				
			TA98	TA1537	TA100	TA1535	TA102
Without S9	DMSO	0.00	12 ± 1.0	7 ± 3.2	108 ± 8.5	9 ± 0.6	291 ± 6.1
	TMC278	7.81	10 ± 3.8	6 ± 3.5	131 ± 22.6	13 ± 2.5	285 ± 12.2
		15.63	13 ± 4.0	10 ± 0.6	146 ± 39.1	11 ± 2.3	311 ± 41.6
		31.25	11 ± 2.6	8 ± 3.8	127 ± 18.2[a]	11 ± 0.6	313 ± 45.5
		62.50	9 ± 0.6[a]	13 ± 1.2[a]	124 ± 13.2[a]	10 ± 3.6[a]	299 ± 43.1[a]
		125.00	11 ± 3.2[a]	8 ± 1.2[a]	128 ± 22.3[a]	11 ± 1.0[a]	257 ± 36.3[a]
		250.00	10 ± 4.0[a]	8 ± 2.3[a]	105 ± 25.7[a]	14 ± 3.2[a]	275 ± 8.3[a]
		500.00	12 ± 0.6[a]	12 ± 4.7[a]	99 ± 12.5[a]	8 ± 3.6[a]	261 ± 12.2[a]
	2-Nitrofluorene	5.00	840 ± 254.0	-	-	-	-
	Sodium Azide	1.00	-	-	469 ± 60.6	207 ± 37.2	-
	9-Amino-Acridine	50.00		123 ± 35.9	-	-	-
	4-Nitroquinoline-N-Oxide	5.00			-	-	2581 ± 531.9

a Precipitation at this dose level and above; DMSO: dimethyl sulfoxide

Test Article: RPV

Metabolic	Control, Test and Reference	Concentration (µg/plate)	Assay # 1 Revertant Colony Counts (Mean ± SD)

Test Article: RPV

Activation	Articles		TA98	TA1537	TA100	TA1535	TA102
With S9 (20 µL)	DMSO	0.00	23 ± 4.6	9 ± 1.2	118 ± 19.5	9 ± 2.5	365 ± 20.1
	TMC278	7.81	23 ± 7.4	6 ± 2.1	124 ± 1.5	9 ± 1.5	327 ± 36.3
		15.63	25 ± 9.5	10 ± 4.6	127 ± 12.7	12 ± 7.4	249 ± 44.1
		31.25	19 ± 1.5	10 ± 0.6[a]	121 ± 6.4[a]	11	243 ± 42.4
		62.50	25 ± 4.6[a]	9 ± 3.5[a]	91 ± 8.7[a]	8 ± 5.5[a]	283 ± 10.1[a]
		125.00	20 ± 4.0[a]	8 ± 4.0[a]	80 ± 14.5 [a]	9 ± 3.2[a]	412 ± 91.7[a]
		250.00	17 ± 10.7[a]	6 ± 0.6[a]	117 ± 6.5[a]	7 ± 2.0[a]	259 ± 83.9[a]
		500.00	9 ± 1.7[a]	8 ± 2.1[a]	109 ± 10.4[a]	11 ± 3.0[a]	147 ± 44.1[a]
	2-Amino-anthracene	1.00	1161 ± 160.5		1765 ± 180.0	211 ± 26.6	
		2.50	-	344 ± 72.0	-	-	1279 ± 47.4

a Precipitation at this dose level and above; DMSO: dimethyl sulfoxide

Test Article: RPV

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/plate)	Assay # 2 Revertant Colony Counts (Mean ± SD)				
			TA98	TA1537	TA100	TA1535	TA102
Without S9	DMSO	0.00	13 ± 2.0	8 ± 2.6	121 ± 14.5	12 ± 4.0	228 ± 42.1
	TMC278	7.81	15 ± 3.5	8 ± 2.5	129 ± 9.2	15 ± 1.5	292 ± 72.8
		15.63	17 ± 2.5	10 ± 2.9	145 ± 24.8	17 ± 2.6	284 ± 52.0
		31.25	13 ± 5.5[a]	7 ± 1.0[a]	109 ± 5.0[a]	15 ± 4.0[a]	261 ± 36.3
		62.50	14 ± 4.0[a]	6 ± 2.1[a]	118 ± 11.1[a]	13 ± 4.9[a]	255 ± 33.3[a]
		125.00	16 ± 4.6[a]	7 ± 2.5[a]	145 ± 21.0[a]	15 ± 4.7[a]	279 ± 12.9[a]
		250.00	11 ± 4.6[a]	6 ± 2.5[a]	123 ± 5.6[a]	15 ± 2.1[a]	200 ± 6.9[a]
		500.00	14 ± 3.5[a]	8 ± 4.4[a]	113 ± 6.5[a]	14 ± 2.5[a]	225 ± 18.0[a]
	2-Nitrofluorene	5.00	805 ± 48.9	-	-	-	-
	Sodium Azide	1.00	-	-	473 ± 16.2	472 ± 28.8	-
	9-Amino-Acridine	50.00	-	84 ± 4.4	-	-	-
	4-Nitroquinoline-N-Oxide	5.00	-	-	-	-	2363 ± 352.8
With S9 (50 µL)	DMSO	0.00	20 ± 6.4	8 ± 2.1	121 ± 12.3	15 ± 2.5	340 ± 14.4
	TMC278	7.81	21 ± 7.0	9 ± 2.9	128 ± 11.2	10 ± 2.1	343 ± 76.4
		15.63	24 ± 2.0	9 ± 5.5	140 ± 12.3	12 ± 2.1	377 ± 63.8
		31.25	27 ± 6.0	9 ± 1.7	115 ± 7.5	10 ± 2.9	309 ± 32.3
		62.50	27 ± 5.0[a]	11 ± 5.0	131 ± 6.0[a]	9 ± 3.1[a]	275 ± 30.6
		125.00	18 ± 2.6[a]	12 ± 1.5[a]	115 ± 5.7[a]	8 ± 4.2[a]	328 ± 59.2[a]
		250.00	21 ± 6.0[a]	8 ± 1.5[a]	130 ± 4.2[a]	12 ± 3.5[a]	304 ± 52.5[a]
		500.00	19 ± 7.2[a]	10 ± 3.1[a]	126 ± 4.5[a]	8 ± 3.5[a]	263 ± 22.0[a]
	2-Amino-anthracene	1.00	320 ± 32.7	-	537 ± 54.3	103 ± 14.7	-
		2.50	-	94 ± 4.6	-	-	1475 ± 20.1

a Precipitation at this dose level and above; DMSO: dimethyl sulfoxide

8.7. RPV: Ames

Test Article: RPV

Report Title: **Mutagenicity Test with RPV in the Salmonella typhimurium Reverse Mutation Assay with a Confirmatory Assay**

Study No.: TMC278-NC335

Test: In Vitro Bacterial Reverse Mutation Test
Species/strain: *S. Typhimurium* strains TA98, TA1537, TA100, TA1535 and TA102
Metabolizing System: Induced rat (Aroclor 1254) liver metabolic system S9-mix
No of independent assays: 2
No of replicate cultures: 3
No of cells analyzed/culture: 10

Treatment: 48 to 72hours
Concentrations: 3.91, 7.81, 15.63, 31.25, 62.5, 125 and 250 µg/plate TMC278.HCl
Test article batch: [redacted]
Vehicle test article: DMSO
Vehicle positive controls: DMSO and water
Positive control: 2-nitrofluorene, sodium azide, 9-aminoacridine, 2-aminoanthracene, 4-nitroquinoline-N-oxide

GLP compliance: Yes

Cytotoxic effects: No
Genotoxic effects: No
Assay # 1 (Revertant Colony Counts (Mean ± SD)

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA1537	TA102
Without S9	DMSO	0	15.7 ± 2.1	119.0 ± 18.0	11.3 ± 2.1	11.0 ± 2.6	365.3 ± 10.7
	TMC278	3.91	11.7 ± 1.5	101.3 ± 7.6	9.0 ± 1.0	11.7 ± 3.5	382.7 ± 5.0
		7.81	11.3 ± 3.5	109.0 ± 16.4	6.7 ± 4.7	9.7 ± 2.1	378.0 ± 9.8
		15.63	22.7 ± 3.8	129.0 ± 4.6	8.0 ± 2.6	8.0 ± 3.0	363.3 ± 26.3
		31.25	20.7 ± 3.2	121.7 ± 23.7	9.7 ± 3.5	6.0 ± 2.6	378.7 ± 27.9
		62.5	13.7 ± 5.9	127.0 ± 16.7	7.3 ± 0.6	5.3 ± 1.2	357.3 ± 18.9
		125	P1 15.7 ± 4.7	110.0 ± 11.8 P1	P1 8.3 ± 2.1	P1 6.0 ± 2.6	P1 364.0 ± 20.3
		250	P1 19.7 ± 4.0	106.3 ± 14.4 P1	P1 9.7 ± 3.2	P1 5.7 ± 1.2	P1 303.0 ± 18.2
	2-Nitrofluorene	5.00	514.7 ±146.8 *	-	-	-	-
	Sodium Azide	1.00	-	* 536.0 ± 4.4	478.3 ± 44.5 *	-	-
	9-Amino-Acridine	50.00	-	-	-	887.0 ± 177.2 *	-

Test Article: RPV

	4-Nitroquinoline-N-Oxide	5.00	-	-	-	-	3309.3 ± 44.8	*

* more than 2-fold increase with TA98, TA100 and TA102 compared to the vehicle control, more than 3-fold increase with TA1535 and TA1537 compared to the vehicle control, P1: Slight precipitation, DMSO: dimethyl sulfoxide, -: not applicable

Assay # 1 (Revertant Colony Counts (Mean ± SD))

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA1537	TA102
With S9	DMSO	0	16.3 ± 2.5	150.7 ± 19.0	14.3 ± 1.2	4.0 ± 0.0	415.3 ± 51.0
	TMC278	3.91	15.7 ± 2.5	163.3 ± 31.5	11.3 ± 0.6	4.0 ± 1.7	476.0 ± 7.2
		7.81	21.7 ± 9.6	176.7 ± 13.9	12.3 ± 4.7	5.5 ± 3.5	448.0 ± 37.5
		15.63	19.7 ± 4.7	161.3 ± 2.5	13.0 ± 4.4	6.0 ± 2.0	456.3 ± 55.6
		31.25	19.3 ± 5.0	180.0 ± 7.8	7.7 ± 0.6	4.7 ± 2.5	478.0 ± 32.1
		62.5	21.0 ± 3.6	222.3 ± 15.2	11.7 ± 0.6	5.3 ± 1.2	483.0 ± 30.3
		125	20.3 ± 3.8 P1	171.3 ± 15.5 P1	10.0 ± 4.6 P1	6.0 ± 4.2 P1	387.0 ± 67.7 P1
		250	22.0 ± 8.5 P2	163.0 ± 14.5 P2	8.7 ± 4.0 P2	4.3 ± 0.6 P2	238.7 ± 71.1 P2
	2-Amino-anthracene	2.5	1244.0 ± 79.5 *	1102.3 ± 338.5 *	116.7 ± 5.5 *	124.0 ± 16.5 *	-
		7.5	-	-	-	-	2516.7 ± 41.0 *

a unless stated otherwise, *more than 2-fold increase with TA98, TA100 and TA102 compared to the vehicle control, more than 3-fold increase with TA1535 and TA1537 compared to the vehicle control, P1: Slight precipitation, P2: Moderate precipitation, DMSO: dimethyl sulfoxide, -: not applicable

Assay # 2 (Revertant Colony Counts (Mean ± SD)

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA1537	TA102
Without S9	DMSO	0	19.3 ± 3.8	113.7 ± 3.2	7.0 ± 2.6	7.3 ± 0.6	288.0 ± 19.1
	TMC278	3.91	14.0 ± 4.6	97.3 ± 10.1	7.3 ± 2.5	4.7 ± 1.5	303.3 ± 7.6
		7.81	13.0 ± 7.8	96.3 ± 21.5	7.3 ± 4.0	8.0 ± 4.4	339.7 ± 12.7
		15.63	14.7 ± 2.5	106.0 ± 18.4	5.3 ± 2.3	7.3 ± 2.5	341.0 ± 21.6
		31.25	14.7 ± 6.5	114.0 ± 6.1	7.3 ± 3.1	6.7 ± 3.8	295.3 ± 13.4
		62.5	15.0 ± 5.0	97.3 ± 8.4	7.3 ± 4.2	8.3 ± 1.2	292.7 ± 22.5
		125	19.0 ± 7.0	113.7 ± 3.8	5.0 ± 2.6	5.3 ± 3.8	288.3 ± 20.5
		250	18.0 ± 3.6	104.3 ± 7.8	6.3 ± 3.5	7.7 ± 3.5	278.7 ± 13.3
	2-Nitrofluorene	5.00	1048.7 * ± 44.8	-	-	-	-
	Sodium Azide	1.00	-	415.7 * ± 2.1	313.0 * ± 10.4	-	-
	9-Amino-Acridine	50.00	-	-	-	124.3 * ± 39.5	-
	4-Nitroquinoline-N-Oxide	5.00	-	-	-	-	3250.0 ± * 168.1

a unless stated otherwise; *more than 2-fold increase with TA98, TA100 and TA102 compared to the vehicle control, more than 3-fold increase with TA1535 and TA1537 compared to the vehicle control; DMSO: dimethyl sulfoxide; -: not applicable

Assay # 2 (Revertant Colony Counts (Mean ± SD)

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA1537	TA102
With S9	DMSO	0	19.3 ± 3.5	135.0 ± 11.3	9.0 ± 1.0	6.7 ± 1.5	363.7 ± 42.7
	TMC278	3.91	29.7 ± 3.5	125.0 ± 13.5	9.0 ± 5.2	4.7 ± 3.8	385.3 ± 29.3
		7.81	25.0 ± 5.3	129.3 ± 4.7	7.3 ± 0.6	2.7 ± 1.5	389.3 ± 25.1
		15.63	25.0 ± 4.4	182.3 ± 26.5	8.7 ± 5.7	5.7 ± 1.5	406.0 ± 19.1
		31.25	26.7 ± 1.2	147.3 ± 14.6	7.7 ± 1.2	3.7 ± 1.2	417.7 ± 9.9
		62.5	30.3 ± 7.2 P1	139.7 ± 15.1 P1	9.7 ± 1.2 P1	8.3 ± 4.9	358.3 ± 17.2
		125	23.0 ± 4.6 P1	152.0 ± 4.6 P1	9.7 ± 1.2 P1	9.7 ± 1.5 P1	268.0 ± 18.3 P1
		250	28.7 ± 5.5 P2	126.0 ± 4.6 P2	7.3 ± 0.6 P2	5.3 ± 1.5 P2	205.3 ± 32.1 P2
	2-Amino-anthracene	7.5	2417.3 ± 156.2 *	2217.7 ± 28.5 *	130.0 ± 7.0 *	242.3 ± 9.5 *	1228.7 ± 90.1 *

a unless stated otherwise; *more than 2-fold increase with TA98, TA100 and TA102 compared to the vehicle control, more than 3-fold increase with TA1535 and TA1537 compared to the vehicle control; P1: Slight precipitation; P2: Moderate precipitation; DMSO: dimethyl sulfoxide

8.8. RPV: Ames

Test Article: RPV

Report Title: **Mutagenicity Test with RPV in the Salmonella typhimurium Reverse Mutation Assay with a Confirmatory Assay**

Study No.: TMC278-NC279

Test: In Vitro Bacterial Reverse Mutation Test **Species/strain:** *Salmonella Typhimurium* strains TA98, TA1537, TA100, TA1535 and TA102

Metabolizing System: Human S9_Mix

No of independent assays: 4

No of replicate cultures: 3

Treatment: 48 to 72h

Concentrations: of 0, 3, 7.81, 10, 15.63, 25, 31.25, 50, 62.5, 100, 125, 200, 250, 400 and 500 µg/plate TMC278 base

Test article batch: [redacted]

Vehicle test article: DMSO

Vehicle positive controls: DMSO and water

Positive controls: 2-nitrofluorene, sodium azide, 9-aminoacridine, 2-aminoanthracene, 4-nitroquinoline-N-oxide

GLP compliance: Yes

Cytotoxic effects: Weak

Genotoxic effects: No

Assay # 1 (Revertant Colony Counts (Mean ± SD)

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100[a]	TA1535	TA1537
Without S9	DMSO	0	14.3 ± 1.2	99.0 ± 7.5	11.7 ± 5.0	5.7 ± 3.2
	2-Nitrofluorene	5.00	557.7 ± 64.8*	-	-	-
	Sodium Azide	1.00	-	-	411.0 ± 26.5*	-
		2.50	-	796.7 ± 26.5*	-	-
	9-Aminoacridine	50.00	-	-	-	20.3 ± 2.1*
	4-Nitroquinoline-N-Oxide	5.00	-	-	-	-

a pre-incubation; * more than 2-fold increase with TA98 and TA100 compared to the vehicle control, more than 3-fold with TA1535 and TA1537 compared to the vehicle control; DMSO: dimethyl sulfoxide; -: Not Applicable

Assay # 1 (Revertant Colony Counts (Mean ± SD))

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA1537
With S9	DMSO	0	25.7 ± 7.8	156.3 ± 17.6	11.0 ± 4.4	13.7 ± 3.8
	TMC278	7.81	33.3 ± 5.7	135.7 ± 17.2	11.3 ± 3.1	16.7 ± 2.1
		15.63	28.0 ± 7.5	153.0 ± 6.9	10.0 ± 4.0	13.7 ± 5.5
		31.25	31.3 ± 5.9	145.3 ± 4.7	17.7 ± 8.5	19.7 ± 3.8
		62.5	19.0 ± 10.8	158.7 ± 28.1	16.0 ± 3.6	30.3 ± 4.9
		125	18.0 ± 1.7 P	161.3 ± 25.6	15.7 ± 7.5 P	26.7 ± 10.2 P
		250	17.0 ± 7.0 P	163.3 ± 27.2 P	10.3 ± 3.1 P	33.0 ± 11.3 P
		500	15.7 ± 1.2 P	132.0 ± 6.0 P	9.3 ± 4.6 P	25.7 ± 10.0 P

a pre-incubation; * more than 2-fold increase with TA98 and TA100 compared to the vehicle control, more than 3-fold with TA1535 and TA1537 compared to the vehicle control; DMSO: dimethyl sulfoxide; -: Not Applicable; P: Precipitation.

Assay # 2 (Revertant Colony Counts (Mean ± SD))

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA102
Without S9	DMSO	0	20.3 ± 0.6	141.0 ± 3.5	20.3 ± 4.9	367.7 ± 18.3
	2-Nitrofluorene	5.00	386.0 ± 20.7*	-	-	-
	Sodium Azide	1.00	-	-	610.0 ± 27.6*	-
		2.50	-	1115.3 ± 40.4*	-	-
	4-Nitroquinoline-N-Oxide	5.00	-	-	-	2127.3 ± 122.2*

a unless stated otherwise; * more than 2-fold increase with TA98, TA100 and TA102 and more than 3-fold with TA1535 compared to the vehicle control; DMSO: dimethyl sulfoxide; -: Not Applicable

Assay # 2 (Revertant Colony Counts (Mean ± SD))

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA98	TA100	TA1535	TA102
With S9	DMSO	0	34.7 ± 8.0	143.3 ± 16.3	24.0 ± 6.6	479.3 ± 44.2
	TMC278	3	31.0 ± 5.0	150.0 ± 9.6	22.7 ± 3.5	467.0 ± 63.5
		10	17.7 ± 1.5	145.3 ± 4.5	20.3 ± 5.1	445.0 ± 56.4
		25	32.3 ± 9.0	137.0 ± 15.4	23.7 ± 3.1	480.0 ± 36.4
		50	41.0 ± 4.4	145.3 ± 23.7	25.3 ± 4.0	477.0 ± 21.2
		100	26.0 ± 3.6 P	141.0 ± 9.5 P	26.3 ± 1.5 P	556.7 ± 57.6 P
		200	36.7 ± 8.3 P	142.0 ± 12.8 P	28.0 ± 11.3 P	473.3 ± 33.9 P
		400	34.0 ± 10.8 P	134.3 ± 22.9 P	20.7 ± 8.1 P	443.3 ± 42.8 P

a unless stated otherwise; P: Precipitation; DMSO: dimethyl sulfoxide

Assay # 3 (Revertant Colony Counts (Mean ± SD))

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA102	TA100
Without S9	DMSO	0	244.0 ± 7.9	82.0 ± 3.6
	Sodium Azide	2.5	-	1542.7 ± 25.8*
	4-Nitroquinoline-N-Oxide	5.00	1744.0 ± 85.1*	-

a unless stated otherwise; DMSO: dimethyl sulfoxide; -: Not Applicable; more than 2-fold increase TA100 and TA102 compared to the vehicle control.

Assay # 3 (Revertant Colony Counts (Mean ± SD)

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA102	
With S9	DMSO	0	273.3 ± 25.5	
	TMC278	3	280.7 ± 33.2	
		7.81	372.7 ± 70.3	
		15.63	330.3 ± 43.0	
		31.25	314.0 ± 31.4	
		62.50	351.7 ± 17.0	
		125	380.3 ± 11.0	
		250	390.0 ± 13.2	P
		500	445.3 ± 24.8	P

a unless stated otherwise; P: Precipitation; DMSO: dimethyl sulfoxide; -: Not Applicable.

Assay # 4 (Revertant Colony Counts (Mean ± SD))

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA1537	TA100
Without S9	DMSO	0	5.7 ± 2.3	71.3 ± 3.2
	Sodium Azide	2.5	-	1435.3 ± 28.0*
	9-Aminoacridine	50.00	288.7 ± 38.7*	-

a unless stated otherwise; * more than 2-fold increase TA100 and more than 3-fold increase TA1537 compared to the vehicle control;
DMSO: dimethyl sulfoxide; -: Not Applicable.

Assay # 4 (Revertant Colony Counts (Mean ± SD)

Test Article: RPV

Metabolic Activation	Test Article	Concentration (µg/plate)[a]	TA1537	
With S9	DMSO	0	13.3 ± 2.3	
	TMC278	3	13.0 ± 2.6	
		10	17.7 ± 2.5	
		25	10.3 ± 0.6	
		50	15.0 ± 4.4	
		100	17.3 ± 1.5	P
		200	11.0 ± 5.3	P
		400	10.0 ± 2.6	P

a unless stated otherwise; P: Precipitation, DMSO: dimethyl sulfoxide; -: Not Applicable

8.9. RPV: Mouse Lymphoma Assay

Test Article: RPV

Report Title: **In Vitro Mammalian Forward Mutation Test with L5178Y Mouse Lymphoma Cells (TK-locus) using the Microtiter® Fluctuation Technique**

Study No.: TMC278-Exp5539

Test: In Vitro Mammalian Forward Mutation Test

Species/strain: L5178Y Mouse Lymphoma Cells (TK-locus)

Metabolizing system: Aroclor 1254 induced rat liver S9 mix

No. of independent assays: 5

No. of replicate cultures: 2 x 10^3cells/well

Treatment: Microtiter® Fluctuation Technique

Concentrations: 0, 0.5, 1, 2, 2.5, 4, 5, 6, 8, 10, 15, 20, 25, 30, 35, 40, 50, 60, 70, 80, 90, 100, 250, 500 µg/mL TMC278 base

Test article batch: [redacted]

Vehicle test article: DMSO

Positive controls: DMN, MMS

GLP compliance: Yes

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/mL)	Assay # 1						
			SG		PE			MF	
			Total	Relative	Total	Relative	RTG	Total	Corrected
3 hour treatment without S9	DMSO	0	32	-	106	-	-	84	74
	TMC278	1	37	114	98	92	105	-	-
		2.5	36	112	116	109	122	-	-
		5	36	112	91	85	95	-	-
		10	32	99	92	87	86	79	80
		25	31	95	100	94	89	104	100
		50	32	99	101	95	94	116	111
		100	18	55	98	92	51	112	110
		250*	12	36	80	76	27	121	147
		500*	7	21	82	77	16	94	109
	MMS	15	36	111	74	69	77	211	309

* precipitation, DMSO: dimethyl sulfoxide; - not applicable, SG: suspension growth, PE : plating efficiency, RTG: relative total growth, MF : mutant frequency, MMS: methyl methanesulfonate, DMN: N-nitrosodimethylamine.

Test Article: RPV

Study: TMC278-Exp5539: In Vitro Mammalian Forward Mutation Test with L5178Y Mouse Lymphoma Cells (TK- locus) using the Microtiter® Fluctuation Technique (continued)

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/mL)	Assay # 1						
			SG		PE			MF	
			Total	Relative	Total	Relative	RTG	Total	Corrected
24 hour treatment without S9	DMSO	0	31	-	95	-	-	68	66
	TMC278	20	21	67	82	86	58	116	138
		30	26	85	92	97	82	100	104
		40	25	83	80	85	70	102	121
		50	24	78	78	82	64	104	127
		60	28	91	79	83	76	93	111
		70*	26	84	78	82	69	111	137

Test Article: RPV

		80*	31	100	65	68	68	-	-
		90*	28	92	79	83	77	-	-
		100*	26	84	61	65	55	-	-
	MMS	7.5	13	42	47	49	20	326	894
			Assay # 2						
3 hour treatment with S9	DMSO	0	34	-	112	-	-	72	60
	TMC278	1	31	91	123	109	100	-	-
		2.5	36	104	138	124	128	-	-
		5	37	107	105	93	100	-	-
		10	36	106	112	100	106	106	91
		15	36	106	95	85	90	110	112
		20	32	95	123	109	104	92	71
		25*	25	72	89	80	57	92	97
		30*	29	86	106	95	82	91	81
		35*	18	54	114	102	55	114	97
	DMN	400 nL/mL	35	103	82	73	75	259	366

*: precipitation, DMSO: dimethyl sulfoxide; - not applicable, SG : suspension growth, PE : plating efficiency, RTG: relative total growth, MF : mutant frequency, MMS: methyl methanesulfonate, DMN: N-nitrosodimethylamine

Test Article: RPV

Study: TMC278-Exp5539: In Vitro Mammalian Forward Mutation Test with L5178Y Mouse Lymphoma Cells (TK- locus) using the Microtiter® Fluctuation Technique (continued)

Metabolic Activation	**Control, Test and Reference Articles**	**Concentration (µg/mL)**	**Assay # 3**						
			SG		**PE**			**MF**	
			Total	**Relative**	**Total**	**Relative**	**RTG**	**Total**	**Corrected**
3 hour treatment without S9	DMSO	0	34	-	110	-	-	110	96
	TMC278	1	36	105	105	95	100	-	-
		2.5	34	98	114	104	102	104	87
		5	34	100	91	82	82	98	103
		10	38	109	98	89	97	106	104
		25	31	91	94	85	78	138	146
		50	27	77	95	86	67	124	128
		100	12	35	94	85	29	110	113
		250*	1	3	78	71	2	-	-
		500*	1	2	60	54	1	-	-
	MMS	15	36	105	78	71	75	421	840
			Assay # 4						
24 hour treatment without S9	DMSO	0	14	-	89	-	-	131	144
	TMC278	2.5	17	120	89	100	120	-	-
		5	18	126	91	102	128	109	116
		10	20	144	92	103	149	113	119
		20	19	139	95	106	148	121	124
		30	20	141	88	98	139	98	106
		40	20	144	96	108	156	110	110
		50*	20	143	85	96	136	122	139
		60*	23	166	84	94	157	-	-

Test Article: RPV

		70*	22	161	89	100	161	-	-
	MMS	7.5	8	56	47	53	30	303	788

*: precipitation, DMSO: dimethyl sulfoxide; - not applicable, SG : suspension growth, PE : plating efficiency, RTG: relative total growth, MF : mutant frequency, MMS: methyl methanesulfonate, DMN: N-nitrosodimethylamine.

Test Article: RPV

Study: TMC278-Exp5539: In Vitro Mammalian Forward Mutation Test with L5178Y Mouse Lymphoma Cells (TK- locus) using the Microtiter® Fluctuation Technique (continued)

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/plate)	Assay # 5						
			SG		PE			MF	
			Total	Relative	Total	Relative	RTG	Total	Corrected
3 hour treatment with S9	DMSO	0	41	-	123	-	-	99	77
	TMC278	0.5	47	115	106	87	100	-	-
		1	45	109	105	85	93	102	93
		2	44	107	108	88	95	95	83
		4	47	116	101	83	95	103	97
		6	42	104	98	80	83	100	97
		8*	36	88	100	81	72	95	90
		10*	23	57	92	75	43	111	116
		15*	0	0	-	-	-	-	-
		20*	0	0	-	-	-	-	-
	DMN	400 nL/mL	43	105	91	74	78	247	309

* precipitation, DMSO: dimethyl sulfoxide; - not applicable, SG : suspension growth, PE : plating efficiency, RTG: relative total growth, MF : mutant frequency, MMS: methyl methanesulfonate., DMN: N-nitrosodimethylamine.

8.10. RPV: Mouse Lymphoma

Test Article: RPV

Report Title: **In Vitro Mammalian Forward Mutation Test with L5178Y Mouse Lymphoma Cells (TK-locus) using the Microtiter® Fluctuation Technique** **TMC278-NC336**

Test: In Vitro Mammalian Forward Mutation Test
Species/strain: L5178Y mouse lymphoma cells
Metabolizing System: Aroclor 1254 induced rat liver S9 mix
No of independent assays: 2
No of replicate cultures: 2

Treatment: Microtiter® Fluctuation Technique
Concentrations: 0, 2.5, 5, 10, 12.5, 15, 20, 25, 30, 35, 40 and 50 µg/mL TMC278.HCl
Test article batch: [redacted]
Vehicle test article: DMSO
Positive controls: MMS, B(a)P

GLP compliance: Yes

Assay # 1

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/mL)	% RTG	MF
3 hours treatment without S9	DMSO	0	100	66.38
	TMC278	15	84	70.52
		20	89	62.17
		25	90	59.76
		30*	74	63.29
		40*	81	42.94
		50*	82	71.34
	MMS	15	47	488.91
		20	32	571.54
3 hours treatment with S9	DMSO	0	100	67.38
	TMC278	2.5	100	50.61
		5	121	50.26
		10	101	70.35
		15	85	63.63
		20	87	47.75
	B(a)P	2	47	673.77
		3	20	858.72

*: precipitation, DMSO: dimethyl sulfoxide; - not applicable, MMS: methyl methanesulfonate, MF : mutant frequency, RTG : relative total growth, B(a)P:benzo (a)pyrene

Test Article: RPV

Study TMC278-NC336: Mutation at the Thymidine Kinase (tk) Locus of Mouse Lymphoma L5178Y Cells (MLA) using the Microtitre® Fluctuation Technique (continued)

Assay # 2

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/mL)	% RTG	MF
3 hours treatment without S9	DMSO	0	100	97.95
	TMC278	5	99	77.11
		10	100	85.77
		15	124	70.85

Test Article: RPV

		20*	142	87.89
		25*	136	100.10
		30*	127	73.93
	MMS	15	75	376.88
		20	56	484.53
3 hours treatment with S9	DMSO	0	100	64.48
	TMC278	2.5	93	68.65
		5	83	68.72
		10	82	56.94
		12.5	76	68.65
		15	70	70.99
		20*	72	86.05
	B(a)P	2	15	983.93
		3	15	440.26

*: precipitation, DMSO: dimethyl sulfoxide; - not applicable, MMS: methyl methanesulfonate, MF : mutant frequency, RTG : relative total growth, B(a)P:benzo (a)pyrene

Test Article: RPV

Study TMC278-NC336: Mutation at the Thymidine Kinase (tk) Locus of Mouse Lymphoma L5178Y Cells (MLA) using the Microtitre® Fluctuation Technique (continued)

Assay # 2

Metabolic Activation	Control, Test and Reference Articles	Concentration (µg/mL)	% RTG	MF
24 hours treatment without S9	DMSO	0	100	104.54
	TMC278	2.5	98	76.96
		5	95	82.34
		10	67	76.78
		20*	30	83.18
		25*	52	100.66
		30*	69	94.34
		35*	85	83.53
		40*	96	79.97
		50*	97	84.74
	MMS	5	44	712.13
		7.5	25	1210.66

* precipitation, DMSO : dimethyl sulfoxide; - : not applicable, MMS : methyl methanesulfonate, MF : mutant frequency, RTG : relative total growth.

8.11. TAF: Ames

Test Article: TAF

Report Title:	**Salmonella – Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmation Assay with GS-7340-02**	**Gilead Study No. V990212 [redacted] Study No. 20900-0-409OECD**
Test for Induction of: Reverse mutation in bacterial cells	**No. of Independent Assays:** 2	
Strains: *Salmonella typhimurium and Escherichia coli*	**No. of Replicate Cultures:** 3	**GLP Compliance:** yes
Metabolizing System: : Aroclor™-induced rat liver S9	**No. of Cells Analyzed/Cultured:** ~10^8	**Lot Number:** [redacted]
Vehicle: Dimethylsulfoxide (DMSO)	**Vehicle for Positive Controls:** Deionized (DI) Water (sodium azide only); DMSO (all other positive controls)	
Treatment: Plate incorporation for 52 ± 4 hours		**Date of Treatment:** initial [redacted] 19[redacted]/confirmatory [redacted] 19[redacted]
Cytotoxic Effects: None		
Genotoxic Effects: None		

Metabolic Activation	**Treatment**	**Dose Level (µg/plate)**	**Plate Incorporation – Initial Assay Revertants/Plate (Mean ± SD)**				
			TA98	**TA100**	**TA1535**	**TA1537**	**WP2*uvr*A**
Without Activation	DMSO	50 µL/plate	17 ± 6 N	96 ± 12 N	9 ± 5 N	5 ± 4 N	16 ± 6 N
	GS-7340-02	100	14 ± 4 N	98 ± 10 N	8 ± 2 N	5 ± 1 N	21 ± 4 N
		333	21 ± 7 N	98 ± 13 N	12 ± 6 N	5 ± 2 N	21 ± 1 N
		1000	18 ± 3 N	90 ± 8 N	10 ± 3 N	7 ± 2 N	16 ± 2 N
		3330	14 ± 7 N	93 ± 2 N	16 ± 5 N	5 ± 2 N	18 ± 3 N
		5000	12 ± 2 R	84 ± 19 N	13 ± 6 N	8 ± 3 N	17 ± 1 N
	2-nitrofluorene	**1.0**	244 ± 32 N				
	sodium azide	**2.0**		796 ± 63 N	668 ± 60 N		
	ICR-191	**2.0**				564 ± 19 N	
	4-nitroquinoline-N-oxide	1.0					184 ± 24 N

Test Article: TAF

Report Title:	**Salmonella – Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmation Assay with GS-7340-02**	**Gilead Study No. V990212 [redacted] Study No. 20900-0-409OECD**

Metabolic Activation	**Treatment**	**Dose Level (µg/plate)**	**Plate Incorporation – Initial Assay Revertants/Plate (Mean ± SD)**				
			TA98	**TA100**	**TA1535**	**TA1537**	**WP2*uvr*A**
With Activation	DMSO	50 µL/plate	31 ± 12 N	146 ± 9 N	18 ± 1 N	9 ± 5	17 ± 3
	GS-7340-02	100	30 ± 8 N	117 ± 9 N	12 ± 1 N	10 ± 4 N	16 ± 13 N
		333	30 ± 2 N	117 ± 18 N	10 ± 3 N	5 ± 3 N	23 ± 2 N
		1000	29 ± 4 N	129 ± 10 N	18 ± 6 N	11 ± 3 N	22 ± 4 N
		3330	26 ± 3 N	116 ± 18 N	14 ± 2 N	9 ± 3 N	20 ± 2 N
		5000	29 ± 3 N	114 ± 26 N	13 ± 7 N	4 ± 4 N	15 ± 3 N
	benzo[a]pyrene	2.5	412 ± 35 N				
	2-aminoanthracene	2.5		862 ± 132 N	162 ± 22 N	174 ± 12 N	
	2-aminoanthracene	25.0					280 ± 36 N

N = Normal
R = Slightly reduced
SD = Standard Deviation

Test Article: TAF

Report Title:	**Salmonella – Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmation Assay with GS-7340-02**	**Gilead Study No. V990212 [redacted] Study No. 20900-0-409OECD**
Test for Induction of: Reverse mutation in bacterial cells	**No. of Independent Assays:** 2	
Strains: *Salmonella typhimurium* and *Escherichia coli*	**No. of Replicate Cultures:** 3	**GLP Compliance:** yes
Metabolizing System: Aroclor™-induced rat liver S9	**No. of Cells Analyzed/Cultured:** ~10^8	**Lot Number:** [redacted]
Vehicle: Dimethylsulfoxide (DMSO)	**Vehicle for Positive Controls:** Deionized (DI) Water (sodium azide only); DMSO (all other positive controls)	
Treatment: Plate incorporation for 52 ± 4 hours		**Date of Treatment:** initial [redacted] 19[redacted]/confirmatory [redacted] 19[redacted]

Cytotoxic Effects: None

Genotoxic Effects: None

Metabolic Activation	**Treatment**	**Dose Level (µg/plate)**	**Plate Incorporation – Confirmatory Assay Revertants/Plate (Mean ± SD)**				
			TA98	**TA100**	**TA1535**	**TA1537**	**WP2*uvr*A**
Without Activation	DMSO	50 µL/plate	20 ± 4^N	85 ± 7^N	13 ± 7^N	4 ± 3^N	15 ± 2^N
	GS-7340-02	100	15 ± 8^N	74 ± 3^N	9 ± 5^N	5 ± 2^N	14 ± 2^N
		333	22 ± 3^N	80 ± 10^N	13 ± 6^N	6 ± 3^N	19 ± 4^N
		1000	23 ± 1^N	92 ± 11^N	9 ± 5^N	4 ± 0^N	16 ± 5^N
		3330	20 ± 4^N	96 ± 7^N	9 ± 4^N	6 ± 5^N	19 ± 3^N
		5000	15 ± 3^N	83 ± 12^N	11 ± 3^N	6 ± 2^N	15 ± 6^N
	2-nitrofluorene	**1.0**	234 ± 17^N				
	sodium azide	**2.0**		752 ± 30^N	615 ± 28^N		
	ICR-191	**2.0**				614 ± 58^N	
	4-nitroquinoline-N-oxide	1.0					196 ± 50^N

Test Article: TAF

Report Title:	**Salmonella – Escherichia coli/Mammalian-Microsome Reverse Mutation Assay with a Confirmation Assay with GS-7340-02**	**Gilead Study No. V990212 [redacted] Study No. 20900-0-409OECD**

Metabolic Activation	**Treatment**	**Dose Level (µg/plate)**	**Plate Incorporation –Confirmatory Assay Revertants/Plate (Mean ± SD)**				
			TA98	**TA100**	**TA1535**	**TA1537**	**WP2*uvr*A**
With Activation	DMSO	50 µL/plate	27 ± 4[N]	109 ± 23[N]	10 ± 2[N]	7 ± 3[N]	15 ± 6[N]
	GS-7340-02	100	25 ± 2[N]	94 ± 14[N]	11 ± 4[N]	5 ± 2[N]	15 ± 2[N]
		333	28 ± 3[N]	103 ± 11[N]	9 ± 4[N]	6 ± 2[N]	20 ± 9[N]
		1000	27 ± 2[N]	89 ± 7[N]	13 ± 5[N]	10 ± 3[N]	23 ± 6[N]
		3330	21 ± 4[N]	107 ± 18[N]	14 ± 3[N]	7 ± 1[N]	20 ± 2[N]
		5000	26 ± 1[N]	90 ± 6[N]	11 ± 2[N]	7 ± 4[N]	21 ± 6[N]
	benzo[a]pyrene	2.5	441 ± 10[N]				
	2-aminoanthracene	2.5		1225 ± 162[N]	158 ± 8[N]	193 ± 24[N]	
	2-aminoanthracene	25.0					242 ± 15[N]

N = Normal
SD = Standard Deviation

8.12. TAF: Mouse Lymphoma Mutation Assay

Test Article: TAF

Report Title: **L5178Y TK+/- Mouse Lymphoma Forward Mutation Assay with a Confirmatory Assay with GS-7340-02**		**Gilead Study No. V990213 [redacted] Study No. 20900-0-431 ICH**
Test for Induction of: Forward gene mutation at TK locus	**No. of Independent Assays:** 2	
Cell Line: : L5178Y/TK$^{+/-}$	**No. of Replicate Cultures:** 1	**GLP Compliance:** yes
Metabolizing System: Aroclor 1254-induced rat liver (S9)	**No. of Cells Analyzed/Culture:** Treated: 6 x 10^6 Cloned: 3 x 10^6	**Lot Number:** [redacted]
Vehicle for Test Article: Dimethylsulfoxide (DMSO)	**Vehicle for Positive Controls:** Cell culture grade water for methyl methanesulfonate; DMSO for methylcholanthrene	**Date of Treatment:** initial [redacted] 19[redacted]/confirmatory [redacted] 19[redacted]
Treatment: 4 hours without and with S9 (Initial Assay)		
Cytotoxic Effects: None		
Genotoxic Effects: None		

Metabolic Activation	**Test Article**	**Concentration (µg/mL)**	**RSG (%)[a]**	**Cloning Efficiency[b]**	**Relative Growth[c]**	**Mutant Frequency (10^{-6})[d]**
Without Activation 4 hour Treatment	DMSO	1% (µL/mL)	113.3	92.5	104.8	57.4
		1% (µL/mL)	103.1	93.9	96.8	57.5
		1% (µL/mL)	83.7	113.6	95.1	53.4
	GS-7340-02	39.3	77.1	92.9	71.6	53.6
		78.5	87.8	89.4	78.5	51.9
		157	74.1	112.5	83.3	53.9

Test Article: TAF

Report Title:	**L5178Y TK+/- Mouse Lymphoma Forward Mutation Assay with a Confirmatory Assay with GS-7340-02**	**Gilead Study No. V990213 [redacted] Study No. 20900-0-431 ICH**

Metabolic Activation	Test Article	Concentration (µg/mL)	RSG (%)[a]	Cloning Efficiency[b]	Relative Growth[c]	Mutant Frequency (10^{-6})[d]
Without Activation 4 hour Treatment	GS-7340-02	313	109.9	93.5	102.8	69.4
		625	88.5	105.2	93.1	70.0
		1250	78.8	94.1	74.2	67.1
		2500	55.1	106.1	58.4	77.3
		5000	28.4	88.3	25.1	106.6
	Methyl methanesulfonate	6.5	88.1	88.1	77.6	149.6
		13	67.7	61.2	41.4	252.8

Test Article: TAF

Report Title:	**L5178Y TK+/- Mouse Lymphoma Forward Mutation Assay with a Confirmatory Assay with GS-7340-02**	**Gilead Study No. V990213 [redacted] Study No. 20900-0-431 ICH**

Metabolic Activation	Test Article	Concentration (µg/mL)	RSG (%)[a]	Cloning Efficiency[b]	Relative Growth[c]	Mutant Frequency (10^{-6})[d]
With Activation 4 hour Treatment	DMSO	1% (µL/mL)	119.9	106.1	127.2	61.2
		1% (µL/mL)	92.5	96.9	89.6	64.3
		1% (µL/mL)	87.7	96.9	85.0	72.9
	GS-7340-02	1250	110.4	93.4	103.2	91.1
		1500	123.1	94.1	115.8	99.3
		2000	91.5	98.7	90.3	82.7
		2500	80.8	86.0	69.5	126.0
		3000	80.2	102.4	82.1	82.2
		3500	53.5	121.5	65.0	85.2
		4000	61.2	103.7	63.5	84.6
		4500	46.1	98.9	45.6	94.5
	Methycholanthrene	2	79.5	81.1	64.5	310.8
		4	75.6	113.6	85.9	272.2

a Cum. RSG = Cumulative Suspension Growth Relative to the Average Vehicle Control Suspension Growth
b Cloning Efficiency = Total Viable Colony Count/Number of Cells seeded * 100
c Relative Growth = (Relative Suspension Growth * Relative Cloning Efficiency) / 100
d Mutant Frequency = (Total Mutant Colonies/Total Viable Colonies) * 2x10E-4
Decimal is moved to express the frequency in units of 10E-6
Expressed as Total Mutant Frequency

8.13. FTC/TDF: Ames

Test Article: FTC + TDF

Report Title: Bacterial Reverse Mutation Assay		**Gilead Study No.** TX-164-2002
Test for Induction of: Reverse mutation in bacterial cells	**No. of Independent Assays:** 2	
Species/Strain: *S. typhimurium, E. coli*	**No. of Replicate Cultures:** 2 (#1), 3 (#2)	**GLP Compliance:** Yes
Date of Treatments: [redacted] and [redacted]	**Treatments:** Plate incorporation	**Lot No.:** [redacted] (TDF); [redacted] (FTC)
Metabolizing System: Aroclor-induced rat liver S9	**Vehicles:** DMSO (FTC/TDF), DMSO, except water for sodium azide (positive controls)	
Cytotoxic Effects: No appreciable toxicity was observed	**Genotoxic Effects:** None	

Metabolic Activation	Test Article	Dose Level (µg/plate)	Assay No. 1 Initial Mutagenicity Assay Revertant Colony Counts (Mean ± SD) TA98	TA100	TA1535	TA1537	WP2 *uvr*A
Without Activation	(Vehicle)	50 µL/plate	13 ± 4	112 ± 12	19 ± 6	5 ± 1	16 ± 4
	Test Article	1.5	13 ± 1	149 ± 16	21 ± 5	5 ± 1	13 ± 4
		5.0	12 ± 1	135 ± 14	17 ± 4	6 ± 1	14 ± 2
		15	15 ± 1	111 ± 8	15 ± 0	5 ± 2	12 ± 4
		50	17 ± 1	127 ± 5	16 ± 1	7 ± 3	13 ± 1
		150	13 ± 3	144 ± 16	16 ± 4	8 ± 1	19 ± 1
		500	15 ± 3	135 ± 16	16 ± 6	5 ± 1	14 ± 3
		1500	13 ± 3	157 ± 3	17 ± 3	6 ± 1	14 ± 2
		5000	11 ± 0	155 ± 6	16 ± 7	7 ± 1	12 ± 3
	2-nitrofluorene	1.0	170 ± 11				
	Sodium azide	1.0		546 ± 11	398 ± 6		
	9-aminoacridine	75				1424 ± 127	
	MMS	1000					128 ± 14

Test Article: FTC + TDF

Report Title: Bacterial Reverse Mutation Assay	**Gilead Study No.** TX-164-2002

Metabolic Activation	**Test Article**	**Dose Level**	**Assay No. 1 Initial Mutagenicity Assay Revertant Colony Counts (Mean ± SD)**				
		(µg/plate)	**TA98**	**TA100**	**TA1535**	**TA1537**	**WP2 *uvr*A**
With Activation	(Vehicle)	50 µL/plate	16 ± 1	143 ± 11	14 ± 8	9 ± 2	15 ± 0
	Test Article	1.5	20 ± 8	131 ± 25	14 ± 1	7 ± 1	13 ± 1
		5.0	17 ± 1	146 ± 5	10 ± 3	8 ± 4	15 ± 2
		15	18 ± 2	143 ± 21	11 ± 1	10 ± 2	16 ± 1
		50	22 ± 8	127 ± 6	15 ± 4	8 ± 1	22 ± 4
		150	18 ± 1	140 ± 1	15 ± 1	6 ± 1	18 ± 2
		500	19 ± 4	148 ± 31	12 ± 4	7 ± 1	14 ± 4
		1500	22 ± 4	148 ± 9	11 ± 1	8 ± 0	11 ± 3
		5000	20 ± 1	127 ± 4	13 ± 1	10 ± 1	17 ± 6
	2-aminoanthracene	1.0	689 ± 145	734 ± 0	107 ± 28	93 ± 33	
	2-aminoanthracene	10					608 ± 76

Test Article: FTC + TDF

Report Title: Bacterial Reverse Mutation Assay	**Gilead Study No.** TX-164-2002

Metabolic Activation	**Test Article**	**Dose Level (µg/plate)**	**Assay No. 2 Confirmatory Mutagenicity Assay Revertant Colony Counts (Mean ± SD)**				
			TA98	**TA100**	**TA1535**	**TA1537**	**WP2 *uvr*A**
Without Activation	(Vehicle)	50µL/plate	22 ± 3	109 ± 22	16 ± 2	6 ± 2	12 ± 4
	Test Article	50	27 ± 2	147 ± 3	19 ± 4	6 ± 4	12 ± 2
		150	29 ± 6	104 ± 23	14 ± 2	5 ± 1	13 ± 4
		500	32 ± 6	142 ± 6	18 ± 3	6 ± 2	13 ± 2
		1500	29 ± 3	127 ± 3	17 ± 2	4 ± 1	14 ± 3
		5000	28 ± 4	132 ± 25	22 ± 4	5 ± 3	10 ± 4
	2-nitrofluorene	1.0	173 ± 23				
	Sodium azide	1.0		569 ± 17	357 ± 20		
	9-aminoacridine	75				1282 ± 126	
	MMS	1000					111 ± 13
With Activation	(Vehicle)	50µL/plate	26 ± 5	136 ± 5	15 ± 3	4 ± 1	11 ± 4
	Test Article	50	30 ± 4	139 ± 14	12 ± 4	6 ± 2	14 ± 1
		150	32 ± 2	143 ± 8	10 ± 3	5 ± 2	11 ± 1
		500	28 ± 3	159 ± 21	13 ± 5	7 ± 4	11 ± 4
		1500	30 ± 3	150 ± 9	13 ± 3	5 ± 3	14 ± 4
		5000	27 ± 3	130 ± 19	13 ± 1	10 ± 2	11 ± 3
	2-aminoanthracene	1.0	640 ± 75	917 ± 64	101 ± 13	79 ± 17	
	2-aminoanthracene	10					581 ± 82

8.14. FTC/TDF: Mouse Lymphoma Assay

Test Article: FTC + TDF

Report Title: In Vitro Mammalian Cell Gene Mutation Test (L5178Y/TK+/-	**Gilead Study No. TX-164-2003** ([redacted])	
Test for induction of : Forward mutations at the thymidine kinase locus	**No of independent assays :** Preliminary Toxicity assay: 1, Mutagenicity assay: 1	**GLP compliance:** Yes
Strains: L5178Y mouse lymphoma cells	**Metabolizing system:** Aroclor 1254-induced rat liver (S9 mix)	**Lot No.:** [redacted] (FTC); [redacted] (TDF)
No. of replicate cultures[a]: Preliminary assay: a) single, b) single; Mutagenicity assay: a) duplicate, single for positive controls, b) duplicate, single for positive controls		
No. of cells analyzed / culture: Adjusted to 3 x 10^5/mL at 24 and 48 hours after treatment; cultures less than 3 x 10^5/mL were not adjusted		
Test Article Vehicle: DMSO	**Positive control:** a) –S9: methyl methanesulfonate (MMS) 15 and 20 µg/mL, b) +S9: 7,12-dimethyl-benz(a)anthracene) 2.5 and 4.0 µg/mL	**Dates of treatment:** Preliminary toxicity assay: [redacted] Mutagenicity assay: [redacted]
Treatment: Treatment period was 4 hours with and without S9 activation at 37±1ºC. Recovery at approximately 24 and 48 hours after treatment (+/-S9)		
Cytotoxic effects: Suspension growth relative to the solvent controls at 3575 µg/mL (10 mM) was 3% without activation and 0% with S9 activation		
Genotoxic effects: FTC/TDF was concluded to be positive in the L5178Y/TK$^{+/-}$ Mouse Lymphoma Mutagenesis Assay		

a Preliminary toxicity assay a) –S9: 0.5, 1.5, 5, 15, 50, 150, 500, 1500, 3575 µg/mL. Vehicle control. b) +S9: (same) Mutagenicity assay a) –S9: 15, 50, 100, 150, 200, 300 µg/mL. Positive and vehicle controls. b) +S9: 50, 100, 200, 300, 400, 500, µg/mL. Positive and vehicle controls.

Test Article: FTC + TDF

Report Title: In Vitro Mammalian Cell Gene Mutation Test (L5178Y/TK+/-	**Gilead Study No.** TX-164-2003 ([redacted])

Assay 1: MUTATION ASSAY WITHOUT ACTIVATION

Test Condition	Daily cell counts (Conc. 10^6 cells)[a]		Mutant colonies avg.	Suspension growth total[b]	Suspension growth control (%)[c]	Viable colonies (VC) avg.	Cloning growth control (%)[d]	Total growth (%)[e]	Mutant frequency (10^6 cells)[f]
	Day 1	Day 2							
Vehicle control									
1	1.366	1.688	78	25.6		221			71
2	1.386	1.654	68	25.5		173			78
Positive control: Methyl methanesulfonate (µg/mL)									
15	1.194	1.590	188	21.1	83	150	76	63	251
20	1.154	1.726	205	22.1	87	162	83	72	253
FTC/TDF (µg/mL)									
15 A	1.265	1.563	53	22.0	86	191	97	84	56
15 B	1.266	1.799	63	25.3	99	188	96	95	67
50 A	1.214	1.735	69	23.4	92	183	93	85	75
50 B	1.307	1.709	69	24.8	97	165	84	82	84
100 A	1.128	1.691	128	21.2	83	154	78	65	167
100 B	1.159	1.651	134	21.3	83	69	35	29	386
150 A	0.823	1.466	289	13.4	52	97	49	26	595
150 B	0.857	1.461	257	13.9	54	109	55	30	474
200 A	0.592	0.980	++	6.4	25	66	33	8	++
200 B	0.612	0.966	247	6.6	26	84	43	11	591

Test Article: FTC + TDF

Report Title: In Vitro Mammalian Cell Gene Mutation Test (L5178Y/TK+/-	**Gilead Study No.** TX-164-2003 ([redacted])

Assay 1: MUTATION ASSAY WITHOUT ACTIVATION

Test Condition	Daily cell counts (Conc. 10^6 cells)[a]		Mutant colonies avg.	Suspension growth total[b]	Suspension growth control (%)[c]	Viable colonies (VC) avg.	Cloning growth control (%)[d]	Total growth (%)[e]	Mutant frequency (10^6 cells)[f]
	Day 1	Day 2							
Vehicle control									
1	0.953	1.462	94	15.5		196			96
2	0.983	1.458	84	15.9		169			100
Positive control: 7,12 Dimethylbenz(a)anthracene (µg/mL)									
2.5	0.770	1.489	229	12.7	81	112	61	50	411
4.0	0.600	1.074	249	7.2	46	77	42	19	650
FTC/TDF (µg/mL)									
50 A	0.890	1.499	141	14.8	94	159	87	82	177
50 B	0.919	1.455	115	14.9	95	158	86	82	146
100 A	0.850	1.598	171	15.1	96	167	91	88	205
100 B	0.846	1.561	176	14.7	93	121	66	62	291
200 A	0.738	1.596	149	13.1	83	141	77	64	211
200 B	0.800	1.628	122	14.5	92	140	77	71	174
300 A	0.592	1.408	197	9.3	59	125	68	40	316
300 B	0.593	1.441	179	9.5	60	129	71	43	277
400 A	0.339	1.133	201	4.3	27	91	50	14	443
400 B	0.345	1.167	227	4.5	28	102	56	16	447

a Cultures containing < 0.3×10^6 cells/mL on day 1 and 2 are considered as having 0% total suspension growth.
b Total suspension growth = (Day 1 cell conc./0.3×10^6 cells/mL) × (Day 2 cell conc./Day 1 adjusted cell conc.)
c % of control growth = (total treatment suspension growth / average solvent control total suspension growth) × 100
d % control cloning growth = (average V.C. of treated culture/average V.C. of solvent control) × 100
e % total growth = (% suspension growth × % cloning growth)/100
f Mutant frequency (per 10^6 surviving cells) = (Average # TFT colonies / average # V.C. colonies) × 200
++ - Too toxic to count, total growth < 10%

9. GENOTOXICITY: IN VIVO

9.1. FTC: Micronucleus in Mice

Test Article: FTC

Report Title: Mammalian Erythrocyte Micronucleus Test		**Gilead Study No. TOX011**
Test for Induction of: Micronucleus formation	**Treatment Schedule:** Single dose 500, 1000 & 2000 mg/kg	
Strains: Mouse / ICR	**Sampling Time:** 24 hours and 48 hours (vehicle control and high dose only)	**GLP Compliance:** Yes
Age: 6 to 8 Weeks Old	**Method of Administration:** Oral gavage	**Lot No.:** [redacted]
Date of Dosing: [redacted], 19[redacted] – [redacted], 19[redacted]	**Vehicle/Formulation:** Sterile distilled water 10 mL/kg + FTC [redacted] (free base)	
Special Features: none	**Cells Evaluated:** Polychromatic erythrocytes in bone marrow	

No. of Cells Analyzed/Animal: Bone marrow was collected from femurs and 2 to 4 slides were prepared from each mouse. The number of micronucleated normochromic erythrocytes in the field of 2000 polychromatic erythrocytes was enumerated.

Toxic/Cytotoxic Effects: No mortality, clinical signs of toxicity, or evidence of bone marrow toxicity were observed in any mice in any FTC treated group during the course of the micronucleus study. All mice appeared normal throughout the observation period.

Genotoxic Effects: FTC did not induce a significant increase in the incidence of micronucleated polychromatic erythrocytes in bone marrow and was concluded to be negative in the micronucleus test using male ICR mice.

Test Article	Dose (mg/kg)	No. of Animals	Harvest Time (hour)	PCE/Total Erythrocytes (mean ± sd)	Number/1000 PCEs (mean ± sd)	Number/PCEs Scored
Vehicle control (water)	Vehicle	5	24	0.60 ± 0.01	0.8 ± 0.27	8/10,000
	Vehicle	5	48	0.59 ± 0.05	0.4 ± 0.42	4/10,000
Positive control (Cyclophosphamide)		5	24	0.57 ± 0.02	40.0 ± 6.62	400/10,000*
Test Article						
Low test dose	500	5	24	0.61 ± 0.02	0.5 ± 0.35	5/10,000
Mid test dose	1000	5	24	0.56 ± 0.06	1.1 ± 0.55	11/10,000
High test dose	2000	5	24	0.58 ± 0.03	1.2 ± 1.52	12/10,000
	2000	5	48	0.57 ± 0.05	0.2 ± 0.27	2/10,000

PCE = polychromatic erythrocytes; * = $p \leq 0.05$, ANOVA followed by Dunnett's t-test

9.2. RPV: Micronucleus in Mice

Test Article: RPV

Report Title: In Vivo Micronucleus Test on Bone Marrow Cells of Mice

Study No.: TMC278-Exp5538

Test: *In vivo* micronucleus test
Species/strain: Mouse/SPF Albino Swiss (CD1)
Age at first dose: Approximately 8 weeks
Cells evaluated: PCEs of bone marrow
No. of cells analyzed/animal: 1000 (PCEs + NCEs) to determine bone marrow proliferation and 2000 PCEs to screen for micronuclei

Treatment: polyethylene glycol (PEG) 400 solution with 100 mg/ml citric acid
Administration route/method: oral/gavage
Doses: 0, 100, 400 and 1600 mg/kg b.w TMC278 base
Test article batch: [redacted]
Vehicle/formulation: Polyethylene glycol 400 solution with 100 mg/mL citric acid

GLP compliance: Yes

Toxic/Cytotoxic effects: no statistically significant bone marrow toxic effects were observed following treatment with 100, 400 or 1600 mg/kg TMC278 base.

Genotoxic effects: no genotoxic effects were observed following treatment with 100, 400 or 1600 mg/kg TMC278. under the conditions described in this test

Dose (mg/kg/day)	**0**		**100**		**400**		**1600**	
No. of animals	**M:5**	**F:5**	**M:5**	**F:5**	**M:5**	**F:5**	**M:5**	**F:5**
Died or killed prematurely	0	0	0	0	0	0	0	0
Dose (mg/kg/day)	**0**		**100**		**400**		**1600**	
No. of animals	**-**	**-**	**M:6**	**F:6**	**M:6**	**F:6**	**M:10**	**F:10**
C_{1h} (µg/mL)	-	-	39	33	59	68	60	58
AUC_{0-6h} Day 1 (µg·h/mL)	-	-	158	130	262	258	307	287

Test Article: RPV

Study: TMC278-Exp5538: In vivo Micronucleus Test on Bone Marrow Cells of Mice (continued)

Noteworthy Findings

Control, Test and Reference Articles	Dose (mg/kg body weight)	Number of Animals and Gender	Sampling Time (Hours)	# MNPCEs per 10000 PCEs	Ratio PCEs / NCEs
PEG 400	0	M/5	24	11	1.04
		F/5	24	14	1.07
		M/5	48	9	0.92
		F/5	48	22	1.27
TMC278	100	M/5	24	15	0.99
		F/5	24	12	1.34
		M/5	48	5	1.23
		F/5	48	17	1.08
	400	M/5	24	10	0.90
		F/5	24	13	1.31
		M/5	48	20	0.95
		F/5	48	28	1.16
	1600	M/5	24	8	1.24
		F/5	24	9	1.48
		M/5	48	15	0.95
		F/5	48	15	1.00
Cyclophosphamide	40 mg/mL	M/5	48	185	0.55
		F/5	48	95	0.73

M: male; F: female, MNPCEs: micronucleated polychromatic erythrocytes; NCEs: normochromatic erythrocytes; PEG: polyethylene glycol, PCEs: polychromatic erythrocytes.

9.3. TAF: Micronucleus in Mice

Test Article: TAF

Report Title: **In Vivo Mouse Micronucleus Assay with GS-7340-02**		**Gilead Study No. M2000113 [redacted] Study No. 21816-0-445OECD**
Test for Induction of: : Bone marrow micronuclei	**Treatment Schedule:** single dose	
Species / Strain: Mouse/CD-1®(ICR) BR	**Sampling Time:** 24 and 48 hours after dosing	**GLP Compliance:** yes
Age: 9 weeks	**Method of Administration:** Oral Gavage	**Date of Treatment:** [redacted]
No. of Cells Evaluated / Animal: 2000	**Vehicle:** 50mM citric acid	**Lot No**. [redacted]
Special Features: None	**Positive Control Vehicle:** Deionized (DI) Water	
Toxic / Cytotoxic Effects: Cytotoxicity to the bone marrow was noted at the 2000 mg/kg dose group at the 24 and 48 hour harvest timepoints.		
Genotoxic Effects: GS-7340-02 was evaluated as negative in the mouse bone marrow micronucleus assay under the conditions of the protocol.		

Treatment	Dose (mg/kg)	Number of Males	Harvest Time (Hours)	Mean Ratio PCE:NCE (±SE)	Mean % MNPCE (±SE)
				Male	Male
50mM Citric Acid	20 mL/kg	5	24	0.76 ± 0.08	0.03 ± 0.01
		5	48	0.78 ± 0.06	0.04 ± 0.02
GS-7340-02	500	5	24	0.74 ± 0.02	0.08 ± 0.03
	1000	5	24	0.74 ± 0.07	0.10 ± 0.05
	2000	5	24	0.49 ± 0.07**	0.09 ± 0.02
		5	48	0.24 ± 0.01**	0.12 ± 0.04
Cyclophosphamide	80	5	24	0.65 ± 0.05	2.71 ± 0.39*

*Significantly greater than the corresponding vehicle control, $p<0.01$
**Significantly less than the corresponding vehicle control, $p\leq0.05$
GLP = Good Laboratory Practice
MNPCE = Micronucleated PCEs
NCE = Normochromatic erythrocyte
PCE = Polychromatic erythrocyte
SE = Standard Error

10. CARCINOGENICITY

10.1. FTC: Mouse

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice							**Gilead Study No.** TOX109 ([redacted] 814-005)	
Species/ Strain: Mouse/ CD-1® (ICR) BR	**Duration of Dosing:** 104 Weeks							
Initial Age: 6 Weeks	**Method of Administration:** Oral gavage						**GLP Compliance:** Yes	
Date of First Dose: [redacted]	**Vehicle/Formulation:** 0.5% Methylcellulose						**Lot No.:** [redacted]; [redacted]	
Special Features: None	**Basis for High-Dose Selection:** Results from 6 month mouse study; Toxicokinetic endpoint							
Dose (mg/kg/day)	**0 (Vehicle)**		**80**		**250**		**750**	
Gender	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Toxicokinetics								
AUC (µg·h/mL) Week 26	nd*	nd*	27.48	23.74	90.94	91.71	287.30	322.65
Number of Animals								
At Start	60	60	60	60	60	60	60	60
Died/Sacrificed Moribund	33	38	31	38	37	35	30	34
Terminal Sacrifice	27	22	29	22	23	25	30	26
Survival (%)	45	37	48	37	38	42	50	43
Body Weight[a] (%)	40.6 g	31.9 g	+1	-1	0	-3	-3	-3
Food Consumption[a] (%)	6.0 g/day	5.8 g/day	0	+3	+3	+7	-2	+7
Number of Animals with Neoplastic Lesions								
Adrenal Glands — Adenoma, subcapsular cell, benign, primary	0	2	0	1	0	0	0	0
Adrenal Glands — Pheochromocytoma, benign, primary	0	0	0	0	0	1	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice						**Gilead Study No. TOX109** ([redacted] 814-005)			
Dose (mg/kg/day)		**0 (Vehicle)**		**80**		**250**		**750**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Bone	Osteoma, benign, primary	0	1	0	0	0	0	0	0
Bone Marrow, Femur	Hemangiosarcoma, malignant, primary	0	0	0	0	0	0	0	1
Bone, Vertebra	Osteosarcoma, malignant, primary	0	0	0	0	0	0	0	1
Brain	Astrocytoma, malignant, primary	0	0	1	0	0	0	0	0
Cavity, Abdominal	Carcinoma, squamous cell, malignant, secondary	0	0	0	0	0	1	0	0
	Fibrosarcoma, malignant, secondary	0	0	0	2	0	0	0	0
	Hemangiosarcoma, malignant, primary	0	0	0	0	0	1	0	0
	Leiomyosarcoma, malignant, primary	0	0	0	0	0	1	0	0
	Liposarcoma, malignant, secondary	0	1	0	0	0	0	0	0
	Osteosarcoma, malignant, primary	0	1	0	0	0	0	0	0
Cavity, Oral	Carcinoma, squamous cell, malignant, primary	1	0	0	0	0	0	0	0
Cavity, Thoracic	Fibrosarcoma, malignant, primary	0	0	0	1	0	0	0	0
	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
	Liposarcoma, malignant, secondary	0	1	0	0	0	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice						**Gilead Study No. TOX109** ([redacted] 814-005)			
Dose (mg/kg/day)		**0 (Vehicle)**		**80**		**250**		**750**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Gall Bladder	Adenoma, benign, primary	0	0	1	0	0	0	1	1
Harderian Glands	Adenocarcinoma, malignant, primary	1	2	1	0	0	1	0	1
	Adenoma, benign, primary	4	1	4	3	6	5	4	5
Heart	Hemangiosarcoma, malignant, primary	0	0	0	0	0	1	0	0
Kidneys	Adenoma, tubular cell, benign, primary	1	0	0	0	2	0	0	0
Liver	Adenoma, hepatocellular, benign, primary	5	0	7	0	7	1	4	2
	Carcinoma, hepatocellular, malignant, primary	3	1	4	0	0	1	1	0
	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
	Hemangiosarcoma, malignant, primary	2	0	1	0	2	2	2	0
Lung	Adenocarcinoma, malignant, secondary	0	0	0	1	0	0	0	
	Adenoma, bronchiolar alveolar, benign, primary	7	9	14	15	15	8	13	6
	Carcinoma, bronchiolar alveolar, malignant, primary	1	1	2	3	0	1	1	1

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice						**Gilead Study No. TOX109** ([redacted] 814-005)			
Dose (mg/kg/day)		**0 (Vehicle)**		**80**		**250**		**750**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Lung, cont.	Fibrosarcoma, malignant, secondary	0	0	0	2	0	1	0	0
Lymph Node, Axillary	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
Lymph Node, Cervical	Lymphangioma, benign, primary	0	0	0	0	0	0	0	1
Lymph Node, Hepatic	Leiomyosarcoma, malignant, secondary	0	0	0	0	0	1	0	0
Lymph Node, Mediastinal	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
	Liposarcoma, malignant, secondary	0	1	0	0	0	0	0	0
Lymph Node, Mesenteric	Leiomyosarcoma, malignant, secondary	0	0	0	0	0	1	0	0
Lymph Node, Renal	Leiomyosarcoma, malignant, secondary	0	0	0	0	0	1	0	0
Mammary Gland	Adenocarcinoma, malignant, primary	0	0	0	1	0	0	0	1
Mesentery/Peritoneum	Leiomyoma, benign, primary	0	1	0	0	0	0	0	0
Multicentric Neoplasm	Leukemia, granulocytic, malignant, muticentric	0	1	0	0	0	0	0	0
	Lymphoma, malignant, multicentric	4	5	0	7	3	6	4	4
	Sarcoma, histiocytic, malignant, multicentric	0	6	0	2	1	6	1	2

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice						**Gilead Study No. TOX109** ([redacted] 814-005)			
Dose (mg/kg/day)		**0 (Vehicle)**		**80**		**250**		**750**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Ovaries	Cystadenoma, benign, primary	NA	0	NA	1	NA	2	NA	1
	Hemangioma, benign, primary	NA	1	NA	0	NA	0	NA	0
	Leiomyosarcoma, malignant, secondary	NA	0	NA	0	NA	1	NA	0
	Sex-cord/stromal tumor, benign, primary[b]	NA	0	NA	0	NA	0	NA	2
	Sex-cord/stromal tumor, malignant, primary	NA	1	NA	0	NA	1	NA	0
Pancreas	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
	Leiomyosarcoma, malignant, secondary	0	0	0	0	0	1	0	0
	Liposarcoma, malignant, secondary	0	1	0	0	0	0	0	0
Pituitary Gland	Adenoma, pars distalis, benign, primary	0	1	0	2	0	0	0	0
Skeletal Muscle	Leiomyosarcoma, malignant, secondary	0	0	0	0	0	1	0	0
Skin	Carcinoma, squamous cell, malignant, primary	0	0	0	0	0	1	1	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice						**Gilead Study No. TOX109** ([redacted] 814-005)			
Dose (mg/kg/day)		**0 (Vehicle)**		**80**		**250**		**750**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Skin, Subcutis	Fibrosarcoma, malignant, primary	1	1	0	2	1	5	1	1
	Liposarcoma, malignant, primary	0	1	0	0	0	0	0	0
Small Intestine, Ileum	Adenocarcinoma, malignant, primary	0	1	0	0	0	0	0	0
Spleen	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
	Hemangiosarcoma, malignant, primary[c]	0	0	1	1	2	0	3	1
Testes	Adenoma, interstitial cell, benign, primary	0	NA	0	NA	2	NA	1	NA
Thymus Gland	Fibrosarcoma, malignant, secondary	0	0	0	1	0	0	0	0
Thyroid Gland	Adenoma, c-cell, benign, primary	0	1	0	0	0	0	0	0
	Adenoma, follicular cell, benign, primary	0	1	0	0	0	0	0	0
Tooth/Teeth	Odontoma, benign, primary	0	0	1	0	0	0	0	0
Urinary Bladder	Leiomyosarcoma, malignant, secondary	0	0	0	0	0	1	0	0
	Mesenchymal tumor, benign, primary	0	1	0	0	0	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD-1 Mice						**Gilead Study No. TOX109** ([redacted] 814-005)			
Dose (mg/kg/day)		**0 (Vehicle)**		**80**		**250**		**750**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Uterus with Cervix	Adenocarcinoma, malignant, primary	NA	0	NA	3	NA	1	NA	0
	Carcinoma, squamous cell, malignant, primary	NA	0	NA	1	NA	0	NA	0
	Hemangiosarcoma, malignant, primary	NA	0	NA	1	NA	0	NA	1
	Leiomyoma, benign, primary	NA	4	NA	4	NA	1	NA	2
	Leiomyosarcoma, malignant, primary	NA	0	NA	0	NA	0	NA	1
	Leiomyosarcoma, malignant, secondary	NA	0	NA	0	NA	1	NA	0
	Polyp, glandular, benign, primary	NA	1	NA	1	NA	0	NA	0
	Polyp, stromal, benign, primary	NA	7	NA	5	NA	2	NA	3
	Sarcoma, stromal, malignant, primary	NA	0	NA	3	NA	1	NA	1
Vagina	Polyp, stromal, benign, primary	NA	1	NA	0	NA	0	NA	0

a At 6 months; for controls group means are shown. For treated groups percent differences from control are shown.
b Peto Test; P-value 0.040.
c Peto Test; P-value 0.031.
NA = not applicable; nd = not determined; * = sample taken at Week 104, concentrations were generally below level of quantitation.
Noteworthy Findings: Gross Pathology, None; Microscopic Pathology – ; Non-neoplastic lesions, None

10.2. FTC: Rat

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats		**Gilead Study No. TOX108 (** 814-006)
Species/ Strain: Rat/ CD® [Crl:CD® (SD) IGS BR]	**Duration of Dosing:** 104 Weeks	
Initial Age: 6 Weeks	**Method of Administration:** Oral gavage	**GLP Compliance:** Yes
Date of First Dose: [redacted]	**Vehicle/Formulation:** 0.5% Methylcellulose	**Lot No.:** [redacted]; [redacted]
Special Features: None	**Basis for High-Dose Selection:** Results from 90 day rat study; toxicokinetic endpoint	

Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Toxicokinetics									
AUC_{0-24} (µg·h/mL) (Week 26)		nd*	nd*	42.87	52.53	137.42	170.68	326.77	404.07
Number of animals									
At Start		60	60	60	60	60	60	60	60
Died/Sacrificed Moribund		31	36	40	28	34	31	32	40
Terminal Sacrifice		29	24	20	32	26	29	28	20
Survival (%)		48	40	33	53	43	48	47	33
Body Weight[a] (%)		584 g	339 g	0	-4	+2	0	+3	+1
Food Consumption[a] (%)		26 g/day	20 g/day	+4	0	+8	+5	+8	+10
Number of Animals with Neoplastic Lesions									
Adipose Tissue, Brown	Hibernoma, benign, primary	1	0	1	0	0	0	0	0
	Hibernoma, malignant, primary	0	1	0	0	0	1	0	1
Adrenal Glands	Adenoma, cortical, benign, primary	0	1	0	1	1	0	1	1
	Carcinoma, cortical, malignant, primary	0	3	1	1	0	0	0	0
	Pheochromocytoma, benign, primary	4	1	7	1	12	1	7	2
	Pheochromocytoma, malignant, primary	0	0	1	0	0	1	0	1

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats						**Gilead Study No. TOX108 (** [redacted] 814-006)			
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Bone, Tibia	Hemangiosarcoma, malignant, secondary	0	0	1	0	0	0	0	0
Brain	Astrocytoma, malignant, primary	2	0	0	0	1	1	0	0
	Carcinoma, pars distalis, malignant, secondary	0	0	0	0	1	0	0	0
	Carcinoma, zymbals gland, malignant, secondary	0	0	0	0	1	1	0	0
	Granular cell tumor, benign, primary	0	1	1	0	0	1	0	0
	Reticulosis, malignant, primary	0	1	0	0	0	0	0	0
Cavity, Abdominal	Carcinoma, malignant, primary	0	0	0	1	0	0	0	0
	Fibrosarcoma, malignant, primary	1	0	0	0	0	0	0	0
	Lipoma, benign, primary	0	0	0	0	0	0	1	1
	Neuroendocrine tumor, benign, primary	1	0	0	0	0	0	0	0
Cavity, Thoracic	Hibernoma, benign, primary	0	1	0	0	0	0	0	0
	Osteosarcoma, malignant, primary	0	0	0	0	1	0	0	0
Coagulating Glands	Adenoma, benign, primary	0	0	0	0	0	0	1	0
Ears	Fibroma, benign, primary	1	0	0	0	0	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats								**Gilead Study No. TOX108 (** [redacted] 814-006)	
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Hard Palate	Fibrosarcoma, malignant, primary	0	0	1	0	0	0	0	0
Harderian Glands	Carcinoma, zymbals gland, malignant, secondary	1	0	0	0	0	0	0	0
Implantation Site	Fibrosarcoma, malignant, primary	0	0	0	0	0	0	1	0
Heart	Adenocarcinoma, malignant, secondary	0	1	0	0	0	0	0	0
Kidneys	Carcinoma, tubular cell, malignant, primary	0	0	1	0	0	1	0	0
	Lipoma, benign, primary	0	1	0	0	0	0	0	0
	Liposarcoma, malignant, primary		1	1	0	0	0	1	0
Large Intestine, Cecum	Fibrosarcoma, malignant, secondary	1	0	0	0	0	0	0	0
Large Intestine, Rectum	Granular cell tumor, malignant, secondary	0	0	0	1	0	1	0	0
Liver	Adenoma, hepatocellular, benign, primary	1	3	0	0	0	1	1	2
	Carcinoma, hepatocellular, malignant, primary	0	0	0	1	0	0	0	0
Lung	Adenoma, bronchiolar alveolar, benign, primary	1	0	0	0	0	0	0	1
	Hibernoma, malignant, secondary	0	1	0	0	0	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats						**Gilead Study No. TOX108 (** [redacted] 814-006)			
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Lung, cont.	Carcinoma, zymbals gland, malignant, secondary	1	0	0	0	0	0	0	0
Lymph Node, Cervical	Carcinoma, c-cell, malignant, secondary	0	0	0	0	0	0	1	0
Lymph Node, Mediastinal	Adenocarcinoma, malignant, secondary	0	1	0	0	0	0	0	0
Lymph Node, Mesenteric	Hemangiosarcoma, malignant, primary	0	0	0	1	0	0	0	0
Mammary Gland	Adenocarcinoma, malignant, primary	0	18	1	9	1	13	0	7
	Adenoma, benign, primary	0	3	0	2	0	3	0	1
	Fibroadenoma, benign, primary	0	22	2	17	1	18	0	16
Multicentric Neoplasm	Lymphoma, malignant, multicentric	1	1	2	0	1	2	1	0
	Sarcoma, histiocytic, malignant, multicentric	0	1	2	0	0	1	1	2
Nose, Level B	Carcinoma, zymbals gland, malignant, secondary	1	0	0	0	0	1	0	0
	Lymphangioma, benign, primary	0	0	0	0	1	0	0	0
Nose, Level C	Carcinoma, zymbals gland, malignant, secondary	1	0	0	0	0	1	0	0
	Papilloma, benign, primary	0	0	0	0	1	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats						**Gilead Study No. TOX108 (** [redacted] 814-006)			
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Nose, Level D	Carcinoma, zymbals gland, malignant, secondary	1	0	0	0	0	1	0	0
	Polyp, benign, primary	0	0	0	0	1	0	0	0
Ovaries	Sex-cord/stromal tumor, benign, primary	NA	1	NA	0	NA	1	NA	0
Pancreas	Adenoma, acinar cell, benign, primary	0	0	0	0	0	0	1	0
	Adenoma, islet cell, benign, primary	2	3	3	0	3	0	3	2
	Carcinoma, islet cell, malignant, primary	0	0	1	0	1	0	1	0
	Carcinoma, malignant, secondary	0	0	0	1	0	0	0	0
	Fibroma, benign, primary	0	0	0	0	0	0	0	1
	Fibrosarcoma, malignant, secondary	1	0	0	0	0	0	0	0
Parathyroid Glands	Adenoma, benign, primary	0	0	0	0	4	1	0	0
	Carcinoma, pars distalis, malignant, primary	0	0	0	0	1	1	0	1
Seminal Vesicles	Carcinoma, squamous cell, malignant, primary	0	NA	0	NA	1	NA	0	NA
	Fibrosarcoma, malignant, secondary	1	NA	0	NA	0	NA	0	NA

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats						**Gilead Study No. TOX108 (** [redacted] 814-006)			
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Skin	Carcinoma, squamous cell, malignant, primary	1	0	1	0	0	0	0	1
	Hemangioma, benign, primary	0	0	0	0	1	0	0	0
	Papilloma, squamous cell, benign, primary	0	0	2	0	1	0	0	0
	Trichoepithelioma, benign, primary	0	0	1	0	1	0	0	0
Skin, Subcutis	Fibroma, benign, primary	0	1	2	2	4	0	2	0
	Fibrosarcoma, malignant, primary	1	1	0	0	0	0	0	1
	Hemangiosarcoma, malignant, primary	0	0	1	0	0	0	0	0
	Hibernoma, benign, primary	0	1	0	0	0	0	0	0
	Lipoma, benign, primary	0	1	0	1	0	0	1	0
	Schwannoma, malignant, primary	1	0	0	0	1	0	0	0
Small Intestine, Duodenum	Carcinoma, malignant, secondary	0	0	0	1	0	0	0	0
Spleen	Carcinoma, malignant, secondary	0	0	0	1	0	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats						**Gilead Study No. TOX108 (** [redacted] 814-006)			
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Stomach, Glandular	Fibrosarcoma, malignant, secondary	1	0	0	0	0	0	0	0
	Osteosarcoma, malignant, primary	0	0	1	0	0	0	0	0
Testes	Adenoma, interstitial cell, benign, primary	0	NA	0	NA	1	NA	1	NA
Thymus gland	Lymphoma, malignant, primary	0	0	0	1	0	0	0	0
	Thymoma, benign, primary	0	0	1	0	0	0	0	0
	Thymoma, malignant, primary	1	0	0	0	1	0	0	0
Thyroid Gland	Ademoma, c-cell, benign, primary	3	5	2	2	7	3	0	1
	Adenoma, follicular cell, benign, primary	7	1	0	0	4	0	1	3
	Carcinoma, follicular cell, malignant, primary	0	1	0	0	0	0	0	0
	Carcinoma, c-cell, malignant, primary	1	0	1	0	0	0	1	0
Tongue	Carcinoma, squamous cell, malignant, primary	0	0	0	1	0	0	0	0
Urinary Bladder	Carcinoma, squamous cell, malignant, primary	0	0	1	0	0	0	0	0
	Carcinoma, squamous cell, malignant, secondary	0	0	0	0	1	0	0	0
	Carcinoma, transitional cell, malignant, primary	0	0	0	0	1	0	0	0

Test Article: FTC

Report Title: TP-0006: Two-Year Oral Oncogenicity Study in CD Rats						**Gilead Study No. TOX108 (** [redacted] 814-006)			
Dose (mg/kg/day)		**0 (Vehicle)**		**60**		**200**		**600**	
Gender:No. of Animals		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Uterus with Cervix	Adenocarcinoma, malignant, primary	NA	0	NA	1	NA	0	NA	1
	Granular cell tumor, benign, primary	NA	6	NA	6	NA	4	NA	2
	Granular cell tumor, benign, secondary	NA	1	NA	0	NA	0	NA	0
	Granular cell tumor, malignant, secondary	NA	0	NA	0	NA	1	NA	0
	Leiomyoma, benign, primary	NA	0	NA	0	NA	1	NA	0
	Polyp, stromal, benign, primary	NA	6	NA	3	NA	4	NA	6
	Sarcoma, stromal, malignant, primary	NA	0	NA	0	NA	1	NA	1
	Schwannoma, malignant, primary	NA	0	NA	0	NA	0	NA	1
Vagina	Granular cell tumor, benign, primary	NA	1	NA	2	NA	4	NA	1
	Granular cell tumor, malignant, primary	NA	0	NA	0	NA	1	NA	0
	Polyp, stromal, benign, secondary	NA	0	NA	0	NA	1	NA	0
	Sarcoma, stromal, malignant, secondary	NA	0	NA	0	NA	0	NA	1
Zymbal's Gland	Carcinoma, zymbals gland, malignant, primary	1	0	1	0	1	1	0	0

a At 6 months; for controls group means are shown. For treated groups percent differences from control are shown.
NA = not applicable; nd = not determined; * = sample taken at Week 104, concentrations were below level of quantitation

10.3. RPV: Mouse

Test Article: RPV

Report Title: Carcinogenicity Study by Oral Gavage Administration to CD-1 Mice for 104 Weeks

Study no.: TMC278-NC120

Species/strain: Mouse/Crl:CD-1™ (ICR)

Age at first dose: 41 to 48 days

Duration of dosing: 104 weeks

Duration of postdose: none

Administration route/method: Oral/gavage

Doses: 0, 20, 60, 160 mg base eq./kg/day TMC278.HCl

Test article batch: [redacted]

Vehicle/formulation: 0.5% w/v Methocel/Suspension

GLP compliance: Yes

No Observed Adverse Effect Level: Not established

Toxicokinetics

Dose (mg eq./kg/day)	0 (vehicle) [a]		20		60		160	
No. of animals	**M:18**	**F:18**	**M:24**	**F:24**	**M:24**	**F:24**	**M:24**	**F:24**
AUC (µg·h/mL)								
Day 1 ($AUC_{0-\infty}$)	/	/	59	61	239	182	440	345
Week 28 (AUC_{0-24h})	/	/	76	51	230	278	505	766

a TMC278 plasma concentrations above the lower limit of quantification (0.002 µg/mL) were found in 10 out of 42 samples (ranging between 0.002 and 0.012 µg/mL)

Noteworthy findings

Dose (mg eq./kg/day)	0 (vehicle)		20		60		160	
No. of animals	**M:60**	**F:60**	**M:60**	**F:60**	**M:60**	**F:60**	**M:60**	**F:60**
Died or Sacrified Moribund	25	40	35	37	39	30	42	43
Body Weight[b]	48.4	37.8	1.02	1.05	1.03	1.09	1.01	1.05
Body Weight Gain[b]	15.4	13.6	1.06	1.13	1.10	1.23*	1.07	1.17*
Food Consumption[b]	35	35	1.06	1.00	1.09	1.06	1.23	1.14
Clinical Observations	-	-	-	-	-	-	-	-
Hematology	-	-	-	-	-	-	-	-
Organ Weights								
Liver (absolute)	2.334	-	1.35**	-	1.69**	-	1.18**	-
Liver (adjusted)	-	1.999	-	1.13	-	1.39**	-	1.94**

Test Article: RPV

Study: TMC278-NC120: Carcinogenicity Study by Oral Gavage Administration to CD-1 Mice for 104 Weeks (continued)

Noteworthy findings

Dose (mg eq./kg/day)	0 (vehicle)	20	60	160

No. of animals	M:60	F:60	M:60	F:60	M:60	F:60	M:60	F:60
Gross Pathology								
Liver:								
Enlarged	2	7	3	7	2	10	11	30
Masses	13	4	21	9	23	13	29	32
Pale area(s)	1	6	4	9	6	10	12	33
Lungs:								
Mass(es)	23	-	21	-	26	-	30	-
Kidney:								
Left kidney								
Granular	-	6	-	6	-	4	-	15
Right Kidney								
Granular	-	6	-	6	-	4	-	14

b At end of dosing period. For vehicle, group means are shown. For treated groups, multiples of control/baseline are shown. Statistical significance is based on actual data (not on the multiples of control/baseline). - No noteworthy findings, * - $p<0.05$ ** - $p<0.01$; F: female; M: male

Test Article: RPV

Study: TMC278-NC120: Carcinogenicity Study by Oral Gavage Administration to CD-1 Mice for 104 Weeks (continued)

Dose (mg eq./kg/day)	**0 (vehicle)**		**20**		**60**		**160**	
No. of animals	**M:60**	**F:60**	**M:60**	**F:60**	**M:60**	**F:60**	**M:60**	**F:60**
Histopathology[a]								
Liver (n°. examined)	60	60	60	60	60	60	60	60
- M-Hepatocellular carcinoma	4	0	7	0	7	2	9	7
- B-Hepatocellular adenoma	8	1	14	0	21	8	19	16
- Eosinophilic foci	0	0	0	0	1	1	3	8
- Basophilic foci	0	0	4	0	0	2	5	10
- Clear cell foci	0	0	3	1	1	1	4	4
- Generalised eosinophilic hepatocyte hypertrophy	0	1	1	0	1	11	5	42
- Brown pigmented macrophages associated with mononuclear cells	0	2	0	3	1	7	3	17
- Brown pigmented Kupffer cells/macrophages	0	5	3	3	2	17	9	17
Lungs (n°. examined)	60	60	60	60	60	60	60	60
- M-Bronchioloaveolar adenocarcinomas	6	-	5	-	0	-	10	-
Kidney (n°. examined)	-	60	-	60	-	60	-	60
Kidney (Left)								
- Glomerulonephritis	-	7	-	7	-	6	-	11
- Nephropathy		27		21		23		37
Kidney (Right)								
- Glomerulonephritis	-	7	-	7	-	6	-	11
- Nephropathy	-	22	-	20	-	20	-	36

- No noteworthy findings; F: female; M: male;
a Histopathology findings: M = malignant; B = benign

10.4. RPV: Rat

Test Article: RPV

Report Title: Carcinogenicity Study by Oral Gavage Administration to CD Rats for 104 Weeks

Study No.: TMC278-NC123

Species/strain: Rat/Crl: CD/Sprague Dawley

Age at first dose: approximately 42 days

Date of first dose: [redacted]

Duration of dosing: 104 Weeks (males),
98 weeks (Group 1, 3, 4 and 5 females),
97 weeks (Group 2 females)

Duration of postdose: None

Administration route/method: Oral/Gavage

Doses: 0, 40, 200, 500, 1500 mg base eq./kg/day TMC278.HCl

Test article batch: [redacted]

Vehicle/formulation: 0.5% w/v Methocel in purified water/suspension

GLP compliance: Yes

No Observed Adverse Effect Level: /

Toxicokinetics:

Dose (mg base eq./kg/day)	0 (vehicle) [b]		40		200		500		1500	
No. of Animals	**M:6**	**F:6**	**M:9**	**F:9**	**M:9**	**F:9**	**M:9**	**F:9**	**M:9**	**F:9**
AUC (µg·h/mL) [a]										
Day 1	/	/	19	17	34	40 [c]	45	63	58	81
Week 27	/	/	4.5	15	11	36	14	70	20	81
Week 39	/	/	6.3	14	8.2	41	14	46	18	84

a $AUC_{0-\infty}$ on Day 1 otherwise AUC_{0-24h}; [b] on Day 1, 2 samples out of 18 were above the limit of quantification (LOQ, 0.001-0.002 µg/mL), at Week 27, 17 samples out of 18 were above LOQ and at Week 39, 7 samples out of 18 were above the LOQ; [c] AUC_{0-24h}.

Noteworthy Findings:

Dose (mg base eq./kg/day)	0 (vehicle)		40		200		500		1500	
No. of Animals	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**
Died or Sacrificed Moribund	40	45	40*	46	38	35	37	33	44	34
Body Weight	-	-	-	-	-	-	-	-	-	-
Body Weight Gain	-	-	-	-	-	-	-	-	-	-
Food consumption	-	-	-	-	-	-	-	-	-	-
Ophthalmoscopy	-	-	-	-	-	-	-	-	-	-

* includes one animal that was found dead during the necropsy period (Week 105), - No noteworthy findings; F: female; M: male

Test Article: RPV

Study: TMC278-NC123: Carcinogenicity Study by Oral Gavage Administration to CD Rats for 104 Weeks (continued)

Dose (mg base eq./kg/day)	0 (vehicle)	40	200	500	1500

No. of Animals	M:65	F:65	M:65	F:65	M:65	F:65	M:65	F:65	M:65	F:65
Clinical Observations										
- Encrustations, head	-	9	-	13	-	14	-	15	-	19
- Brown staining, head	-	19	-	13	-	14	-	22	-	31
- Brown staining, dorsal body surface	7	-	9	-	6	-	10	-	15	-
- Yellow staining, ventral body surface	4	-	4	-	1	-	2	-	14	-
- Hairloss, forelimbs	1	6	6	9	2	10	4	9	10	12
- Hairloss, head	-	22	-	22	-	20	-	24	-	33
- Ungroomed	12	-	17	-	21	-	20	-	23	-
Palpable swellings	-	-	-	-	-	-	-	-	-	-
Hematology	-	-	-	-	-	-	-	-	-	-
Serum Chemistry – Week 52 of treatment[a]										
- Alkaline phosphatase U/L	53	24	1.06	1.04	1.13	1.21	1.42**	1.08	1.92**	1.29*
- Alanine aminotransferase U/L	43	-	1.02	-	1.02	-	1.49**	-	2.63**	-
- Aspartate aminotransferase U/L	74	-	1.16	-	1.09	-	1.50*	-	1.92**	-
- gamma glutamyltransferaseGT U/L	1	-	1.00	-	2.00	-	2.00*	-	2.00*	-
- Urea mmol/L	4.10	-	0.94	-	1.01	-	1.08	-	1.14**	-
- Creatinine µmol/L	41	-	1.02	-	1.12*	-	1.22**	-	1.20**	-
- Cholesterol mmol/L	2.45	2.99	1.00	0.89	0.91	0.86*	0.84	0.80**	0.91	0.77**
- Triglycerides mmol/L	1.71	2.12	0.84	0.98	0.73*	0.78	0.80*	0.57**	0.76*	0.57**
- Chloride mmol/L	99	-	1.00	-	1.00	-	1.00	-	0.98**	-
- Calcium mmol/L	2.69	2.80	0.99	0.99*	0.99*	0.97**	0.98**	0.97**	0.97**	0.96**
- Albumin g/L	31	-	1.00	-	1.03	-	1.06**	-	1.10**	-
- a1 g/L	-	14	-	0.93*	-	0.86**	-	0.86**	-	0.93**
- a2 g/L	4	-	0.75**	-	0.75**	-	0.75**	-	0.50**	-
- Beta g/L	17	15	0.94	1.00	0.94*	0.93*	0.88**	0.93**	0.88**	0.93**
- A/G ratio	0.74	1.13	1.05	1.01	1.08*	1.07	1.19**	1.13**	1.18**	1.06**

a At end of dosing period. For vehicle, group means are shown. For treated groups, multiples of control/baseline are shown. Statistical significance is based on actual data (not on the multiples of control/baseline), - No noteworthy findings, * - $p<0.05$, ** - $p<0.01$, *** - $p<0.001$; F: female; M: male

Test Article: RPV

Study: TMC278-NC123: Carcinogenicity Study by Oral Gavage Administration to CD Rats for 104 Weeks (continued)

Dose (mg base eq./kg/day)	0 (vehicle)		40		200		500		1500	
No. of Animals	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**
Serum Chemistry - TERM[a]										
- Alkaline phosphatase U/L	58	-	1.10	-	1.41*	-	1.81**	-	2.19**	-
- Creatinine µmol/L	33	40	1.06	1.18**	1.21#	0.98	1.21**	1.15*	1.15**	1.33**
- Glucose mmol/L	5.72	-	1.11		1.15		1.11		1.19**	
- Cholesterol mmol/L	3.42	-	0.98	-	0.88*	-	0.82*	-	0.85*	-
- Triglycerides mmol/L	-	1.27	-	1.23	-	0.86	-	0.83	-	0.72
- Potassium mmol/L	-	3.7	-	1.08	-	1.08	-	1.11*	-	1.19**
- Sodium mmol/L	143	-	1.01	-	1.01	-	1.00	-	0.99*	-
- Chloride mmol/L	100	-	1.00	-	1.02**	-	1.02**	-	1.02**	-
- Calcium mmol/L	-	2.83	-	0.98*	-	0.98	-	0.98*	-	0.98*
- Phosphorus mmol/L	-	1.41	-	1.06	-	0.96	-	0.99	-	1.15**
- Albumin g/L	30	-	1.00	-	0.97	-	1.03	-	1.07**	-
- Beta g/L	21	-	0.90*	-	0.90*	-	0.86**	-	0.81**	-
- A/G ratio	0.64	-	1.06	-	1.02	-	1.11*	-	1.22**	-
Urinalysis - Week 51 of treatment[a]										
- pH	-	6.9	-	0.94	-	0.96	-	1.01	-	0.87**
- Protein g/L	-	0.27	-	0.59	-	0.89	-	0.56	-	1.11**
- Potassium mmol/L	-	160.4	-	0.94	-	0.86	-	0.86	-	0.81*
- Chloride mmol/L	45.1	31.6	1.10	1.13	1.14	1.59**	1.80**	2.75**	3.63**	4.80**
Urinalysis – TERM										
- pH	7.9	6.6	1.03	0.97	1.03	1.00	1.00	1.03	0.90**	0.91**
- Potassium mmol/L	130.7	107.2	0.98	1.06	0.81*	0.97	0.80*	0.82**	0.85*	0.85**
- Chloride mmol/L	42.2	34.9	0.95	1.01	1.29	1.47**	1.47**	1.94**	3.48**	3.46**
Organ Weights, Unadjusted (absolute)										
Thyroid and Parathyroid	0.033	0.027	-	-	-	-	-	-	1.64*	1.37**

a At end of dosing period. For vehicle, group means are shown. For treated groups, multiples of control/baseline are shown. Statistical significance is based on actual data (not on the multiples of control/baseline), TERM: End of treatment period, - : No noteworthy findings, # : With exclusion of 3M, value 148 µmol/L, *: $p<0.05$, ** : $p<0.01$, *** : $p<0.001$; F: female; M: male

Test Article: RPV

Study: TMC278-NC123: Carcinogenicity Study by Oral Gavage Administration to CD Rats for 104 Weeks (continued)

Dose (mg base eq./kg/day)	0 (vehicle)		40		200		500		1500	
No. of Animals	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**
Gross Pathology										
Number Examined:	65	65	65	65	65	65	65	65	65	65
- Lungs and Bronchi, pale area(s)	22	17	37	25	31	23	31	28	40	42
- Liver, dark area(s)	12	-	12	-	20	-	11	-	17	-
- Liver, mass(es)	-	5	-	11	-	8	-	10	-	7
- Thyroids, mass(es)	2	-	6	-	6	-	3	-	4	-
- Thyroids, enlarged	-	2	-	3	-	3	-	4	-	12
- Abnormal contents (pale/ white) of GI Tract	6	4	9	6	18	14	15	13	32	19
Histopathology										
Liver (n°. examined)	65	65	65	65	65	65	65	65	65	65
- Hepatocelluar adenoma	0	1	1	5	3	5	3	6	3	4
- Sinuosoidal dilatation:										
Minimal	0	11	-	6	-	13	-	13	-	12
Slight	0	8	-	22	-	19	-	14	-	28
Moderate	0	6	-	2	-	4	-	4	-	3
Total	0	25	-	30	-	36	-	31	-	43
Thyroid (n°. examined)										
- Follicular cell adenoma	1	0	3	0	4	3	5	3	5	4
- Follicular cell carcinoma	0	0	0	1	2	1	1	0	2	1
- Follicular cell hypertrophy:										
Minimal	1	0	0	1	2	3	1	1	1	6
Slight	0	0	1	1	2	1	3	1	3	2
Total	1	0	1	2	4	4	4	2	4	8
- Cystic follicular cell hyperplasia:										
Minimal	3	0	1	1	3	2	0	2	5	2
Slight	3	0	4	1	2	0	2	3	1	3
Moderate	0	0	0	0	0	0	0	0	1	0
Total	6	0	5	2	5	2	2	5	7	5

- : No noteworthy findings ; F: female; M: male

Test Article: RPV

Study: TMC278-NC123: Carcinogenicity Study by Oral Gavage Administration to CD Rats for 104 Weeks (continued)

Dose (mg base eq./kg/day)	0 (vehicle)		40		200		500		1500	
No. of Animals	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**
Histopathology (cont'd)										
Nasal turbinates (n°. examined)	65	65	65	65	65	65	65	65	65	65
- Inflammatory exudate	1	0	2	0	2	0	3	1	39	26
- Respiratory epithelium hyperplasia:										
Minimal	0	0	0	0	0	0	0	1	8	4
Slight	0	0	0	0	1	0	0	0	11	15
Moderate	0	0	0	0	0	0	0	0	5	2
Total	0	0	0	0	1	0	0	1	24	21
- Respiratory epithelium inflammation:										
Minimal	2	0	0	0	0	0	0	1	5	2
Slight	0	0	0	0	0	0	1	0	17	11
Moderate	0	0	0	0	0	0	0	0	3	4
Total	2	0	0	0	0	0	1	1	25	17
- Olfactory epithelium respiratory metaplasia:										
Minimal	0	0	0	0	0	0	0	0	0	2
Slight	0	0	0	0	0	0	0	0	5	7
Moderate	0	0	0	0	0	0	0	0	3	2
Total	0	0	0	0	0	0	0	0	8	11
Lungs (n°. examined)	65	65	65	65	65	65	65	65	65	65
- Foamy alveolar macrophages:										
Minimal	31	31	32	32	33	38	30	36	24	35
Slight	11	4	18	3	11	3	16	9	19	12
Moderate	2	0	1	2	2	1	1	0	2	6
Marked	0	0	0	0	1	0	0	0	0	0
Total	44	35	51	37	47	42	47	45	45	53

F: female; M: male

Test Article: RPV

Study: TMC278-NC123: Carcinogenicity Study by Oral Gavage Administration to CD Rats for 104 Weeks (continued)

Dose (mg base eq./kg/day)	**0 (vehicle)**		**40**		**200**		**500**		**1500**	
No. of Animals	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**	**M:65**	**F:65**
Histopathology (cont'd)										
Lungs (n°. examined)	65	65	65	65	65	65	65	65	65	65
- Alveolar epithelial hyperplasia:										
Minimal	15	9	20	9	15	8	13	16	15	17
Slight	4	2	7	2	5	4	3	3	7	11
Moderate	0	0	0	0	1	0	0	0	0	0
Total	19	11	27	11	21	12	16	19	22	28

F: female; M: male

10.5. TDF: Mouse

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Mouse		**Gilead Study No.** M990205
Species/ Strain: Mouse, Swiss Crl:CD^{R}-1(ICR)BR	**Duration of Dosing:** 104 Weeks	
Initial Age: 6 Weeks (approx)	**Method of Administration:** Oral gavage	**GLP Compliance:** Yes
Date of First Dose: [redacted] (Main)	**Vehicle/ Formulation:** Suspension vehicle	**Lot No.:** [redacted]

Basis for High-Dose Selection: The dose levels were selected according to the potential human exposure, existing toxicity data and limitations imposed by the test article. A previous 13 week oral toxicity study indicated that 600 mg/kg/day (60 mg/mL) is the highest dose level of TDF that may be practically administered to albino mice.

Special Features: Separate populations for toxicokinetic evaluations were included in the study: Groups of 30 males and 30 females were sampled on Days 1 and 180 for the high dose only. Animals were euthanized on completion of the blood collection & subjected to necropsy. Before assignment of animals to a treatment group, 10 males & 10 females animals, selected randomly, were subjected to a health screen.

Dose (mg/kg/day)		**0 (Vehicle)**		**100**		**300**		**600**	
		M: 60	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Toxicokinetics									
AUCss 0-24 (µg·h/mL) Day 1		-	-	-	-	-	-	30.9	35.1
AUCss 0-24 (µg·h/mL) Day 180		-	-	-	-	-	-	44.3	50.9
Number of Animals									
At Start		60	60	60	60	60	60	60	60
Died/Sacrificed Moribund		33	39	37	32	31	29	40	43
Terminal Sacrifice		27	21	23	28	29	31	20	17
Survival (%)		45	35	38	47	48	52	33	28
Body Weight (%)[a]		38.65g	30.79 g	-4.4*	+1.5 %	-3.4	-1.6%	-3.7%	-0.07%
Food Consumption (%)[a]		37.4g	39.4g	+8.6 %	+2.2 %	+5.1%	+0.3 %	+9.9%	-0.3%
Abdomen	Leiomyosarcoma	-	-	1	-	-	-	-	1
	Osteosarcoma	-	-	-	1	-	-	-	-
	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
	Sarcoma	-	-	-	1	-	-	-	-
Adrenal	Adenoma: cortical	-	1	-	1	1	1	-	-
	Sarcoma: fusiform cell	1		-	1	-	-	-	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Mouse						**Gilead Study No.** M990205			
Dose (mg/kg/day)		**0 (Vehicle)**		**100**		**300**		**600**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions									
Adrenal, cont.	Benign pheochromocytoma	-	-	-	2	-	-	-	-
	Sarcoma (metastasis)	-	-	-	1	-	-	-	-
Bone - Femur	Hemangiosarcoma	-	-	1	-	-	-	-	-
Bone - Misc	Hemangiosarcoma	1	-	-	-	-	-	-	-
	Osteosarcoma	-	-	-	1	-	-	-	-
	Rhabdomyosarcoma	-	-	-	-	-	1	-	-
Brain	Malignant meningioma	1	-	-	-	-	-	-	-
Cecum	Leiomyoma	-	-	-	-	-	1	-	-
Diaphragm	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
	Sarcoma(metastasis)	-	-	-	1	-	-	-	-
Duodenum	Adenoma	-	-	-	-	-	-	-	1
	Adenocarcinoma	-	-	-	-	-	-	1	1
Epididmydis	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
Eye	Meningioma	1	-	-	-	-	-	-	-
	Carcinoma (metastasis)	1	-	-	-	-	-	-	-
Fat	Leiomysarcoma (metastasis)	-	1	-	-	-	-	-	-
Harderian Gland	Adenoma	1	2	5	-	3	3	7	3
Head	Hemangiosarcoma	-	-	1	-	-	-	-	-
Hemolym Tissue	Malignant lymphoma	3	10	1	17	3	4	2	5
	Histiocytic sarcoma	-	9	1	4	-	5	1	1
Jejunum	Adenoma	-	-	-	-	1	-	-	-
	Adenocarcinoma	-	-	-	1	1	-	-	1
Kidney	Carcinoma: tubular cell	-	-	1	-	-	-	-	-
	Sarcoma (metastasia)	-	-	-	1	-	-	-	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Mouse								**Gilead Study No.** M990205	
Dose (mg/kg/day)		**0 (Vehicle)**		**100**		**300**		**600**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Liver	Adenoma: hepatocellular	5	1	1	1	2	-	4	9
	Carcinoma: hepatocellular	6	-	2	-	-	-	-	-
	Hemangioma	1	-	-	-	-	-	2	-
	Hemangiosarcoma	2	-	1	-	-	2	-	1
	Leiomyosarcoma (metastasis)	-	1	1	-	-	-	-	-
	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
	Carcinoma: metastasis	1	-	-	-	-	-	-	-
Lung	Carcinoma: metastasis	1	1	-	-	-	1	-	-
	Adenoma: alveolar/bronchiolar	10	10	8	9	11	5	10	7
	Adenocarcinoma: Alveolar/bronchiolar	9	5	7	2	9	2	9	1
	Mammary carcinoma (metastasis)	-	1	-	-	-	-	-	-
	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
	Sarcoma (metastasis)	-	-	-	1	-	-	-	-
	Carcinosarcoma (metastasis)	1	-	-	-	-	-	-	1
	Fibroma	-	-	-	-	-	1	-	-
Lymph Node	Hemangioma	1	-	-	-	-	-	-	-
	Carcinoma (metastasis)	1	-	-	-	-	-	-	-
	Mammary Carcinoma (metastasis)	-	1	-	-	-	-	-	-
Lymph Node Mesenteric	Hemangioma	1	1	-	-	-	-	-	-
	Rhabdomyosarcoma	-	-	1	-	-	-	-	-
Mammary Gland	Adenocarcinoma	-	3	-	-	-	-	-	-
	Adenoacanthoma	-	1	-	-	-	1	-	-
Muscle Skeletal	Hemangioma	-	1	-	-	-	-	-	-
Muscle Skeletal Misc.	Rhabdomyosarcoma	-	-	1	-	-	-	-	-
	Sarcoma (Metastasis)	-	-	-	1	-	-	-	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Mouse								**Gilead Study No.** M990205	
Dose (mg/kg/day)		**0 (Vehicle)**		**100**		**300**		**600**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Ovary	Cystadenoma	-	1	-	1	-	-	-	-
	Carcinoma: tubulostromal	-	-	-	1	-	-	-	-
	Malignant granulosa-theca cell tumor	-	1	-	-	-	-	-	-
	Leiomyosarcoma (metastasis)	-	1	-	-	-	-	-	-
	Carcinoma (metastasis)	-	-	-	-	-	1	-	-
Pancreas	Adenocarcinoma (metastasis)	-	-	-	-	1	-	-	-
	Carcinoma (metastasis)	-	-	-	-	-	1	-	-
	Leiomyosarcoma (metastasis)	-	1	-	-	-	-	-	-
	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
Pituitary	Adenoma: pars distalis	1	1	-	3	-	3	-	-
	Adenoma: pars intermedia	-	-	-	-	-	1	-	-
Skin Misc.	Keratoacanthoma	-	-	-	-	-	1	-	-
Spleen	Hemangioma	-	-	1	-	-	-	-	-
	Hemangiosarcoma	1	-	3	-	1	1	-	-
	Sarcoma (metastasis)	-	-	1	1	-	-	-	-
Stomach	Adenoma	1	-	-	-	-	-	-	-
	Carcinoma (metastasis)	-	-	-	-	-	1	-	-
	Papilloma	-	1	-	-	-	1	-	-
	Leiomyosarcoma (metastasis)	-	1	-	-	-	-	-	-
	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	-	-	-
	Sarcoma (metastasis)	-	-	-	1	-	-	-	-
Subcutaneous Tissue	Hemangiosarcoma	1	-	-	-	-	-	-	-
	Fibrosarcoma	1	-	-	-	-	-	1	-
	Rhabdomyosarcoma (metastasis)	-	-	1	-	-	1		

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Mouse						**Gilead Study No.** M990205			
Dose (mg/kg/day)		**0 (Vehicle)**		**100**		**300**		**600**	
Number Evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Tail	Hemangioma	1	-	-	-	-	1	-	-
Testis	Adenoma: interstitial cell	1	-	1	-	2	-	-	-
	Adenoma: rete testis	2	-	-	-	-	-	-	-
Thyroid	Adenoma: follicular cell	1	-	-	-	-	-	-	-
	Carcinoma: follicular cell	-	1	-	-	-	1	-	-
Urinary Bladder	Carcinoma: transitional cell	1	-	-	-	-	-	-	-
Uterus	Adenocarcinoma: endometrial	-	-	-	-	-	1	-	1
	Carcinosarcoma	-	-	-	-	-	-	-	1
	Sarcoma: endomertrial stromal	-	1	-	-	-	2	-	1
	Leiomyoma	-	-	-	1	-	-	-	2
	Leiomysarcoma	-	2	-	2	-	-	-	-
	Fibroma	-	-	-	1	-	-	-	-
	Carcinoma: squamous cell	-	-	-	-	-	1	-	-
	Benign granular cell Tumor	-	-	-	-	-	1	-	-
	Hemangioma	-	-	-	1	-	-	-	-
	Hemangiosarcoma	-	1	-	2	-	2	-	1
	Sarcoma (metastasis)	-	-	-	1	-	-	-	-
	Polyp: endometrial stromal	-	6	-	5	-	3	-	4
Vagina	Osteosarcoma (metastasis)	-	-	-	1	-	-	-	-
Gross Pathology									
Duodenum	Dilation	3	-	4	3	1	6	9	11
	Thickening	4	1	2	2	3	2	5	9
	Mass	-	-	-	-	-	-	1	2
Jejunum	Dilatation	2	2	5	3	2	7	8	9
Ileum	Dilatation	2	1	5	3	3	5	6	6
Cecum	Dilatation	1	1	3	2	1	2	4	5
Liver	Surface irregular	-	2	1	2	-	5	4	9
	Mass	13	2	4	3	-	2	5	10

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Mouse								**Gilead Study No.** M990205	
Dose (mg/kg/day)		**0 (Vehicle)**		**100**		**300**		**600**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Non-Neoplastic Lesions									
Bone Marrow	Hypercellularity: myeloid	6	11	4	7	8	12	21	20
Duodenum	Hyperplasia: Villous/mucosal	-	2	7	6	26	23	32	27
Kidney	Karyomegaly: tubular	-	-	-	-	-	1	3	8
Liver	Basophilic cell focus	3	1	-	1	-	3	-	4
	Eosinophillic cell focus	2	-	1	2	3	7	8	8
	Hypertrophy; Hepatocellular	9	6	14	6	22	27	39	46
	Cyto/karyomegaly	16	2	9	4	21	27	35	36
	Inclusions: nuclear	4	1	8	-	12	13	33	26
	Necrosis: single cell	7	5	1	2	11	7	22	18
	Proliferation: sinusoidal lining cells	3	2	3	3	7	15	20	25
Testis	Mineralization: vascular	-	-	1	-	6	-	17	-
Tumor analysis									
Liver-hepatocellular adenomata								-	P=0.0037 [b]
Harderian gland								P=0.0098 [b]	-

a At 6 months, for controls group means are shown. For treated groups percent differences from controls are shown. Statistical differences are based upon the actual data not percent differences.
b Pair wise test.
* = $p < 0.05$, Dunnetts "t" test

10.6. TDF: Rat

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Rat		**Gilead Study No.** R990204
Species/ Strain: Rat, Sprague Dawley CD (Crl: CD^{R} (SD) BR)- VAF	**Duration of Dosing:** 104 Weeks	
Initial Age: 6 Weeks (approx)	**Method of Administration:** Oral gavage	**GLP Compliance:** Yes
Date of First Dose: [redacted] (Main)	**Vehicle/ Formulation:** Suspension vehicle	**Lot No.:** [redacted]

Basis for High-Dose Selection: Toxicity-based endpoint. The high dose was based on toxicity (GI tract toxicity, kidney tubular karyomegaly and bone toxicity) observed in a 13/42 week rat study. On day 10 the dose level for group 2 was increased to 300 mg/kg/day.

Special Features: Separate populations for toxicokinetic evaluation were included in the study: Initial TK group (Set 1) comprised of 16 males and 16 females. Blood samples were taken on days 1 & 30. Animals euthanized on day 30 & subjected to necropsy. An additional group of TK animals were added to the study (Group 6, Set 2) to obtain plasma data from Day 1, 30, 60, and 90 at the revised dose level of 300 mg/kg/day. Animals euthanised on day 91 & subjected to necropsy. Additional populations of animals of 24 males and 24 females were added to Groups 1 to 4 to evaluate TK response for a period of 26 Weeks (Set 3). Animals euthanized on day 182 & subjected to necropsy. Before assignment of animals to a treatment group, 10 males & 10 females animals, selected randomly, were subjected to a health screen.

Dose (mg/kg/day)		**0 (Vehicle)**		**10/300[a]**		**30**		**100**	
		M: 60	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Toxicokinetics									
AUCss 0-24 (µg·h/mL) Day 180		-	-	16.69	15.72	2.70	3.06	6.60	7.46
Number of Animals									
At Start		60	60	60	60	60	60	60	60
Died/Sacrificed Moribund		42	44	42	32	42	45[b]	38	43
Terminal Sacrifice		18	16	18	28	18	15	22	17
Survival (%)		30	27	30	47	30	25	37	28
Body Weight (%)		644.8g	363.2g	-5.5%*	-3.7%	+2.0%	+0.6%	-2.9%	-1.1%
Food Consumption (%)		206.4	144.0	-1.5%	+5.6%	-0.2%	+2.8%	-3.5%	+3.9%
Number of Animals with Neoplastic Lesions									
Abdomen	Sarcoma	-	-	-	-	-	-	1	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Rat | **Gilead Study No.** R990204

Dose (mg/kg/day)		**0 (Vehicle)**		**10/300[a]**		**30**		**100**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions, cont.									
Adrenal	Adenoma: cortical	-	2	-	-	1	-	-	-
	Malignant Pheochromocytoma	1	-	-	2	1	-	-	-
	Benign pheochromocytoma	-	-	3	1	1	1	3	-
	Metastasis	-	1	-	-	-	-	-	-
Brain	Malignant astrocytoma	-	1	1	1	3	-	2	1
	Malignant mixed glioma	-	-	-	-	-	-	1	-
	Metastasis	2	2	-	1	-	2	-	1
	Benign granular cell Tumor	-	1	-	-	-	-	-	-
Cavity Cranial	Metastasis	-	-	-	1	-	-	-	-
Clitoral Gland	Carcinoma: squamous cell	-	-	-	-	-	-	-	1
Eye	Malignant schwannoma	-	-	-	1	-	-	-	-
Harderian Gland	Metastasis	-	-	-	1	-	-	-	-
Heart	Metastasis	-	1	-	-	-	-	-	-
	Benign schwannoma: Endocardial	-	1	-	-	-	-	2	-
Hemolym	Malignant lymphoma	-	2	1	-	-	2	-	1
Tissue	Histiocytic sarcoma	2	0	1	1	2	2	2	-
Ileum	Adenocarcinoma	1	-	-	-	1	-	-	-
Jejunum	Adenocarcinoma	-	-	-	-	1	-	-	-
Kidney	Carcinoma: tubular cell	-	-	-	-	1	-	-	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Rat	**Gilead Study No.** R990204

Dose (mg/kg/day)		**0 (Vehicle)**		**10/300[a]**		**30**		**100**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions									
Kidney, cont.	Adenoma: tubular cell	1	-	-	-	-	-	-	-
	Renal mesenchymal tumor	-	-	-	-	1	-	-	-
Liver	Carcinoma: hepatocellular	3	-	1	-	1	-	1	-
	Adenoma: hepatocellular	1	1	2	-	-	-	-	-
	Cholangioma	-	-	-	-	-	-	1	-
	Cholangiocarcinoma	1	-	1	-	-	-	-	-
	Metastasis	1	-	-	-	-	-	-	-
Lung	Carcinoma: metastasis	-	-	1	1	1	2	-	2
	Adenoma: Alveolar/bronchiolar	-	-	2	-	2	-	3	-
	Metastasis	-	1	-	-	-	-	-	-
Lymph node	Metastasis	1	-	-	-	2	-	-	-
Lymph Node Mandibular	Metastasis	-	-	-	1	-	-	-	-
Mammary Gland	Fibroadenoma	1	23	2	25	1	23	-	20
	Adenoma	-	3	-	2	-	1	-	2
	Adenocarcinoma	-	14	-	11	-	13	-	10
Nerve sciatic	Metastasis	-	-	-	1	-	-	-	
Ovary	Benign luteoma	-	1	-	-	-	-	-	-
	Benign granulosa-theca cell tumor	-	-	-	2	-	2	-	2

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Rat | **Gilead Study No.** R990204

Dose (mg/kg/day)		**0 (Vehicle)**		**10/300[a]**		**30**		**100**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions									
Ovary, cont.	Malignant schwannoma	-	-	-	1	-	-	-	-
Pancreas	Adenoma-acinar cell	-	-	-	-	1	-	-	-
	Carcinoma: islet cell	8	3	7	1	10	2	12	6
	Adenoma: islet cell	4	-	6	2	5	2	2	1
Parathyroid Gland	Adenoma	-	-	-	-	1	-	-	-
Pituitary	Carcinoma: pars distalis	3	2	-	2	-	2	-	1
	Adenoma: pars distalis	35	52	25	39	30	47	28	42
	Adenoma: pars intermedia	3	-	-	1	-	-	-	-
Skin Misc.	Papilloma: squamous cell	1	-	1	-	3	-	-	-
	Carcinoma: squamous cell	1	2	1	-	1	-	1	1
	Keratoacanthoma	-	-	1	1	1	-	4	-
	Adenoma: sebaceous	-	-	-	-	-	-	1	-
	Carcinoma: basal cell	-	1	1	-	-	-	2	-
	Adenoma: basal cell	-	-	-	-	-	-	1	-
	Fibrosarcoma	-	-	-	-	1	-	-	-
	Histiocytoma	-	1	-	-	-	-	-	-
Spleen	Hemangioma	-	-	-	-	1	-	-	-
Spinal Cord, Cervical	Benign astrocytoma	-	-	-	1	-	-	-	-
Subcutaneous Tissue	Sarcoma	-	-	1	-	-	-	-	-
	Fibroma	2	1	2	-	-	-	2	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Rat | **Gilead Study No.** R990204

Dose (mg/kg/day)		**0 (Vehicle)**		**10/300[a]**		**30**		**100**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Number of Animals with Neoplastic Lesions									
Subcutaneous Tissue	Fibrosarcoma	1	1	1	-	-	1	-	-
	Lipoma	-	-	3	2	-	2	-	1
Tail	Fibrosarcoma	1	-	-	-	-	-	1	-
	Osteosarcoma	-	-	-	-	-	1	-	-
Testis	Adenoma: intestitial cell	2	-	2	-	1	-	-	-
	Carcinoma: interstitial cell	-	-	-	-	1	-	-	-
	Adenoma: rete testis	-	-	-	-	1	-	-	-
Thorax	Liposarcoma	-	-	-	-	1	-	-	-
Thyroid	Adenoma: follicular cell	2	-	3	-	-	1	1	1
	Carcinoma: follicular cell	-	-	-	-	2	1	-	-
	Adenoma: C-cell	4	4	4	6	2	3	3	2
	Carcinoma: C-cell	2	1	1	-	-	-	1	-
Tongue	Papilloma: squamous cell	-	-	-	-	1	-	1	-
	Sarcoma	-	-	-	-	-	-	-	1
Urinary Bladder	Papilloma: transitional cell	-	-	-	-	-	1	1	-
Uterus	Polyp: endometrial stromal	-	2	-	11	-	4	-	3
	Benign granular cell tumor	-	3	-	-	-	-	-	-
Vagina	Polyp	-	-	-	1	-	-	-	1
	Benign granular cell tumor	-	-	-	-	-	1	-	-
	Papilloma: squamous cell	-	-	-	1	-	-	-	-

Test Article: TDF

Report Title: An Oral Carcinogenicity Study of Tenofovir Disoproxil Fumarate (TDF) in the Albino Rat	**Gilead Study No.** R990204

Dose (mg/kg/day)		**0 (Vehicle)**		**10/300[a]**		**30**		**100**	
Number evaluated		**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**	**M: 60**	**F: 60**
Gross Pathology		-	-	-	-	-	-	-	-
Histopathology- Non-Neoplastic Findings									
Kidney	Karyomegaly, tubular	-	-	29	17	1	-	5	-
	Chronic progressive nephropathy	46	23	29	11	47	16	38	13
Tumor Analysis									
Uterus – Polyp:endometrial Stromal Data			P= 0.0053**		P= 0.0065**				

a Dose increased on Day 10 from 10 to 300 mg/kg/day.
b Terminated at Week 103.
* = Pair wise test; ** = P < 0.05, Dunnett's

10.7. TAF

No carcinogenicity studies have been conducted with TAF, but carcinogenicity studies with TDF are included in Sections 2.6.7.10.7 and 2.6.7.10.8.

11. REPRODUCTIVE AND DEVELOPMENTAL TOXICITY (NONPIVOTAL STUDIES)

11.1. FTC

Test Article: FTC

Species/ Strain	Method of Administration (Vehicle/ Formulation)	Dosing Period	Dose (mg/kg/day)	No. per Group	Noteworthy Findings	Study Number
Mouse / CD-1	Oral Gavage	GD 6-15	0, 250, 500, 1000, divided doses spaced by approximately six hours	5 F	One fetus in the control group had exencephaly without open eyelids (bilateral). No other external malformations and no developmental variations were observed in fetuses in this study. No other significant effects were reported. NOAEL = 1000 mg/kg/day for maternal and developmental toxicity of FTC administered twice daily.	TOX033[a]
Rabbit / New Zealand White	Oral Gavage	GD 7-19	0, 250, 500, 1000, divided doses spaced by approximately six hours	5 F	All animals survived to the scheduled necropsy on gestation day 29. No treatment-related clinical signs were observed at any dose level. Mean maternal body weights, body weight changes, net body weights, net body weight changes, gravid uterine weights, and food consumption were unaffected by treatment at all dose levels.Intrauterine growth and survival were also unaffected by treatment. Parameters evaluated included postimplantation loss, mean fetal body weights, viable litter size, and mean numbers of corpora lutea and implantation sites. No external malformations or developmental variations were observed in fetuses. No maternal toxicity or developmental toxicity was noted at dosage levels up to 1000 mg/kg/day.	TOX034[a]

a Indicates GLP compliant.
GD = gestation day; F = female

11.2. RPV

Test Article: RPV

Species/ Strain	Method of Administration (Vehicle/ Formulation)	Dosing Period	Dose (mg/kg/day)	No. per Group	Noteworthy Findings	Study Number
Rat/Sprague	Oral/gavage (PEG400 + CA)	GD 6-	0 (vehicle), 40, 120,	8 F	No effects	TMC278-

Test Article: RPV

Dawley		17	400 mg/kg/day TMC278 base			NC127[a]
Rabbit / New Zealand White	Oral Gavage (0.5% HPMC)	GD 6-19	0 (vehicle) , 25, 75, 150 mg/kg/day TMC278 base	6 F	25 mg/kg/day: 1 F sacrificed during the study because of abortion of its litters. 75 mg/kg/day: 100% postimplantation loss. 150 mg/kg/day: 1F sacrificed during the study because of abortion of it's litter, ↓ fecal output in all animals, ↓ body weight and food consumption, 100% post-implantation loss	TMC278-NC128
Rabbit / New Zealand White	Oral Gavage (0.5% HPMC)	GD 6-19	0 (vehicle) , 5, 20, 60 mg/kg/day TMC278 base	6F	5 mg/kg/day: No effects. 20 mg/kg/day: No effects. 60 mg/kg/day: ↓ Body weight, 2 F aborting prior to term, 2 F exhibiting total litter resorption (early resorptions) at term and 2 F with live litters exhibiting increased (33 and 43%) postimplantation loss.	TMC278-NC129

a Indicates GLP compliant.
GD = gestation day; F = female

11.3. TAF

Test Article: TAF

Species / Strain	Method of Administration (Vehicle / Formulation)	Dosing Period	Doses (mg/kg)	No. Per Group	Noteworthy Findings	Gilead Study No. (CRO Name and Study No.)
Rats/ Sprague-Dawley Crl:CD(SD)	Oral Gavage (0.1% (v/v) Tween 20, 0.1% (w/v) (HPMC) K100LV prepared in ultra-pure water)	12 days (GD 6-17)	5, 100, 200	8F/group	Decreased body weight gain (-18%) and food consumption at 200 mg/kg/day. NOAEL for maternal toxicity: 100 mg/kg/day NOAEL for embryo-fetal development:200 mg/kg/day (AUC0-t = 35.7µg•hr/mL)	TX-120-2001[a] ([redacted] 902244)
Rabbits/New Zealand White	Oral Gavage (0.1% (v/v) Tween 20, 0.1% (w/v) (HPMC) K100LV prepared in ultra pure water)	13 days (GD 7-20)	0, 5, 25, 50, 100	6F/group	Maternal effects: decreased body weights and food intake at 100 mg/kg/day. No evidence of embryolethality, fetotoxicity or teratogenicity at any dose level. NOEL for maternal toxicity : 50 mg/kg/day (AUC0-t = 11.8 and 5.7 µg•hr/mL for R-PMPA and GS-7340, respectively) NOEL for embryo-fetal development: 100 mg/kg/day (AUC0-t = 23.5 and 10.7 µg•hr/mL for R-PMPA and GS 7340, respectively).	TX-120-2004[a] ([redacted] 902247)

a Indicates GLP compliant
GD= gestation day; NOEL = No-Observed- Effect Level; NOAEL = No-Observed-Adverse-Effect Level; F = female; R-PMPA = tenofovir

12. REPRODUCTIVE AND DEVELOPMENTAL TOXICITY: FERTILITY AND EARLY EMBRYONIC DEVELOPMENT TO IMPLANTATION

12.1. FTC: Fertility and Early Embryonic Development in Mice

Test Article: FTC

Report Title: Study of Fertility and Early Embryonic Development of TP-0006 Administered by Gavage to CD-1 Mice		**Gilead Study No.** TOX036
Duration of Dosing: M: received 4 Weeks pre-breeding exposure, 21 days during mating and 21 days post-mating, for a total of 70 days; **F:** 14 days prebreed, 21 days mating, 7 days post coitum (gestational day 0–6) for total maximum of 42 days.		**GLP Compliance:** Yes
Species/Strain: Mouse / CD-1	**Day of Mating:** 21 Days	**Design similar to ICH 4.1.1:** Yes
Initial Age: 51 days old upon arrival	**Day of C-Section:** GD 13	**Vehicle/Formulation:** 0.5% methylcellulose
Date of First Dose: M: [redacted] or [redacted], 19[redacted] **F:** [redacted], 19[redacted]		**Lot No.:** [redacted]; [redacted]
Special Features: none	**Method of Administration:** Oral gavage, divided doses spaced by approximately six hours	
No Observed Adverse-Effect Level: F_0 M/F =1000 mg/kg/day; F_1 Litters =1000 mg/kg/day		

Dose (mg/kg/day)	**0 (Vehicle)**	**250**	**500**	**1000**
Toxicokinetics: AUC (males)	NA	NM	NM	NM
No. Evaluated	21	20	21	21
Mortality				
No. Died	0/20	0/20	0/20	0/20
Sacrificed Moribund[a]	0	1	1	1
Clinical Observations	N	N	N	N
Necropsy Observations				
Paired testes weight (g)	0.247	0.248	0.252	0.231
Paired epididymides weight (g)	0.116	0.119	0.120	0.111
Body Weight (%)	N	N	N	N
Food Consumption (%)	N	N	N	N
Reproductive Toxicity				
Mating index (%)	100	100	100	100

Test Article: FTC

Report Title: Study of Fertility and Early Embryonic Development of TP-0006 Administered by Gavage to CD-1 Mice				**Gilead Study No.** TOX036
Dose (mg/kg/day)	**0 (Vehicle)**	**250**	**500**	**1000**
Fertility index (%)	96	90	100	90
Pregnancy index (%)	100	94.7	100	94.7
Epididymal sperm count (10^6/g)	620.1	692.2	667.3	757.2
Sperm motility (% motile)	69.3	68.9	69.7	71.1
Toxicokinetics: AUC (females)	NA	NM	NM	NM
No. Evaluated	20	20	20	20
Mortality				
No. Died[b]	0/20	0/20	0/20	0/20
Sacrificed moribund	0	0	0	0
Clinical Observations	N	N	N	N
Necropsy Observations				
Gravid uterine weight (g)	6.65	6.92	6.30	6.59
Paired ovary weight (g)	0.049	0.055	0.064	0.058
Body Weight (%)	N	N	N	N
Food Consumption (%)	N	N	N	N
Reproductive Toxicity				
Mating index (%)	100	100	100	100
Fertility index (%)	96.0	90.0	100	90.0

Test Article: FTC

Report Title: Study of Fertility and Early Embryonic Development of TP-0006 Administered by Gavage to CD-1 Mice				**Gilead Study No.** TOX036
Dose (mg/kg/day)	**0 (Vehicle)**	**250**	**500**	**1000**
Gestational index (%)	100	100	100	100
Preimplantation loss/litter (%)	5.74	4.08	11.46	9.04
Postimplantation loss/litter (%)	4.10	4.98	13.42	2.56
Parents				
Females that mated (Mating Index, %)	20 (100.0)	20 (100.0)	20 (100.0)	20 (100.0)
Fertility Index (No. females pregnant/No. females mated, %)	96.0	90.0	100.0	90.0
Evaluated pregnant females	20	18	20	18
Litter Means (mean per litter ± standard deviation)				
Corpora lutea	13.25 ± 0.42	13.22 ± 0.46	14.10 ± 0.69	14.06 ± 0.89
Implantations (sites per litter)	12.50 ± 0.34	12.89 ± 0.42	12.35 ± 0.50	12.61 ± 0.52
Live fetuses (live implants per litter)	12.00 ± 0.36	12.22 ± 0.39	10.90 ± 0.73	12.28 ± 0.52
Dead fetuses (nonlive implants per litter)	0.50 ± 0.14	0.67 ± 0.18	1.45 ± 0.45	0.33 ± 0.11
Resorptions	0.45 ± 0.14	0.61 ± 0.16	1.35 ± 0.45	0.33 ± 0.11
Weight of fetuses (g)	NA	NA	NA	NA
Sex ratio of fetuses	NA	NA	NA	NA

Conclusion: Exposure of F_0 male and female mice by oral gavage during prebreeding and mating for both sexes and through implantation (on gestation day 6) for females, resulted in no toxicity for body weight or feed consumption. There were no effects of treatment on F_0 male or female reproduction or on development of the F_1 offspring through gestation day 13.

a Three males were euthanized moribund due to injury or infection.
b No females died, were euthanized moribund, or were removed from study.
N = Normal or no change from control group value; NM = Not measured; NA = not applicable

12.2. FTC: Male Fertility in Rats

Test Article: FTC

Report Title: A Fertility Study in Male Rats Given 524W91 by Gavage		**Gilead Study No.** TTEP/95/0028 (TOX679)
Duration of Dosing: M: 73 days (prior to mating and throughout mating, dose days 1–95); **F:** untreated		
Species/Strain: Rat / CD (Sprague-Dawley)	**Day of Mating:** Bred-dams day 0 (date of insemination)	**GLP Compliance:** Yes
Initial Age: M: F_0 males approx. 17 Weeks of age at time of pairing, **F:** F_0 approx. 12 Weeks of age at time of pairing	**Day of C-Section:** GD 13	**Design similar to ICH 4.1.1:** Yes
	Method of Administration: Oral gavage	**Lot No.:** [redacted]
Date of First Dose: M: [redacted], 19[redacted] **F:** [redacted], 19[redacted]	**Vehicle/Formulation:** 524W91 + 0.5% methylcellulose, 20 mL/kg/dose	**Special Features:** none
No Observed Adverse-Effect Level: F_0 Males: NOEL = 750 mg/kg/day, NOAEL = 3000 mg/kg/day; **F_0 Females:** 3000 mg/kg/day; **F_1 Litters:** 3000 mg/kg/day		

Results for F_0 Generation Males:

Dose (mg/kg/day)	0 (Vehicle)	150	750	3000
No. Evaluated	25	25	25	25
Mortality	0	0	0	1 (dosing accident)
Clinical Observations	None	None	Transient postdose salivation	Postdose salivation
Body weight (%)	-	No change	No change	No change
Food consumption (%)	-	Normal	Normal	Normal
Necropsy Observations				
Histopathology	1/10 nephritis 2/10 prostatitis	-	-	3/10 prostatitis
Gross lesions	1[a]	0	0	0
Organ weight (%)	-	No change	No change	No change
Sperm count and motility	-	Normal	Normal	Normal

Test Article: FTC

Report Title: A Fertility Study in Male Rats Given 524W91 by Gavage			**Gilead Study No.** TTEP/95/0028 (TOX679)	
Dose (mg/kg/day)	**0 (Vehicle)**	**150**	**750**	**3000**
Results for F_0 Generation Untreated Females:				
No. Evaluated	25	25	25	25
Body weight change (gestation)	-	No change	No change	No change
Food consumption (gestation)	-	No change	No change	No change
Maternal gross lesions[a]	None	None	None	None
Reproductive performance				
Fertility index, %	87.5	96.0	92.0	100.0
Pregnancy rate, %	88.0	96.0	92.0	100.0
Number of pregnant females	22	24	23	25
Total resorption	0	1	0	0
Implantation sites/dam	15.6	15.6	17.3	17.0
Viable fetuses/dam	14.4	14.8	15.7	16.2
Post-implantation loss/dam	1.2	0.8	1.6	0.8

Test Article: FTC

Report Title: A Fertility Study in Male Rats Given 524W91 by Gavage			**Gilead Study No.** TTEP/95/0028 (TOX679)	
Dose (mg/kg/day)	**0 (Vehicle)**	**150**	**750**	**3000**
Parents				
Females with sperm (Fertility index, %)	24 (87.5)	25 (96.0)	25 (92.0)	25 (100.0)
Pregnant females (Pregnancy rate, %)	21 (88.0)	24 (96.0)	23 (92.0)	25 (100.0)
Evaluated pregnant females	21	24	23	25
Litter Means (mean per litter ± standard deviation)				
Corpora lutea	18.2 ± 3.4	17.4 ± 3.9	18.3 ± 1.8	19.0 ± 2.4
Implantations	15.6 ± 3.5	15.6 ± 5.2	17.3 ± 1.7	17.0 ± 1.5
Live fetuses	14.4 ± 3.4	14.8 ± 5.3	15.7 ± 2.9	16.2 ± 1.6
Dead fetuses	NA	NA	NA	NA
Resorptions	1.2 ± 1.3	0.8 ± 1.2	1.6 ± 2.0	0.8 ± 0.9
Weight of fetuses (g)	NA	NA	NA	NA
Sex ratio of fetuses	NA	NA	NA	NA

a Raised white areas on the kidneys
NA = not applicable; NM = not measured; N = normal or no change from control group value

Conclusion: Male rats given 150, 750 or 3000 mg 524W91/kg/day orally for 73 days prior and throughout the period of mating to untreated female rats exhibited postdose salivation primarily at the high dose, but did not show any other evidence of treatment-related toxicity. No adverse effects on fertility, reproductive performance, sperm count, and sperm motility or pathological changes in the reproductive organs were observed.

12.3. RPV: Male Fertility in Rats

Test Article: RPV

Report Title: Oral Gavage Study of Fertility and Early Embryonic Development to Implantation with RPV in Rats

Study No.: TMC278-NC124

Species/strain: Rat/Sprague-Dawley

Age/weight at first dose: 6-7 weeks (M, first dose), 9-10 weeks (F, mating)

Duration of dosing: 14 weeks

Duration of postdose: 0 days

Administration route/method: Oral/Gavage

Doses: 0 (vehicle), 100, 400, 1600 mg base eq./kg/day TMC278.HCl

Test article batch: [redacted] and [redacted]

Vehicle/formulation: 0.5% w/v HPMC

GLP Compliance: Yes

Date start of study: [redacted]

No Observed Adverse Effect Level: 1600 mg base eq./kg/day

Toxicokinetics: Not determined

Noteworthy Findings:

Dose (mg base eq./kg/day)	**0**		**100**		**400**		**1600**	
No. of animals	**25M**	**25F**	**25M**	**25F**	**25M**	**24F**	**25M**	**25F**
Died or killed prematurely	-	-	-	-	-	-	-	-
Clinical observations	-	/	-	/	-	/	-	/
Body weight (g)	-	/	-	/	-	/	-	/
Food consumption (g)	-	/	-	/	-	/	-	/
Copulation index (%)	-	-	-	-	-	-	-	-
Fertility index (%)	-	-	-	-	-	-	-	-
Sperm motility (%)	-	/	-	/	-	/	-	/
Sperm morphology	-	/	-	/	-	/	-	/
Mean no. corpora lutea	/	-	/	-	/	-	/	-
Pre-implantation loss (% of corpora lutea)	/	-	/	-	/	-	/	-
Post-implantation loss (% of implantation sites)	/	-	/	-	/	-	/	-
Mean gravid uterus weight	/	-	/	-	/	-	/	-
Organ weights	-	/	-	/	-	/	-	/

F: female; HPMC: hydroxypropylmethylcellulose; M: male; - no noteworthy findings. /: not done.

12.4. RPV: Female Fertility in the Rat

Test Article: RPV

Report Title: Oral Gavage Study of Fertility and Early Embryonic Development to Implantation with RPV

Study No.: TMC278-NC125

Species/strain: Rat/Sprague-Dawley

Age at first dose: 11 - 12 weeks

Duration of dosing: 14 days prior to pairing + 7 days of pregnancy

Duration of post-dose: None

Administration route/method: Oral/gavage

Doses:0, 40, 120, 400 mg base eq./kg/day TMC278.HCl

Test article batch: [redacted]

Vehicle/formulation: 0.5% w/v HPMC

GLP compliance: Yes

Date start of study: [redacted]

No Observed Adverse Effect Level: 400 mg base eq./kg/day

Dose (mg base eq./kg/day)	**0 (vehicle)**	**40**	**120**	**400**
No. of animals	**F: 24**	**F: 24**	**F: 24**	**F: 24**
Mortality/Sacrifice	0/24	0/24	0/24	0/24
Behavior and appearance	N	N	N	N
Body weight gain	N	N	N	N
Corrected maternal weight gain	N	N	N	N
Food consumption	N	N	N	N
Weight of gravid uterus	N	N	N	N
Gross pathology	N	N	N	N
Pre-coital interval	N	N	N	N
Copulation rate	N	N	N	N
Fertility rate	N	N	N	N
Implantations	N	N	N	N
Corpora lutea of pregnancy	N	N	N	N
Pre-implantation loss	N	N	N	N
Embryos	N	N	N	N
Resorptions	N	N	N	N
Post-implantation loss	N	N	N	N

F: female; HPMC: hydroxymethylcellulose; N: no relevant changes

12.5. TAF: Fertility and General Reproduction in Rats

Test Article: TAF

Report Title: **Oral Gavage Study of Fertility and Early Embryonic Development to Implantation with GS 7340-03 in Rats**		**Gilead Study No. TX-120-2012** [redacted] **Study No.: 8264528**
Species / Strain: Rat/Sprague-Dawley	**Duration of Dosing:** M: at least 28 days prior to mating and throughout the mating period through the day prior to termination (at least 10 weeks prior to sacrifice)	
Initial Age: Males: Approximately 10 weeks Females: Approximately 12 weeks	F: at least 14 days prior to mating, throughout mating through Day 7 of gestation (confirmed mated) or gestation Day 13 (confirmed non-mated)	**GLP Compliance:** yes
Date of First Dose: Males: [redacted] Females: [redacted]	**Day of Mating:** GD 0	**Lot Number:** [redacted] (hemifumarate salt)
No-Observed-Adverse-Effect Level	**Day of C-Section:** GD 13	**Design Similar to ICH 4.1.1?** yes
F_0 Males: 80 mg/kg/day	**Method of Administration:** Oral Gavage	
F_0 Females: 80 mg/kg/day	**Vehicle:** 0.1% hydroxypropylmethylcellulose K100LV (HPMC),0.1% Tween 20 in reverse osmosis (RO) water	**Dose Volume:** 10 mL/kg
F_1 Litters: 160 mg/kg/day	**Special Features:** early embryonic toxicity	

Daily Dose (mg/kg/day)		**0 (Control)**	**20**	**80**	**160**	
Males	No. Evaluated	22	22	22	22	
	No. Died or Sacrificed Moribund	0	0	0	0	

Daily Dose (mg/kg/day)		**0 (Control)**	**20**	**80**	**160**	
	Clinical Observations					
	Discolored haircoat – left periorbital brown, perineal brown, front legs brown, and/or periorbital red	0	1	1	0	
	Rough haircoat	0	2	2	2	
	Body Weight (%)[a]	608	-0.5	-4.8	-12.3$^{\#H}$	
	Food Consumption (%)[a]	225.1	-0.2	-2.0	-9.6^{+H}	
	Necropsy Observations	-	-	-	-	
	Mean No. Days Prior to Mating	3.2	3.0	3.2	3.6	
	No. of Males that Mated	22	21	20	21	
	No. of Fertile Males	21	21	19	20	

Test Article: TAF

Report Title:	**Oral Gavage Study of Fertility and Early Embryonic Development to Implantation with GS 7340-03 in Rats**			**Gilead Study No. TX-120-2012 [redacted] Study No.: 8264528**		
	Sperm Motility (Motlile %)	91.8 (± 8.43)	95.2 (± 3.08)	95.5 (± 4.01)	94.0 (± 6.10)	
Females						
	No. Evaluated	22	22	22	22	
	No. Died or Sacrificed Moribund	0	0	0	0	
	Clinical Observations					
	Discolored haircoat (perineal yellow, shoulder(s) – yellow or brown, midline abdomen yellow, or front legs orange)	1	1	2	4	
	Premating Body Weight (%)[a]	305	-2.0	-3.6	-5.9*H	
	Gestation Body Weight (%)[a]	380	-0.5	-1.6	-5.3	
	Premating Food Consumption (%)[a]	169.2	0.2	-2.4	-10.0+H	
Daily Dose (mg/kg/day)		**0 (Control)**	**20**	**80**	**160**	
	Gestation Food Consumption (%)[a]	95.1	-0.1	0.7	-2.6	
	Necropsy Observations	-	-	-	-	
	Mean No. Estrous Cycles/14 days	2.8	2.6	2.3	2.5	
	Mean No. Days Prior to Mating	3.2	3.0	3.2	3.6	
	No. of Pregnant Females	21	22	21	21	
	No. Aborted or with Total Resorption of Litter	0	1	0	0	
	Mean No. Corpora Lutea	16.3	16.0	15.8	16.0	
	Mean No. Implantations	16.2	15.0	15.1	15.7	
	Mean % Preimplantation Loss	0.5	7.1	4.3	1.8	
	Mean No. Live Conceptuses	15.3	14.5	13.9	14.5	
	Mean No. Early Resorptions	0.9	0.5	1.3	1.0	
	Mean % Postimplantation Loss	5.8	7.9	8.6	7.3	

a For controls, group means are shown. For treated groups, percent differences or fold change from controls are shown. Statistical significance is based on actual data not on the percent.

- No noteworthy findings; + Mild, ++ Moderate, +++ Marked; * = $p \leq 0.05$ (<site statistical method used here>)
+H = Dunnett Exact Homogeneous Test Significant: 0.01 level
#H = Dunnett Exact Homogeneous Test Significant: 0.001 level
*H = Dunnett Exact Homogeneous Test Significant: 0.05 level

13. REPRODUCTIVE AND DEVELOPMENTAL TOXICITY: EFFECTS ON EMBRYO-FETAL DEVELOPMENT

13.1. FTC: Embryo/Fetal Development in Mice

Test Article: FTC

Report Title: A Study of the Effects of TP-0006 (FTC) on Embryo/Fetal Development in Mice		**Gilead Study No.** TOX037
Species/Strain: Mouse/CD-1	**Duration of Dosing:** GD 6–15	
Initial Age: 72 day old upon receipt	**Day of Mating:** Day 0	**GLP Compliance:** Yes
Date of First Dose: [redacted] 19[redacted]	**Day of C-Section:** GD 18	**Lot No.:** [redacted]
Method of Administration: Oral, divided doses spaced by approximately six hours		**Design similar to ICH 4.1.3:** Yes
Special Features: none	**Vehicle/Formulation:** 0.5% methylcellulose	
No Observed Adverse-Effect Level: 1000 mg/kg/day for maternal and developmental toxicity		

Dose (mg/kg/day)	**0 (Vehicle)**	**250**	**500**	**1000**
F_0 Toxicokinetics[a]: Conc. mean ± SD (µg/mL)	NA	52 ± 15	88 ± 14	186 ± 14
Females				
Females with Sperm	ND	ND	ND	ND
Pregnant Females	23	25	23	22
Evaluated Pregnant Females	23	25	23	22
Litters				
No. Corpora Lutea	15.3 ± 3.59	15.3 ± 2.69	14.3 ± 2.37	14.9 ± 3.00
No. Implantations	12.3 ± 2.42	12.6 ± 2.58	11.9 ± 3.23	12.1 ± 2.47
Live Fetuses	11.4 ± 2.68	11.7 ± 2.84	11.0 ± 3.20	11.7 ± 2.36
Dead Fetuses	0	0	0.0 ± 0.21	0.0 ± 0.21
Resorptions (early)	0.8 ± 1.17	0.8 ± 1.09	0.7 ± 0.70	0.4 ± 0.58
Resorptions (late)	0.1 ± 0.29	0.2 ± 0.37	0.1 ± 0.34	0.0 ± 0.21
Weight of Fetuses (g)	1.34 ± 0.088	1.34 ± 0.113	1.34 ± 0.117	1.26 ± 0.102
Sex of Fetuses (% of live fetuses, m|f)	48.0 | 52.0	51.8 | 48.2	47.6 | 52.4	49.8 | 50.2
Litters with any Fetal Alteration (%)	1 (1)	4 (3)	1 (1)	3 (3)
Fetuses available for Morphological Evaluation (%)	263 (23)	292 (25)	254 (23)	257 (22)

a A toxicokinetic phase was conducted with 5 female mice/group administered 250, 500, and 1000 mg/kg/day as a single dose on gestation day 15. The mean ± standard deviation for plasma concentrations were determined. The increase in plasma levels was roughly in proportion to the increase in dose administered at 0.5 hr.

NA = not applicable; ND = not determined; Conc = concentration; SD = standard deviation

13.2. FTC: Embryo/Fetal Development in Rabbits

Test Article: FTC

Report Title: A Study of the Effects of TP-0006 on Embryo/Fetal Development in Rabbits		**Gilead Study No. TOX038**
Species/Strain: Rabbit/New Zealand White	**Duration of Dosing:** GD 7–19	
Initial Age: approximately 4.5 months old	**Day of Mating:** Day 0	**GLP Compliance:** Yes
Date of First Dose: [redacted], 19[redacted]	**Day of C-Section:** GD 29	**Lot No.:** [redacted]
Method of Administration: Oral, divided doses spaced by approximately six hours		**Design similar to ICH 4.1.3:** Yes
NOAEL: F_0 Fem. =100 mg/kg/day for maternal toxicity; F_1 Litters =1000 mg/kg/day for developmental toxicity		**Vehicle/Formulation:** 0.5% methylcellulose

Dose (mg/kg/day)	**0 (Vehicle)**	**100**	**300**	**1000**
F_0 Toxicokinetics[a]: AUC_{0-12} (µg·h/mL) GD 19	NA	43.6	157.6	628.9
AUC_{0-24} (µg·h/mL) GD 19	NA	87.3	315	1258
Females				
Females with Sperm	ND	ND	ND	ND
Pregnant Females	18	19	19	17
Evaluated Pregnant Females	17	18	18	16
Litters (means per litter ± standard deviation)				
No. Corpora Lutea	9.9 ± 1.73	10.4 ± 2.31	10.2 ± 2.29	12.1 ± 3.86
No. Implantations	8.1 ± 2.37	7.6 ± 2.52	7.2 ± 2.04	7.0 ± 2.90
Live Fetuses	7.6 ± 2.37	6.8 ± 3.01	6.2 ± 2.60	6.9 ± 2.70
Dead Fetuses	0	0	0	0
Resorptions (early)	0.4 ± 0.70	0.7 ± 1.28	0.8 ± 1.76	0.1 ± 0.25
Resorptions (late)	0.2 ± 0.39	0.1 ± 0.32	0.1 ± 0.32	0.1 ± 0.25
Weight of Fetuses (g)	44.4 ± 4.86	45.7 ± 6.44	47.0 ± 5.72	42.4 ± 5.77
Sex of Fetuses (% of live fetuses, m|f)	ND	ND	ND	ND
Litters with Any Fetal Alteration (%)	0	0	0	0
Fetuses Available for Morphological Evaluation (%)	129 (17)	123 (17)	112 (18)	110 (16)

a Additional evaluations: On gestation day 19, satellite groups of 5 rabbits per dose were used to provide plasma samples to evaluate drug absorption. Animals were bled pre-dose, 1, 3, 7, and 12 hours after the first dose (the second dose was withheld until after plasma collection).ND = not determined; NA = not applicable: NOAEL = no-observed-adverse-effect level; GD = gestation day

13.3. RPV: Developmental Toxicity Study in Rats

Test Article: RPV

Report Title: A Study of the Effects of TP-0006 on Embryo/Fetal Development in Rats

Study No.: TMC278-NC105

Species/strain: Rat/SPF Sprague Dawley
Age/weight: about 11-12 weeks/245 - 301 g
Duration of dosing: Gestation Days (GD) 6-17
Day of mating: GD0
Day of c-section: GD21

Administration route/method: Oral/Gavage
Doses: 0, 40, 120 and 400 mg/kg/day TMC278 base
Test article batch: [redacted]
Vehicle/formulation: PEG400 + CA

GLP Compliance: Yes
Date start of study: [redacted]

No Observed Adverse Effect Level: 40 mg/kg/day (maternal and embryo-fetal)

Toxicokinetics:

Doses in mg/kg/day	**0 (vehicle)**	**40**	**120**	**400**
No. of animals	**-**	**F:18**	**F:18**	**F:18**
C_{max} (µg/mL) GD6	-	4.9	6.0	14
C_{max} (µg/mL) GD16	-	5.6	7.2	13
$AUC_{0-\infty}$ (µg·h/mL) GD6	-	33	65	182
AUC_{0-24h} (µg·h/mL) GD16	-	37	63	152

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**	**40**	**120**	**400**
No. of animals	**24F**	**24F**	**24F**	**24F**
Mortality	0	0	0	0
Pregnancy rate	21/24	24/24	23/24	23/24
Behavior and appearance	N	N	N	N
Body weight gain	N	N	(+)D	(+)D (GD10-13)
Corrected maternal weight gain	N	N	(+)D	(+)D
Food consumption	N	N	+D (GD10-17)	+D (GD10-17)
Weight of gravid uterus	N	N	N	N
Thyroid weight	N	N	+I	+I
Gross pathology	N	N	N	N

CA: citric acid; N = no relevant changes; D = decreased; F: female; I = increase; (+) = minimal; + = slight; GD = gestational day; PEG400: plyethylene glycol 400

Test Article: RPV

Study TMC278-NC105: Oral Developmental Toxicity Study in the Rats (continued)

Noteworthy Findings:

Dose (mg/kg/day)	**0 (vehicle)**	**40**	**120**	**400**
No. of animals	**24F**	**24F**	**24F**	**24F**
Litter Data				

Implantations	N	N	N	N
Corpora lutea of pregnancy	N	N	N	N
Pre-implantation loss	N	N	N	N
Live fetuses	N	N	N	N
Dead fetuses	N	N	N	N
Mean litter size	N	N	N	N
Early resorptions	N	N	N	N
Late resorptions	N	N	N	N
Post-implantation loss	N	N	N	N
Body weight of live fetuses	N	N	N	N
Sex ratio	N	N	N	N
Fetal observations				
Dilated renal pelvis	N	N	+I	+I

N = no relevant changes; I = increase; + = slight; F: female.

13.4. RPV: Developmental Toxicity Study in the Rabbit

Test Article: RPV

Report Title: A Study of the Effects of TP-0006 on Embryo/Fetal Development in Rabbits

Study No.: TMC278-NC130

Species/strain: Rabbit/New Zealand White

Age at first hormone injection/weight at first dose: between 18-21 weeks/ 2869 to 3794 g

Duration of Dosing: GD6 - 19

Day of Mating: GD0

Day of C-Section: GD28

Administration route/method: Oral/gavage

Doses: 0, 5, 10 and 20 mg/kg/day, as TMC278 base

Test article batch: [redacted]

Vehicle/formulation: 0.5% w/v HPMC

GLP Compliance: Yes

Date start of study: [redacted]

Special features: None

No Observed Adverse Effect Level: 20 mg/kg/day (maternal) and 10 mg/kg/day (fetal).

Toxicokinetics:

Doses in mg/kg/day	**0 (vehicle)**	**5**	**10**	**20**
No. of animals	**-**	**F:3**	**F:3**	**F:3**
C_{max} (µg/ml) GD6	-	6.4	9.7	13
C_{max} (µg/ml) GD19	-	6.7	10	15
$AUC_{0-\infty}$ (µg·h/mL) GD6	-	95	162	219
AUC_{0-24h} (µg·h/mL) GD19	-	105	170	232

Test Article: RPV

Dose (mg/kg/day)	**0**	**5**	**10**	**20**
No. of animals	**F:20**	**F:20**	**F:20**	**F:20**
Mortality	-	0/20	0/20	0/20
Clinical observations	-	N	N	N
Pregnancy rate[a]	-	16/20	14/20	18/20
Body weight gain	-	N	N	N
Corrected mean maternal weight gain	-	N	N	N
Food consumption	-	N	N	N
Weight of gravid uterus	-	N	N	N
Adrenal weights	-	N	N	N
Gross pathology	-	N	N	N

a Number of pregnant females/ terminally sacrificed females; HPMC: hydroxypropylmethylcellulose; N = no relevant changes, GD = gestational day

Test Article: RPV

Study: TMC278-NC130: Oral Developmental Toxicity Study in the Rabbit (continued)				
Dose (mg/kg/day)	**0**	**5**	**10**	**20**
No. of animals	**F:20**	**F:20**	**F:20**	**F:20**
Litter data				
Implantations	-	N	N	N
Corpora lutea of pregnancy	-	N	N	N
Pre-implantation loss	-	N	N	N
Live fetuses	-	N	N	N
Dead fetuses	-	N	N	N
Mean litter size	-	N	N	N
Early resorptions	-	N	N	N
Late resorptions	-	N	N	N
Post-implantation loss	-	N	N	N
Weight of live fetuses	-	N	N	N
Sex ratio	-	N	N	N
Fetal observations				
left subclavian artery branches arising from the aorta	-	N	N	+I
hypoplastic interparietal bone	-	N	N	+I

I = increase; + = slight; N = no relevant changes.

13.5. TAF: Developmental Toxicity in Rat

Test Article: TAF

Report Title: An Embryo-fetal Development Study of GS-7340-02 by Oral Gavage in Rats		**Gilead Study No. TX-120-2002** [redacted] **Study No. 902245**
Species / Strain: rats / Sprague-Dawley Crl:CD(SD)	**Duration of Dosing:** 12 days (GD 6 to 17)	
Initial Age: 11 to 12 weeks at treatment start	**Day of Mating:** Time-mated	**GLP Compliance:** yes
Date of First Dose: [redacted] (GD 6)	**Day of C-Section:** GD 21	
Special Features: Toxicokinetic evaluation	**Method of Administration:** Oral Gavage	**Batch Number:** [redacted]
No Observed Adverse Effect Level: **F_0 Females:** 100 mg/kg/day (Targeted dose level)	**Vehicle:** 0.1% (v/v) Tween 20, 0.1% (w/v) hydroxypropylmethylcellulose (HPMC) K100LV prepared in ultra pure water	
F_1 Litters: 100 mg/kg/day (Targeted dose level)		

Dams	**Daily Dose (mg/kg/day)**	**0 (Control)**	**25 [b]**	**100[b]**	**250**
	Number of Animals (toxicokinetic)	3	9	9	9
	Toxicokinetics (GS-7340): AUC_{0-t} (µg*hr/mL)				
	Gestation Day 6	-	NC	NC	137.6
	Gestation Day 17	-	NC	0.242	138.2
	Toxicokinetics (GS-7340): C_{max} (µg/mL)				
	Gestation Day 6	-	NC	0.038	0.756
	Gestation Day 17	-	NC	0.149	0.597
	Toxicokinetics (R-PMPA): AUC_{0-t} (µg*hr/mL)				
	Gestation Day 6	-	2.715	0.009	0.054
	Gestation Day 17	-	2.803	0.017	0.056

Test Article: TAF

Report Title:	An Embryo-fetal Development Study of GS-7340-02 by Oral Gavage in Rats			**Gilead Study No. TX-120-2002** [redacted] **Study No. 902245**	
Dams	**Daily Dose (mg/kg/day)**	**0 (Control)**	**25 [b]**	**100[b]**	**250**
	Number of Animals (toxicokinetic)	3	9	9	9
	Toxicokinetics (R-PMPA): C_{max} (µg/mL)				
	Gestation Day 6	-	1.110	0.003	0.010
	Gestation Day 17	-	0.870	0.004	0.007
	Number of Animals (main study)	25	25	25	25
	Noteworthy Findings				
	No. Pregnant (excluding animal with ammonium sulfide)	25	22	24	24 (23)
	No. Died or Sacrificed Moribund	0	0	0	0
	No. Aborted or with Total Resorption of Litter	0	0	0	1
	Clinical Observations	-	-	-	Transient salivation (7/25)
	Body Weight Change GD 6 to 18 (%)[a]	103.4	0	-2	-41***
	Corrected Body Weight, GD 21 (%)[a]	339.4	-3	-1	-10***
	Corrected Body Weight Gain, GD 6 to 21 (%)[a]	51.8	-11	-8	-56***
	Overall Food Consumption GD 6 to 18 (%)[a]	371.4	-2	-1	-17***
	Necropsy Observations	-	-	-	-
	Mean No. Corpora Lutea	17.4	17.9	18.6	17.3
	Mean No. Implantations	15.0	16.2	16.0	14.5
	Mean % Preimplantation Loss	13.2	9.8	13.4	15.3
	Number of Animals (main study)	25	25	25	25
	Noteworthy Findings				

Test Article: TAF

Report Title:	An Embryo-fetal Development Study of GS-7340-02 by Oral Gavage in Rats			**Gilead Study No. TX-120-2002** [redacted] **Study No. 902245**	
	Daily Dose (mg/kg/day)	**0 (Control)**	**25 [b]**	**100[b]**	**250**
Litters	Litters Evaluated	25	22	24	23
	Live Fetuses	14.3	15.7	15.6	14.0
	Dead Fetuses	0	0	0	0
	Mean No. Resorptions	0.7	0.5	0.3	0.5
	Mean % Postimplantation Loss	4.70	2.99	2.01	7.12
	Mean Fetal Body Weight (total) (g)	5.83	5.62	5.53**	4.94***
	Fetal Sex Ratios (% Males)	49.9	49.3	48.8	44.0
	Overall Major Malformations	No treatment related findings.			
	Overall Minor External and Visceral Anomalies	No treatment related findings.			
	Overall Minor Skeletal Anomalies: litters (fetuses)	20 (48)	14 (35)	20 (49)	13 (19)***
	Interparietal Bone Incomplete Ossification: litters (fetuses)	11 (20)	7 (16)	9 (19)	2** (2)***
	Hyoid Bone Incomplete Ossification: litters (fetuses)	9 (16)	7 (15)	7 (20)	0** (0)***
	Common Skeletal Variants Sternebra 5 and Xiphisternum (unossified/incomplete/irregular/semi-bitartite/bi-partite)	14.74	17.33	18.90	31.43 **

GD = gestation day; - = no noteworthy findings; NC = not calculated

a For controls, group means are shown. For treated groups, percent differences from controls are shown. Statistical significance is based on actual data not on the percent.

b From GD 6 to 9, all Group 3 animals (100 mg/kg/day) received 65% of the target dose (65 mg/kg/day), and from GD 13 to 17, all Group 2 animals (25 mg/kg/day) received 77% of the target dose (19.3 mg/kg/day).

** = $p \leq 0.01$; *** = $p \leq 0.001$

13.6. TAF: Developmental Toxicity in Rabbits

Test Article: GS-7340-02

Report Title: **An Embryo-fetal Development Study of GS-7340-02 by Oral Gavage in Rabbits**		**Gilead Study No. TX-120-2005 [redacted] Study No. 902248**
Species / Strain: New Zealand White Rabbits (Oryctolagus cuniculus)	**Duration of Dosing:** Days 7 to 20 postcoitum (i.e., gestation)	
Initial Age: 5 months	**Day of Mating:** Time-mated	**GLP Compliance:** Yes
Date of First Dose: [redacted]	**Day of C-Section:** Day 29 postcoitum (i.e., gestation)	**Batch Number:** [redacted]
Special Features: Toxicokinetics	**Method of Administration:** Oral Gavage	
	Vehicle: 0.1% (v/v) Tween 20, 0.1% (w/v) hydroxypropylmethylcellulose (HPMC) K100LV prepared in ultra pure water	

Daily Dose (mg/kg/day)		**0 (Control)**	**10**	**30**	**100**
Number of Animals		**3**	**3**	**3**	**3**
Does:	Toxicokinetics:				
	GS-7430 - C_{max} (µg/mL) – GD 7	BLLQ	0.008	0.077	0.846
	GS-7430 - C_{max} (µg/mL) – GD 20	BLLQ	0.155	0.937	9.190
	R-PMPA - C_{max} (µg/mL) – GD 7	BLLQ	0.261	0.398	2.193
	R-PMPA - C_{max} (µg/mL) – GD 20	BLLQ	0.260	0.676	2.970
	GS-7430 - AUC_{0-t} (µg·h/mL) – GD 7	BLLQ	NC	NC	0.918
	GS-7430 - AUC_{0-t} (µg·h/mL) – GD 20	BLLQ	NC	1.135	11.04
	R-PMPA - AUC_{0-t} (µg·h/mL) – GD 7	BLLQ	1941	3.013	13.44
	R-PMPA - AUC_{0-t} (µg·h/mL) – GD 20	BLLQ	2018	5.005	27.25

Test Article: GS-7340-02

Report Title:	**An Embryo-fetal Development Study of GS-7340-02 by Oral Gavage in Rabbits**			**Gilead Study No. TX-120-2005 [redacted] Study No. 902248**	
Daily Dose (mg/kg/day)		**0 (Control)**	**10**	**30**	**100**
Does:	No. Pregnant	18	17	19	20
	No. Died or Sacrificed Moribund	0	0	0	0
	No. with Total Resorption of Litter	0	1	0	0
	Clinical Observations				
	Feces Output Decreased	2	3	2	8
	Fur staining yellow	1	2	6	7
	Body Weight Gain (total per interval) (%)[a]				
	GD 7 to 21	0.28 g	-17.9	-14.3	-60.7***
	GD 7 to 29	0.36 g	-8.3	13.9	-16.7
	GD 21 to 29	0.08 g	25.0	112.5*	137.5**
	Food Consumption (total per interval) (%)[a]				
	GD 7 to 21	2341 g	-4.1	-5.5	-17.6***
	GD 7 to 29	3332 g	-3.1	-1.2	-11.6
	GD 21 to 29	991 g	-0.6	7.8	1.9
	Necropsy Observations	No treatment-related findings			
	Mean No. Corpora Lutea[b]	9.3	8.6	9.0	9.4
	Mean No. Implantations[b]	8.8	8.2	8.4	8.9
	Mean % Pre-implantation Loss[b]	5.6	6.9	6.5	5.2

Test Article: GS-7340-02

Report Title:	**An Embryo-fetal Development Study of GS-7340-02 by Oral Gavage in Rabbits**			**Gilead Study No. TX-120-2005 [redacted] Study No. 902248**	
Daily Dose (mg/kg/day)		**0 (Control)**	**10**	**30**	**100**
Litters:	Litters Evaluated[b]	18	17	19	20
	Live Fetuses[b]	8.6	8.1	8.0	8.5
	Dead Fetuses[b]	0.0	0.0	0.0	0.0
	Mean No. Resorptions[b]	0.2	0.2	0.4	0.4
	Mean % Postimplantation Loss[b]	2.6	7.2	4.5	3.8
	Mean Fetal Body Weight (total) (g)	41.2	42.7	43.6	40.5
	Fetal Sex Ratios (% Males)	52.0	49.9	56.4	47.5
	Fetal Abnormalities (external, visceral, skeletal)	No treatment-related findings			

a For controls, group means are shown. For treated groups, percent differences from controls are shown. Statistical significance is based on actual data not on the percent.
b Including animal with total resorption
- No noteworthy findings; * = $p \leq 0.05$ (Dunnett) & = $p \leq 0.05$ (Wilcoxon); ** = $p \leq 0.01$ (Dunnett), *** = $p \leq 0.001$ (Dunnett), GD = gestation day; NC = Not calculable; BLLQ = Below lower limit of quantification;
R-PMPA = Tenofovir

14. REPRODUCTIVE AND DEVELOPMENTAL TOXICITY: EFFECTS ON PRE- AND POST-NATAL DEVELOPMENT, INCLUDING MATERNAL FUNCTION

14.1. FTC: Pre- and Post-natal Development in Mice

Test Article: FTC

Report Title: Pre-and Postnatal Development Study in CD-1 Mice Given TP-0006 by Gavage		**Gilead Study No. TOX039**
Species/Strain: Mouse/CD-1	**Duration of Dosing: F:** GD 6 to PND 20	
Initial Age: 58 days old upon arrival	**Day of Mating:** F_0 GD 0; F_1 GD 0	**GLP Compliance:** Yes
Date of First Dose: [redacted], 19[redacted]	**Vehicle/Formulation:** 0.5% methylcellulose	**Lot No.:** [redacted]; [redacted]
Method of Administration: Oral, divided doses spaced by approximately six hours		**Design similar to ICH 4.1.2:** Yes
Special Features: none		
No Observed Adverse-Effect Level: F_0 Females = 1000 mg/kg/day for maternal and reproductive toxicity; F_1 M/F = 1000 mg/kg/day		

Dose (mg/kg/day)	**0 (Vehicle)**	**250**	**500**	**1000**
F_0 Toxicokinetics: AUC	ND	ND	ND	ND
Females				
Females with Sperm	25/25	25/25	24/25[a]	25/25
Pregnant Females	24	24	24	25
Females with Delivery	24	24	22[b]	25
F_0 Maternal Toxicity				
Mortality (number): Gestation	0	0	0	0
Lactation[c]	0	2	0	2
Gestational Body Weights	No treatment-related findings.			
Gestational Weights Changes	No treatment-related findings.			
Gestational Feed Consumption	No treatment-related findings.			
Gestational Clinical Observations	No treatment-related findings.			
Lactational Body Weights	No treatment-related findings.			
Lactation Weight Changes	No treatment-related findings.			
Lactational Feed Consumption	No treatment-related findings.			
Lactational Clinical Observations	No treatment-related findings.			
F_0 Reproductive Indices	No treatment-related findings.			
Parental Gross Necropsy				
F_0 Female Terminal Body Weights and Findings	No treatment-related findings.			

Test Article: FTC

Report Title: Pre-and Postnatal Development Study in CD-1 Mice Given TP-0006 by Gavage					**Gilead Study No. TOX039**			
Dose (mg/kg/day)	**0 (Vehicle)**		**250**		**500**		**1000**	
Litters								
Implantation Sites per Litter	12.54 ± 0.41		12.83 ± 0.52		12.25 ± 0.37		12.12 ± 0.54	
Live Births	12.54 ± 0.48		12.63 ± 0.47		12.91± 0.41		12.76 ± 0.29	
Stillbirths	0.04 ± 0.04		0		0.05 ± 0.05		0	
Survivors Index pnd 4[d]	94.93 ± 4.16		94.66 ± 4.15		98.67 ± 0.62		97.92 ± 0.79	
Lactation Index pnd 21[e]	97.75 ± 1.05		98.21 ± 1.30		99.43 ± 0.57		98.91 ± 0.75	
Weight at Birth (g) – Male	1.60 ± 0.03		1.62 ± 0.03		1.56 ± 0.02		1.63 ± 0.02	
Weight at Birth (g) – Female	1.56 ± 0.03		1.55 ± 0.03		1.52 ± 0.02		1.55 ± 0.02	
Weight at Weaning (g) – Male	13.97 ± 0.32		14.21 ± 0.29		13.22 ± 0.21		14.01 ± 0.29	
Weight at Weaning (g) – Female	13.51 ± 0.29		13.50 ± 0.27		12.70 ± 0.22		13.33 ± 0.28	
Sex ratio of Live Newborns (% of live newborns m|f)	47.0	53.0	48.1	51.9	49.6	50.4	49.2	50.8
F_1 Offspring Toxicity								
Lactational Survival (number of litters died)	1		1		0		0	
Postwean Developmental Landmarks:								
Estrous Cyclicity	N		N		N		↑**	
Postwean Body Weights	N		N		↓**[f]		N	
Postwean Body Weight Changes	N		N		↓**[g]		↓*[h]	
Lactational Body Weights/Litter	No treatment-related findings.							
Lactational Sex Ratio/Litter	No treatment-related findings.							
Lactational Clinical Observations	No treatment-related findings.							
Postwean Developmental Landmarks (vaginal patency, pupillary reflex, motor activity, learning and memory, postwean clinical observations)	No treatment-related findings.							

Test Article: FTC

Report Title: Pre-and Postnatal Development Study in CD-1 Mice Given TP-0006 by Gavage			**Gilead Study No. TOX039**	
Dose (mg/kg/day)	**0 (Vehicle)**	**250**	**500**	**1000**
F_1 Reproductive Toxicity				
Reproductive Indices	No treatment-related findings.			
Gestational Body Weights	No treatment-related findings.			
Gestational Body Weight Changes	No treatment-related findings.			
Gestational clinical observations	No treatment-related findings.			
Lactational body weights (PND 0 and 1)	No treatment-related findings.			
Lactation body weight changes	No treatment-related findings.			
Lactational clinical observations	No treatment-related findings.			
Parental Gross Necropsy				
F_0 male terminal body weights	N	N	↓*	N
F_0 female terminal body weights and findings	No treatment-related findings.			
F_0 male findings	No treatment-related findings.			
F_2 Offspring Toxicity				
Lactational survival (PND 0–1)	No treatment-related findings.			
Lactational body weight/litter (PND 0)	No treatment-related findings.			
Lactational sex ratio/litter	No treatment-related findings.			

a Female 3 was removed due to a dosing error (misdirected dose) on gestational day 12.
b Female 6 had implant sites only and was included as a pregnant female that did not have a live litter. Female 14 had started to deliver but had no pups the next morning. She was noted as a pregnant female but was not included in the live litter since it was not observed if live pups were present when she was in the process of delivering.
c The causes of 4 deaths were not determined; the lack of a dose-response relationship indicates that they were not treatment-related.
d Number surviving 4 days/number live on PND 0.
e Number surviving 21 days/number live on pnd 4.
f On study days 63 and 70 and at sacrifice in males, but not at 1000 mg/kg/day.
g For study days 56–63 only in males, but not at 1000 mg/kg/day.
h For study days 14–21 only in males.
GD = gestation day; PND = postnatal day; ND = not determined; N = normal; no statistically significant or biologically relevant change; ↓ or ↑ = increase or decrease; * = $p < 0.05$ versus control; ** = $p < 0.01$ versus control

14.2. RPV: Pre- and Post-natal Developmental Toxicity and Juvenile Toxicity Dose Range Finding Study in the Rat

Test Article: RPV

Report Title: Pre-and Postnatal Development Toxicity and Juvenile Toxicity Dose Range Finding Study in CD-1 in Rats

Study No.: TMC278-NC168

Species/strain: Rat/Sprague Dawley

Initial Age/weight: 10 weeks

Duration of dosing: GD6-LD7 (dams); D12-D25 of age (pups)

Day of mating: GD1

Administration route/method: Oral/Gavage

Doses: 0, 40, 120, 400 mg base eq./kg/day TMC278.HCl

Test article batch: [redacted]

Vehicle/formulation: 0.5% w/v HPMC.

GLP Compliance: No

Date start of study: [redacted]

No Observed Effect Level: 400 mg base eq./kg/day (maternal); 120 mg base eq./kg/day (fetal)

Toxicokinetics:

Maternal Dose (mg base eq./kg/day)	**0 (vehicle)**		**0 (vehicle)**		**40**		**120**		**400**	
Pup Dose (mg base eq./kg/day) LD12-D25	**0 (vehicle)**		**400**		**40**		**120**		**400**	
No. of Pups	**-**	**-**	**M: 8**	**F:8**	**M: 8**	**F:8**	**M: 8**	**F:8**	**M: 8**	**F:8**
Died or killed prematurely	-	-	0	0	0	0	0	0	0	0
C_{max} LD7 (µg/mL) exposure by lactation	-	-	-	-	0.05	0.07	0.08	0.08	0.16	0.14
C_{max} D25 of age (µg/mL) oral gavage	-	-	6.4	4.9	2.6	5.8	3.7	3.6	9.1	7.3
AUC_{0-24h} LD7 (µg·h/mL) exp. by lactation	-	-	-	-	0.62	0.74	0.94	0.91	1.9	1.8
AUC_{0-24h} D25 of age (µg·h/mL) oral gavage	-	-	43	39	12	18	34	28	50	53

- not determined; F: female; M: male; - not determined; GD: gestation day; HPMC: hydroxypropylmethylcellulose; LD: lactation day.

Test Article: RPV

Noteworthy Findings					
Dose (mg base eq./kg/day)	**0 (vehicle)**	**0 (vehicle)**	**40**	**120**	**400**
F0 Females: No. of Animals	**6**	**6**	**6**	**6**	**6**
No. Pregnant	6	6	6	6	6
No. died or sacrificed moribund	0	0	0	0	0
Clinical observations	-	-	-	-	-
Gestation body weight	-	-	-	-	-
Lactation body weight	-	-	-	-	-
Gestation food consumption	-	-	-	-	-
Lactation food consumption	-	-	-	-	-
Mean duration of gestation (days)	-	-	-	-	-
Total no of litters	-	-	-	-	-
Mean no of pups born	-	-	-	-	-
Mean live birth index	-	-	-	-	-
Total litter death during LD	-	-	-	-	-
Maternal Dose (mg base eq./kg/day)	**0 (vehicle)**	**0 (vehicle)**	**40**	**120**	**400**
Pup Dose (mg base eq./kg/day) LD12-D25	**0 (vehicle)**	**400**	**40**	**120**	**400**
F1 litters	**8M/8F**	**8M/8F**	**8M/8F**	**8M/8F**	**8M/8F**
Number of pups born	76	88	65	76	74
Pup Mortalities LD12-D25	-	--		1	2
Pup clinical signs LD12-D25					
Poor condition	-	-	-	-	2
Decreased activity	-	-	-	-	2
Pup body weight	-	-	-	-	-
Pup necropsy observations	-	-	-	-	-

D: day; HPMC: hydroxypropylmethylcellulose; LD: lactation day; - no noteworthy findings; F: female; M: male

14.3. RPV: Pre- and Post-natal Developmental Toxicity Study in the Rat

Test Article: RPV

Report Title: Pre-and Postnatal Development Toxcity Study in the Rat **Study No.:** TMC278-NC131

Species/strain: Rat/Sprague-Dawley **Administration route/method:** Oral/gavage **GLP Compliance:** Yes
Initial Age/weight: 9-10 weeks **Doses**: 0, 40, 120, 400 mg base eq./kg/day TMC278.HCl **Date start of study:** [redacted]
Duration of dosing: GD6 – LD20 **Test article batch:** [redacted]
Day of mating: GD0 **Vehicle/formulation:** 0.5% w/v HPMC
No Observed Adverse Effect Level: 400 mg base eq./kg/day (maternal and fetal)
Noteworthy Findings

Dose (mg base eq./kg/day)	**0**	**40**	**120**	**400**
F0 Females: No. of Animals	**25**	**25**	**25**	**25**
No. died or sacrificed moribund	0	2	0	2
Clinical observations	-	-	-	-
Gestation body weight	-	-	-	-
Lactation body weight	-	-	-	-
Gestation food consumption	-	-	-	-
Lactation food consumption	-	-	-	-
Macroscopic observations	-	-	-	-
Mean duration of gestation (days)	-	-	-	-
Mean no. implantations	-	-	-	-
Dose (mg/kg/day)	**0**	**40**	**120**	**400**
F1 generation (pre-weaning)	**25**	**25**	**25**	**23**
Mean no of pups born	-	-	-	-
Mean live birth index (%)	100	100	100	99.2*
Mean viability index (%)	99.8	99.4	97.6*	93.5**
Mean lactation index (%)	-	-	-	-

* $p \leq 0.05$; ** $p \leq 0.01$ (Analysis of variance). GD: gestation day; HPMC: hydroxypropylmethylcellulose; LD: lactation day; - no noteworthy findings

Test Article: RPV

Noteworthy Findings

Dose (mg base eq./kg/day)	**0**	**40**	**120**	**400**
F1 generation (post-weaning)	**20**	**20**	**20**	**20**
No. died or sacrificed moribund	0	0	0	0
Pup clinical observations	-	-	-	-
Pup body weight	-	-	-	-
Pup body weight gain	-	-	-	-
Auditory acuity	-	-	-	-
Locomotor coordination	-	-	-	-
Sexual developmental observations	-	-	-	-
Fertility/mating performance	-	-	-	-
Macroscopic observations	-	-	-	-

- no noteworthy findings.

14.4. TDF: Developmental and Perinatal/Postnatal Reproduction in Rats

Test Article: TDF

Report Title: Oral (Gavage) Developmental and Perinatal/Postnatal Reproduction Toxicity Study of GS-4331-05 (不純物TE*) in Rats Including a Postnatal Behavioral/Functional Evaluation		**Gilead Study No.** R990202
Species/Strain: Rat/Crl:CD(SD)IGS BR VAF/Plus	**Duration of Dosing: F:** From day 7 of presumed gestation to LD 20 or day 24 of presumed gestation (in rats that did not deliver a litter)	
Initial age: 59 Days	**Day of mating:** Day 0	**GLP Compliance:** Yes
Date of First Dose: [redacted]	**Method of Administration:** Oral gavage	**Lot No.:** [redacted]
Litters Culled/Not Culled: At weaning (day 21 lactation) 25 pups per sex, per group, where possible, were randomly chosen for continued evaluation. All other pups were sacrificed.		**Design similar to ICH 4.1.2:** Yes
NOEL: F_o Females = 50mg/kg/day; F_1 General Toxicity = 50 mg/kg/day; F_1 Behavioral, reproductive & developmental = 150 mg/kg/day		**Special Features:** None

Dose (mg/kg/day)		**0 (Vehicle)**	**50**	**150**	**450**	**600**
F_0	Toxicokinetics (From study R990202-PK)					
Females	C_{max} (µg/mL) Day 20 post partum	-	1.46	2.26	4.93	2.89
	$AUC_{0-\tau}$ (µg·h/mL) Day 20 post partum	-	5.88	11.7	26.8	27.3
	No. Evaluated	25	25	25	25	25
	Mortality[a]					
	Number died	0	0	1	0	0
	Moribund sacrificed	1	0	0	1	1
	Presumed Gestation					
	Maximal Possible Incidence (days x rats)/No. of Rats/Group	561/25	556/25	559/25	562/25	569/25
	Red Perioral Substance	0/0	1/1	37/14*	123/24**	124/25**
	Excess Salivation	0/0	0/0	7/5	12/10**	28/13**
	Urine Stained Abdominal Fur	0/0	0/0	0/0	9/4*	13/8**
	Ungroomed Coat	0/0	0/0	0/0	6/1	17/5**

Test Article: TDF

Report Title: Oral (Gavage) Developmental and Perinatal/Postnatal Reproduction Toxicity Study of GS-4331-05 (不純物TE*) in Rats Including a Postnatal Behavioral/Functional Evaluation					**Gilead Study No.** R990202	
Dose (mg/kg/day)		**0 (Vehicle)**	**50**	**150**	**450**	**600**
F_0 Females, cont.	Lactation					
	Maximal Possible Incidence (days x rats)/No. of Rats/Group	504/24	525/25	494/24	382/24	260/22
	Dehydration	4/4	1/1	2/2	10/6	16/8**
	Emaciation	0/0	6/1	0/0	5/4*	12/7**
	Urine Stained Abdominal Fur	4/2	6/3	8/3	45/9**	28/11**
	Red of Dried Perioral Substance	0/0	0/0	3/3	9/8**	12/7**
	Ungroomed Coat	0/0	1/1	0/0	6/4*	11/7**
	Cold to Touch	0/0	0/0	0/0	0/0	9/4**
	Necropsy Observations					
	Gestation/Lactation Body Weight (%)[b]	327.8g	0	0	0	0
	Gestation/Food Consumption (%)[c]	23.8g/day	0	0	-12**	-18**
	Lactation/Food Consumption (%)[c]	54.2g/day	0	0	-7%*	-15**
	Delivery Observations					
	Females with Sperm	25	25	25	25	25
	Pregnant Females (%)	25 (100)	25 (100)	24 (96)	24 (96)	23 (92)
	Females with Delivery (%)	24 (100)[d]	25 (100)	24 (100)	24 (100)	22 (100)[e]
	Mean Duration of Gestation Days (SD)	23.0 (0.2)	23.0 (0.2)	23.2 (0.4)	23.3 (0.5)*[f]	23.3 (0.5)
F_1 Litters	Implantations per Delivered Litter (SD)	16.3 (2.1)	15.5 (2.6)	15.2 (1.6)	15.4 (1.7)	15.5 (1.6)
	Live Births per Litter (SD)	14.7(2.2)	14.4 (2.9)	14.2 (1.7)	13.8 (3.0)	12.8 (2.9)
	Stillbirths (SD)	0.2 (0.6)	0.2(0.4)	0 (0.2)	0.7 (2.3)	2.0 (2.8)*
	Survivors Day 4 pp (%) Viability Index	99.4 351/353	99.2 357/360	99.1 338/341	80.8 256/317	53.9 152/282**

Test Article: TDF

Report Title: Oral (Gavage) Developmental and Perinatal/Postnatal Reproduction Toxicity Study of GS-4331-05 (不純物TE*) in Rats Including a Postnatal Behavioral/Functional Evaluation											**Gilead Study No.** R990202
Dose (mg/kg/day)		**0 (Vehicle)**		**50**		**150**		**450**		**600**	
F_1 Litters Means											
F_1 Litters	Survivors at Weaning (%) Lactation Index	98.6 346/351		98.0 350/357		98.1 319/325		95.0 230/242		97.4 148/152	
	Weight at Birth (g)	6.7 (0.5)		6.5 (0.5)		6.4 (0.4)		5.5 (0.7)**		5.1 (0.6)**	
	Weight at Weaning g (SD)	37.4 (5.4)		35.9 (7.1)		35.4 (5.8)		27.1(3.6) **		25.7 (2.5) **	
		M	**F**	**M**	**F**	**M**	**F**	**M**	**F**	**M**	**F**
	Sex Ratio of Fetuses	49.9	50.1	46.5	53.5	52.4	47.6	48.0	52.0	47.0	53.0
	Clinical Observations (Total Frequency: Days x Pups)										
	Cold to Touch	3/1		1/1		0/0		64/6		80/11**	
	Pale	3/1		0/0		0/0		32/3		80/11**	
	Not Nesting	0/0		0/0		0/0		0/0		31/4**	
	Umbilical Hernia	0/0		0/0		0/0		28/4**		3/1	
	Necropsy Observations										
	Incidence Pup Appearance Normal (%)	295 (99.7)		298 (99.3)		269 (100)		169 (94.4) **		97 (99)	
	Incidence Pup Liver Appearance Pale (%)	0 (0)		0 (0)		0 (0)		10 (5.6)*		0 (0)	
	Incidence Pup Testes Small & Flaccid (%)	0 (0)		0 (0)		0 (0)		0 (0)		1 (1.0)**	
	Kidney Weights g (mean (SD)	0.54 (0.08)		0.52 (0.09)		0.51(0.09)		0.40 (0.04) **		0.36 (0.05)**	

a No deaths were related to test article. All deaths were related to intubation errors with the exception of the rat in control group, which had dystocia.
b Terminal weights for control groups (means, at day 21, postpartum) are shown. For treatment groups, % differences from controls are shown.
c Maternal absolute food consumptions for control groups (during gestation & lactation) are shown. For treatment groups, % differences from controls are shown.
d Excludes 1 rat which was scarified moribund day 22 gestation.
e Excludes 1 rat which was scarified moribund day 19 gestation.
f Not considered test article related.
* = $p \leq 0.05$; ** = $p \leq 0.01$

14.5. TAF

No pre- and post-natal development toxicity studies have been conducted with TAF, but a pre- and post-natal development toxicity study with TDF is included in Section 2.6.7.14.2.

15. STUDIES ON JUVENILE ANIMALS

No juvenile toxicity studies have been conducted with FTC or TAF. For RPV (TMC278), studies in juvenile animals are described in this document in Sections 7.2.7 and 14.2.

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16. LOCAL TOLERANCE

16.1. RPV

Test Article: RPV

Type of Study	In vitro System or Species/Strain	Route/Method of Administration	Gender and No. per Group	Dose (Batch) (Reference/ Controls)	Noteworthy Findings	Study Number
In Vitro Tests						
Bovine corneal opacity - permeability eye irritation assay	In vitro	NA	NA	20% w/w TMC278.HCl in 0.9% NaCl ([redacted]) (Reference: imidazole)	Increase in opacity and no increase in permeability. TMC278 considered a moderate eye irritant	TMC278-NC202
Phototoxicity - Balb/c mouse fibroblast assay	In vitro	NA	NA	0 (vehicle), 0.001 to 2.16 mg/L TMC278.HCl ([redacted]) (positive control: Chlorpromazine) In presence and absence of UVA	TMC278.HCl was not phototoxic	TMC278-NC188[a]
In Vivo Tests						
Primary skin irritation	Rabbit/New Zealand White	Gauze patch applied to intact skin for 4 hours	1M + 2F	0.5 g of TMC278.HCl ([redacted])	No skin reactions, staining of skin or corrosive effects were observed. Classified as "non-irritant" to rabbit skin.	TMC278-NC159[a]

DMSO = dimethyl sulfoxide; F = female; M = male; NA = not applicable; UVA = ultraviolet A
a GLP compliant

16.2. TAF

Test Article: TAF

Type of Study	Species / Strain	Method of Administration (Vehicle / Formulation)	Duration of Dosing	Dose (mg/kg)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name & Study No.)
BCOP	Bovine Cornea	In Vitro (prepared in 0.9% saline)	Opacity = 4 ± 5 hr.; Permeability = 90 ± 5 min.	20% (w/w) in 0.9% sodium chloride solution	3 Corneas	GS-7340-03 elicited an in vitro irritancy score of 21.0 ± 8.7 and was predicted to be a non-corrosive/non-severe eye irritant.	TX-120-2013[a] ([redacted] EUN0017)
Dermal Irritation	Rabbit/NZW	Dermal (moistened in 0.5 – 0.6 mL distilled water)	4 hours	0.5 g/ rabbit	3M	No signs of erythema or edema were noted. Test article was considered nonirritating/ noncorrosive.	TX-120-2011[a] ([redacted] 1000078)

a Indicates GLP compliant
BCOP = bovine corneal opacity and permeability; NA = not applicable; RO = reverse osmosis; M = male; F = female; w/w = weight/weight

17. OTHER TOXICITY STUDIES

17.1. FTC

Test Article: FTC

Type of Study	Species/ Strain	Method of Administration	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study Number
Immuno-toxicity	Rat/ Sprague-Dawley	Oral Gavage	28 Days	0, 60, 240, 1000	5 M, 5 F	The objective of the study was to assess the immunotoxicity of TP-0006 (FTC) in the rat following once daily oral gavage administration for 28 days. No adverse clinical signs were noted during the dosing period. Body weight was unaffected by treatment with TP-0006. There was no effect of FTC at dose levels up to 1000 mg/kg/day on the immune response to sheep red blood cells.	TOX146[a]
Impurities	Mouse/ CD-1 (ICR)	Oral Gavage	28 Days (13 day post dose recovery)	FTC/ 不純物FA*: 0, 50/1, 150/3, 450/9	20 (10M, 10F)	All mice survived to the scheduled necropsy. There were no observations related to treatment with the combination of TP-0006 (FTC) and TP-0296 (不純物FA*) at any dose level. Body weight, food consumption, hematology, serum biochemistry values and organ weights were unaffected by administration of the test article combination at any dose level. There were no gross or microscopic findings attributed to the test article combination. The no-observed-effect level (NOEL) for oral administration of TP-0006 in combination with TP-0296 was 450/9 mg/kg/day TP-0006/TP-0296, the highest dose combination tested.	TOX153[a]

Test Article: FTC

Type of Study	Species/ Strain	Method of Administration	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study Number
Other	Mouse/ CD-1 (ICR)	Oral Gavage	28 Days	0, 50, 150, 450	20 (10M, 10F)	The NOEL for oral gavage administration of TP-0006 test article (FTC produced by the [redacted] chemistry method of synthesis) to mice for 28 consecutive days was 450 mg/kg/day, the highest dose tested.	TX-162-2001[a]
Other/ Reverse Mutation Assay	*S. typhimurium & E. Coli*	In Vitro	NA	For TP-0006: 1.5 to 5000 µg/plate. For TP-0296: 0.015 to 50 µg/plate. Mutagenicity assay: For TP-0006: 50 to 5000 µg/plate. For TP-0296: 0.50 to 50 µg/plate	NA	**Cytotoxic Effects:** no positive mutagenic responses were observed with any of the tester strains in either the presence or absence of S9 activation. The maximum dose of TP-0006 (FTC) and TP-0296 tested were 5000 and 50 µg per plate respectively. Neither precipitate nor appreciable toxicity was observed. **Genotoxic Effects:** No positive mutagenic responses were observed with any of the tester strains in either presence or absence of S9 activation. Mutagenic responses were observed with the positive control articles.	TOX151[a]

Test Article: FTC

Type of Study	Species/ Strain	Method of Administration	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study Number
Other/ Chromo-some Aberration	CHO-K_1 cells	In Vitro	NA	0.5 to 5000 µg/mL with 1% TP-0296	NA	**Cytotoxic Effects:** Substantial toxicity (i.e., at least 50% cell growth inhibition, relative to the solvent control) was not observed at any dose level in both the non-activated and S9 activated 4 hour exposure groups, or in the non-activate 20 hour continuous exposure group. Based upon the results of the toxicity study, the dose levels selected for testing in the chromosome aberration assay were ranging from 313 to 5000 µg/mL for TP-0006 (FTC) and from 3.13 to 50 µg/mL for TP-0296. **Genotoxic Effects:** Based on the result of this study, it was concluded that TP-0006 with 1% TP-0296 was negative for the induction of structural and numerical chromosome aberrations in CHO cells.	TOX152[a]

a Indicates GLP Study.
TP-0006 = FTC produced by the [redacted] chemistry method of synthesis; TP-0296 = 不純物FA*

17.2. RPV

Test Article: RPV

Species/ Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch No.	Doses (mg base eq./kg/day)	Noteworthy Findings	Study Number
Local lymph node	Mouse/CBA/ Ca/Ola/Hsd	Topical application (3 consecutive days) 4/group	[redacted]	5, 10 and 35% TMC278.HCl in an acetone:olive oil 4:1 (positive control: streptozotocin)	TMC278 has no sensitizing properties.	TMC278 -NC199
Mechanistic Studies						
Guinea Pig Adrenal Cell Test (Evaluation of cortisol biosynthesis)	In vitro (DMSO 1%)	-	[redacted]	10^{-9} to 10^{-5}M (0.37 ng/mL to 3.7 µg/mL) TMC278 base. Positive control: Etomidate (10^{-9} to 10^{-5} M)	TMC278 inhibited cortisol synthesis in one experiment with an IC_{50} value of 6 µM but showed no significant inhibition in the second experiment. Etomidate rendered an IC_{50} value of 0.5 nM	TMC278 - Exp5653
Dog adrenal cortex (Evaluation of the cortisol in crude subcellular fraction)	In vitro	-	[redacted]	0.03 to 75 µM (0.011 to 27.75 µg/mL) TMC278 base Positive control: Etomidate (0.28 µM)	**75 µM**: 39% inhibition of the conversion of pregnenolone. Progesterone and 17-α OH-progesterone concentrations increased with increasing concentrations of TMC278 whereas 11-deoxycorticosterone, 11-deoxycortisol, and corticosterone concentrations decreased with increasing concentrations of TMC278. No changes in cortisol. Etomidate showed a 38% inhibition of the corticosteroid synthesis.	TMC278 -FK4790

Test Article: RPV

Species/ Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch No.	Doses (mg base eq./kg/day)	Noteworthy Findings	Study Number
Dog/Beagle (endocrinology study) 6M/group	Oral/gavage (PEG400 + CA)	Single dose	[redacted]	0 (vehicle), 20, or 80 TMC278 base	No mortality. No relevant clinical signs. No effects on body weight, cortisol, ACTH, aldosterone and CRF. **20 mg/kg**: C_{max}: 2.8 µg/mL; $AUC_{0-\infty}$: 87 µg·h/mL **80 mg/kg**: C_{max}: 2.8 µg/mL; $AUC_{0-\infty}$:105 µg·h/mL.	TMC278 - Exp5627

a Unless indicated differently; ACTH: adreno corticotropic hormone ; CA: citric acid; CRF: corticotrophin releasing factor; DMSO: dimethylsufoxide; F: female; M: male; PEG400: polyethylene glycol 400.

Test Article: RPV

Species/Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch No.	Doses (mg/kg/day)	Noteworthy Findings	Study Number
Impurities (RPV)						
Qualification of 不純物A* and 不純物B* according to ICH Q3A						
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	-	[redacted] (TMC278HCl) + [redacted] (不純物A*)	0 (vehicle), 3.91, 7.81, 15.63, 31.25, 62.5, 125 and 250 µg/plate TMC278.HCl spiked with 4% 不純物A*	Precipitation at 55.69 µg/mL and above. No evidence of mutagenic properties/	TMC278 -NC308[a]
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	-	[redacted] (TMC278HCl) + [redacted] (不純物B*)	0 (vehicle), 3.91, 7.81, 15.63, 31.25, 62.5, 125 and 250 µg/plate TMC278.HCl spiked with 4% 不純物B*	Precipitation at 100 µg/plate and above/ No evidence of mutagenic properties/	TMC278 -NC309[a]

a GLP compliant; DMSO: dimethyl sulfoxide; Exp: experiment.

Test Article: RPV

Species/Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch No.	Doses (mg/kg/day)	Noteworthy Findings	Study Number
不純物A* and 不純物B* (continued)						
Mouse lymphoma L5178Y/tk$^{+/-}$	In vitro (DMSO)	-	[redacted] (TMC278HCl) + [redacted] 不純物A*)	**Exp 1**: 0 (vehicle), 2.5, 5, 10, 15, 20, 25, 30, 40 µg/mL 3 hours + S9 3 hours – S9 **Exp 2:** 0 (vehicle), 2.5, 5, 7.5, 10, 12.5, 15, 20, 25, 30, 35, 40, 50, 75 µg/mL. 3 hours +S9 24 hours –S9 TMC278 HCl spiked with 4% 不純物A*	Precipitation at 15 µg/mL and above/ No evidence of mutagenic or clastogenic properties.	TMC278-NC311[a]

a GLP compliant; DMSO: dimethyl sulfoxide; Exp: experiment.

Test Article: RPV

Species/Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch No.	Doses (mg/kg/day)	Noteworthy Findings	Study Number
Qualification of 不純物A* and 不純物B* according to ICH Q3A (continued)						
Mouse lymphoma L5178Y/tk$^{+/-}$	In vitro (DMSO)	-	[redacted] (TMC278HCl) + [redacted] (不純物B*)	**Exp 1**:0 (vehicle), 2.5, 5, 10, 15, 20, 25, 30 µg/mL 3 hours + S9 3 hours – S9 **Exp 2:** 0 (vehicle), 2.5, 5, 7.5, 10, 12.5, 15, 20, 25, 30, 35, 40, 50, 75 µg/mL. 3 hours +/-S9 24 hours –S9 TMC278 HCl spiked with 4% 不純物B*	Precipitation at 15 µg/mL and above. No mutagenic or clastogenic properties.	TMC278-NC312[a]
Rat/Sprague Dawley 10M and 10F/group	Oral/Gavage (0.5% w/v HPMC)	1-month	[redacted] + [redacted] (不純物A*) Or [redacted] + [redacted] (不純物B*)	0 (vehicle), 10 TMC278 HCl, 10 TMC278 HCl spiked with 4% 不純物A*, 10 TMC278 HCl spiked with 4% 不純物B*	No effects in any group different from the negative control group.	TMC278-NC314[a]

a GLP compliant; DMSO: dimethyl sulfoxide; F: female; HPMC: hydroxypropyl methylcellulose; M: male.

Test Article: RPV

Species/Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch no.	Doses (mg/kg/day)	Noteworthy Findings	Study No.
不純物H*						
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	---	[redacted]	不純物H* **Exp 1and 2**: 0 (vehicle), 78.13, 156.25, 312.5, 625, 1250, 2500 and 5000	Increased mutation frequency in strains TA98 and TA100 in the presence of rat liver S9-mix.	TMC278-NC179[a]
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	---	[redacted]	不純物I* **Exp 1, 2, 3, 4, 5 and 6**: 0 (vehicle), 78.13, 156.25, 312.5, 625, 1250, 2500 and 5000	Increasede mutation frequency in strain TA1537 in the absence of S9-mix and in strain TA98 in the presence of S9-mix.	TMC278-NC165
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	---	[redacted]	不純物H* **Exp 1 (2)**: 0 (0), 78.13 (93.75), 156.25 (187.5), 312.5 (375), 625 (750), 1250 (1500), 2500 (3000) and 5000 (5000)	Increased mutation frequency in the absence and the presence of S9-mix in TA98 and TA1537 and in the presence of S9-mix in TA100.	TMC278-1646_0016451[a]

a GLP compliant; DMSO: dimethyl sulfoxide

Test Article: RPV

Species/Strain No. and Gender/Group	Route/Method of Administration (Vehicle/ Formulation)	Duration of Dosing	Batch No.	Doses (mg/kg/day)	Noteworthy Findings	Study No.
Human lymphocytes +/- S9	In vitro/ (DMSO)	4 and 24 hours exposure	Not reported	不純物H* 0 (vehicle), 65.22, 130.44, 260.88 and 521.75 µg/mL	Statistically significant increase in the frequency of cells with aberrations in the absence of metabolic activation (S9) after 4 hours exposure group at 521.75 µg/mL.	TMC278-NC184
不純物E*						
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	-	[redacted]	**Exp 1 (2)**: 0 (0), 78.13 (93.75), 156.25 (187.5), 312.5 (375), 625 (750), 1250 (1500), 2500 (3000) and 5000 (5000)	No evidence of mutagenic properties with any of the tested strains both in the absence and the presence of S9-mix.	TMC278-1646_0015298[a]
不純物F*						
S. typhimurium TA98, TA100, TA1535, TA1537, TA102 +/- S9	In vitro (DMSO)	-	[redacted]	**Exp 1 (2)**: 0 (0), 78.13 (93.75), 156.25 (187.5), 312.5 (375), 625 (750), 1250 (1500), 2500 (3000) and 5000 (5000) **Exp 3**: 0, 78.13, 156.25, 312.5, 625, 1250, 2500 and 5000	No evidence of mutagenic properties with any of the tested strains both in the absence and the presence of S9-mix.	TMC278-1646_0015299[a]

a GLP compliant; DMSO: dimethyl sulfoxide

17.3. TAF/TVF

Test Article: TAF

Type of Study	Species / Strain	Method of Administration (Vehicle / Formulation)	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name & Study No.)
Other/ Reverse Mutation Assay	*S. typhimurium* & *E. Coli*	In Vitro Tenofovir	NA	Up to 5000 µg/plate with and without S9 activation	NA	Increase in revertants in TA1535 only in presence of S9	95-TOX-1278-006 ([redacted] 17444-0-409)
Other/ Mouse lymphoma L5178Y	Mouse lymphoma L5178Y cells	In Vitro Tenofovir	NA	Up to 5000 µg/mL with and without S9 activation	NA	Negative with activation; equivocal with activation at 5000 µg/mL	95-TOX-1278-007 ([redacted] 17444-0-431)
LLNA	Mouse	Topical GS-7340-03 (acetone:olive oil, 4:1 v/v)	3 days	25 µL application of 10, 25, 50% (w/v)	5F	TAF not regarded as a potential skin sensitizer.	TX-120-2014[a] [redacted] UK EUN0018)
Impurity Qualification	Rat/Crl:CD (SD)	Oral Gavage (0.1% (v/v) Tween 20 and 0.1% (w/v) HPMC in RO water	2 weeks	0, 5, 50 Lot-1: [redacted] (97.7% purity) Lot-2: [redacted] (83.1% purity)	10M	No GS-7340-02-related effects on clinical signs, body weights, food consumption, clinical or anatomic pathology for either lot. At 50 mg/kg/day the Day 14 tenofovir C_{max} was 0.2 µg/mL and the AUC_{0-t} was 2.0 µg·hr/mL for Lot No.1, and for Lot No. 2 the respective C_{max} was 0.4 µg/mL and the AUC_{0-t} was 2.5 µg·hr/mL.	TX-120-2008[a] ([redacted] 8246754)

Test Article: TAF

Type of Study	Species / Strain	Method of Administration (Vehicle / Formulation)	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name & Study No.)
Impurity Qualification	Rat/Crl:CD (SD)	Oral Gavage (1% (w/v) HPMC cellulose K100LV and 0.1% (v/v) Tween® 20 in 99.8% (v/v) reverse osmosis (RO) water)	4 weeks	0, 25, 50 (free base equivalents) Lot-1: [redacted] (99.3% purity) Lot-2: [redacted] (98.0% purity) Lot-3: [redacted] (97.8% purity)	10M; 10F	No GS-7340-03-related clinical observations, ophthalmic observations, or effects on clinical pathology parameters, bone biomarker parameters, or macroscopic and microscopic observations were noted. NOAEL = 50 mg/kg/day (free base equivalents) At 50 mg/kg/day the Day 28 R-PMPA combined male/female C_{max} was 1.79 µg/mL and the combined male/female AUC_{0-t} was 6.76 µg·hr/mL for Lot No.1, for Lot No. 2 the respective C_{max} was 1.38 µg/mL and the AUC_{0-t} was 6.03 µg·hr/mL, and for Lot 3 the respective C_{max} was 1.95 µg/mL and the AUC_{0-t} was 6.04 µg·hr/mL.	TX-120-2021[a] ([redacted] 8298810)
Bone Metabolism	Rat/Harlan:CD (SD)	Oral Gavage GS-7340-02 (50mM citrate buffer)	6 days	0, 400	4M	Serum 1,25-dihydroxycholcalciferol was decreased by 80% and urinary calcium/creatinine and phosphorus/ creatinine ratios were increased 444% and 202%, respectively, compared to controls.	R990177 (NA)

Test Article: TAF

Type of Study	Species / Strain	Method of Administration (Vehicle / Formulation)	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name & Study No.)
Bone Metabolism	Rat/Crl:CD (SD)	Oral Gavage GS-7340-02 (50 mM citric acid)	6 days	0, 100, 400	8M	Statistically significant decreases in serum 1,25-dihydroxyvitamin, calcium and phosphate were observed at 100 and 400 mg/kg/day. Urinary calcium:creatinine ratios were significantly decreased on Days 1 and 2, at 100 and on Day 1 at 400mg/kg/day, but were then significantly increased by Day 6, at 400 mg/kg/day.	R2000044 (NA)
Renal Effect	Rat/Crl:CD (SD)	Oral Gavage GS-7340-02 (50 mM citric acid)	1 day	0, 100, 1000[b]	10M	No adverse effect on renal function. Urinary output of calcium was increased at 1000 mg/kg. This correlated with an increase in serum calcium concentration and indicated that the kidneys were functioning in order to reduce the serum calcium load.	R990186[a] ([redacted] 56520)

Test Article: TAF

Type of Study	Species / Strain	Method of Administration (Vehicle / Formulation)	Duration of Dosing	Dose (mg/kg/day)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name & Study No.)
Bone Marker Assessment	Dog, Beagle	Oral Gavage GS-7340-02 (50 mM citric acid)	5 days	0, 37.5, 75	3M	Emesis, excessive salivation and diarrhea at 37.5 and 75 mg/kg/day. Decreased body weight and food consumption at both dose levels. Marked decreases in neutrophils, lymphocytes, monocytes and eosinophils were observed at both levels. Increases were noted in total protein, globulin, triglyceride, cholesterol, ALP, AST and CK at both dose levels. Test-article related findings were noted in the GI, lymph nodes, thymus, tonsil and bone marrow at both dose levels	D990142[a] ([redacted] 9529.3)

a Indicates GLP compliant
b No effect level
M = male; F = female; NA = not applicable; w/w = weight/weight; w/v = weight/volume; v/v = volume/volume
ALP = alkaline phosphatase, AST = serum aspartate aminotransferase, CK = creatine kinase

17.4. FTC/TDF

Test Article: FTC/TDF

Type of Study	Species/ Strain	Method of Administration	Duration of Dosing	Dose (mg/kg)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name and Study No.)
Impurities	Rat/Sprague-Dawley	Oral Gavage	14 Days	TDF/FTC: 30/20, 100/67, 300/200 (each dose level with non-degraded and degraded)	10 M	There were no toxicologically significant differences between groups treated with non-degraded and degraded TDF/FTC. A minor increase in adrenal weights at necropsy (all dose levels, except those receiving degraded TDF/FTC at the lowest dose) and a hyperplasia of the anterior duodenal mucosa overlying Brunner's glands was seen in animals treated at the high-dose level with non-degraded and degraded TDF/FTC. The NOAEL in this study was considered to be 100/67 mg/kg/day.	TX-164-2001[c] ([redacted] 800392)

Test Article: FTC/TDF

Type of Study	Species/ Strain	Method of Administration	Duration of Dosing	Dose (mg/kg)	Gender and No. per Group	Noteworthy Findings	Gilead Study No. (CRO Name and Study No.)
Impurities	Rat/Sprague-Dawley	Oral Gavage	14 Days	TDF/FTC: 0, 30/20[a], 100/67[a], 300/200[a], 30/20[b], 100/67[b], 300/200[b]	10 M	Minimal, non-adverse changes in red cell parameters noted in animals given non-degraded or degraded TDF/FTC at 300/200 mg/kg/day. Non-adverse, minimal, dose level-dependent increases in ALT activity were observed in rats given non-degraded TDF/FTC at ≥ 100/67 mg/kg/day and degraded TDF/FTC at 300/200 mg/kg/day. Minimal cryptal epithelium hyperplasia and minimal single cell necrosis in the duodenum with both degraded and non-degraded TDF/FTC at 300/200 mg/kg/day. NOAEL = 300/200 mg/kg/day for both non-degraded and degraded TDF/FTC, with no difference in toxicity between the non-degraded and degraded material. TDF Day 14 C_{max} 1.4 and 1.2 µg/mL, AUC_{0-24} 13.6 and 13.7 µg·h/mL; FTC Day 14 C_{max} 15.8 and 13.4 µg/mL, AUC_{0-24} 163 and 133 µg·h/mL for non-degraded and degraded tablets, respectively.	TX-164-2005[c] ([redacted] 805427)

a Non-degraded TDF/FTC.
b Degraded TDF/FTC.
c Indicates GLP compliant.