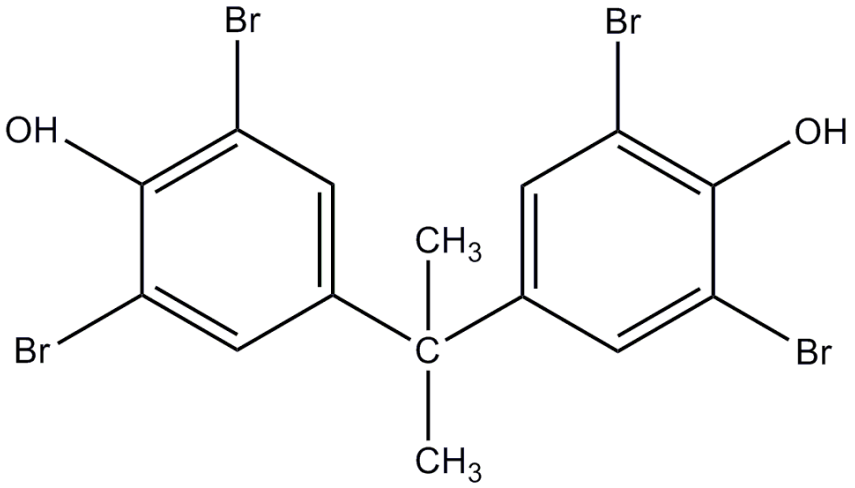
Draft NTP Technical Report TR 587
Tetrabromobisphenol A (TBBPA)
CAS # 79-94-7



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TBBPA – flame retardant

- Nominated by NIEHS - Widely used flame retardant – >100,000 tons worldwide/year
- TBBPA - reactive or additive flame retardant in epoxy and polycarbonate resins, and acrylonitrile, butadiene-styrene (ABS) resins and phenolic resins.
- Flame retardant in plastics, paper, electronics, textiles, adhesives
- Household products: computers, TVs, mobile phones

TBBPA ADME Information

- Rapidly absorbed following oral administration to rats or humans
- Rapidly metabolized with terminal serum half-life in rats < 5 hours; the half-life is longer in humans
- Primarily conjugated with glucuronic acid and sulfate and excreted in feces through the bile
- TBBPA has some potential to form reactive intermediates through redox cycling

NTP TBBPA Genetic Toxicity Studies

- Negative in *Salmonella typhimurium* strains TA98, TA100, TA1535, or TA1537 with and without exogenous metabolic activation
- No increase in micronucleated erythrocytes in male or female mice after 3 month exposure

TBBPA: 3-month rat and mouse study design

- **F344/NTac rat** - administered in corn oil by oral gavage to male and female rats at 0,10, 50, 100, 500, 1000 mg/kg for up to 3-month (5X week) - 5 ml/kg
- **B6C3F1/N mouse** - administered in corn oil by oral gavage to male and female rats at 0,10, 50, 100, 500, 1000 mg/kg for up to 3-month (5X week) - 10 ml/kg
- 1000 mg/kg – maximum dose due to solubility (NTP Chemistry)
- NTP 2000 diet

TBBPA: Summary of 3-month study results F344/NTac rat & B6C3F1/N1 mice

- No treatment-related mortality, clinical signs, or body weight effect (± 10% of controls) in rats or mice
- No treatment-related lesions except for minimal kidney cytoplasmic alteration in male mice at 500 &1000 mg/kg
- No treatment-related SMVCE findings

TBBPA: Summary of 3-month study results F344/NTac rat & B6C3F1/N1 mice

- ↑Liver weights (9-14%) in 500 and 1000 mg/kg rats
- ↑Liver weights (11-13%) in 500 and 1000 mg/kg male mice and 1000 mg/kg female mice
- Consistent, progressive, dose-related decrease in T4 (rats)
  - ↓T4 at 4, 23, 93 days
    - at 93 days ↓T4 17%-50% at 100, 500, & 1000 mg/kg
  - No change in TSH or T3

TBBPA: Dose selection for the 2-year study Wistar Han rat [Crl:WI(Han)] & B6C3F1/N mice

- None of the effects in the 3-month study were considered severe enough to compromise the conduct of a 2-year study
- Doses for 2-year study: 0, 250, 500, 1000 mg/kg

- Interim 3-month sacrifice to compare F344/N rat and Wistar Han rat findings (0 & 1000 mg/kg)

TBBPA: Summary of 3-month study in Wistar Han rats

- No treatment-related mortality or clinical signs
- Body weights within ± 10% of controls
- No treatment-related lesions
- ↑Liver weights (4-7%) at 1000 mg/kg in male & female Wistar Han rats
- ↓Thymus weight (19-20%) at 1000 mg/kg in male and female Wistar Han rats

TBBPA 2-year rat study
Final survival & mean body weight (at day 701)

Dose (mg/kg)	0	250	500	1000
Male rats Survival	33	28	38	39
Mean BW (gm) (% control)	663 -	615 (93)	599 (90)	584 (88)
Female rats Survival	35	34	29	33
Mean BW (gm) (% control)	375 -	403 (107)	368 (98)	375 (100)

N = 50

TBBPA female rat - Uterus

Original transverse read

Dose (mg/kg)	0	250	500	1000
Uterus – Adenoma	0**	0	3	4
Uterus – Adenocarcinoma (includes multiple)	3*	3	8	9
Uterus - MMMT	0	4	0	2
Uterus - Adenoma, adenocarcinoma, or MMMT[a]	3**	7	11*	13**

N = 50 *p ≤0.05, **p≤0.01

[a]Historical control incidence (all routes): 7/150 (4.7% ±2.3%), range 2%-6%

MMMT – Malignant mixed Müllerian tumor

TBBPA female rat – Uterus

Residual longitudinal review

Dose (mg/kg)	0	250	500	1000
Endometrium, hyperplasia, atypical	2	13**	11**	13**
Uterus - Adenoma	3	2	1	3
Uterus – Adenocarcinoma (includes multiple)	4**	9	15**	15**
Uterus - MMMT	0	0	0	1
Uterus - Adenoma, adenocarcinoma, or MMMT	6**	10	16**	16*

N = 50 *p≤0.05 **p≤0.01
MMMT – Malignant mixed Müllerian tumor

TBBPA female rat – Uterus

Original transverse & residual longitudinal review

Dose (mg/kg)	0	250	500	1000
Endometrium, hyperplasia, atypical	2	13**	11**	13**
Uterus - Adenoma	3	2	4	6
Uterus – Adenocarcinoma (includes multiple)	4**	10	15**	16**
Uterus - MMMT	0	4	0	2
Uterus - Adenoma, adenocarcinoma, or MMMT	6**	11	16**	19**

N= 50 **p≤0.01
MMMT – Malignant mixed Müllerian tumor

Molecular pathology investigation

- Molecular analysis of uterine adenocarcinomas was performed to better understand underlying genetic alterations in TBBPA-exposed animals
- *TP53* mutation is an important event in a large number of rodent and human cancers
  - TP53 mutation associated with some human endometrial cancers
- Mutational analysis of rat *Tp53* was performed
  - Spontaneous uterine carcinomas (5 NTP Wistar Han studies, n=10)
  - Uterine carcinomas from TBBPA-exposed rats (n=16)

Pattern of *Tp*53 mutations in Uterine adenocarcinomas in Wistar Han rats

	Dose (mg/kg)	Mutation Frequency
Total control incidence (%)	0	2/10 (20%)
TBBPA	250	3/3
	500	3/7
	1000	4/6
TBBPA all dose groups combined		**10/16 (63%)**

M. Hoenerhoff, Investigative Pathology Group -– studies based on analysis of DNA extracted from formalin fixed tissues
Silent mutations not included

TBBPA female rat – Ovary

Dose (mg/kg)	0	250	500	1000
Rete ovarii, cyst	1	0	6*	6*

N = 49-50 *p ≤0.05

TBBPA male rat – Testis

Dose (mg/kg)	0	250	500	1000
Germinal epithelium, atrophy	0	4	1	2
Interstitial cell, adenoma, bilateral	0	0	1	0
Interstitial cell, adenoma (includes bilateral)[a]	0*	0	1	3

N = 50 *p ≤0.05

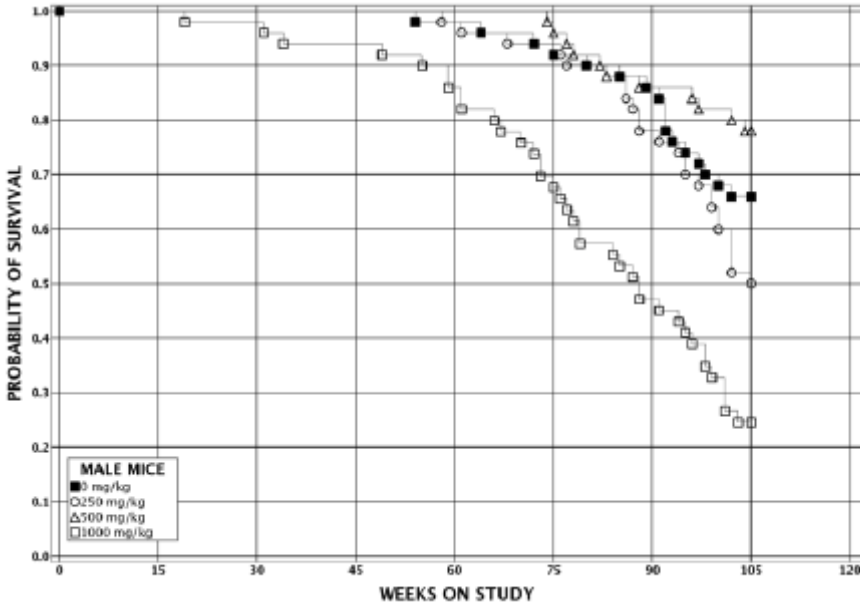
[a]Historical control incidence (all routes): 4/150 (2.7% ± 2.3%), range 0%-4%

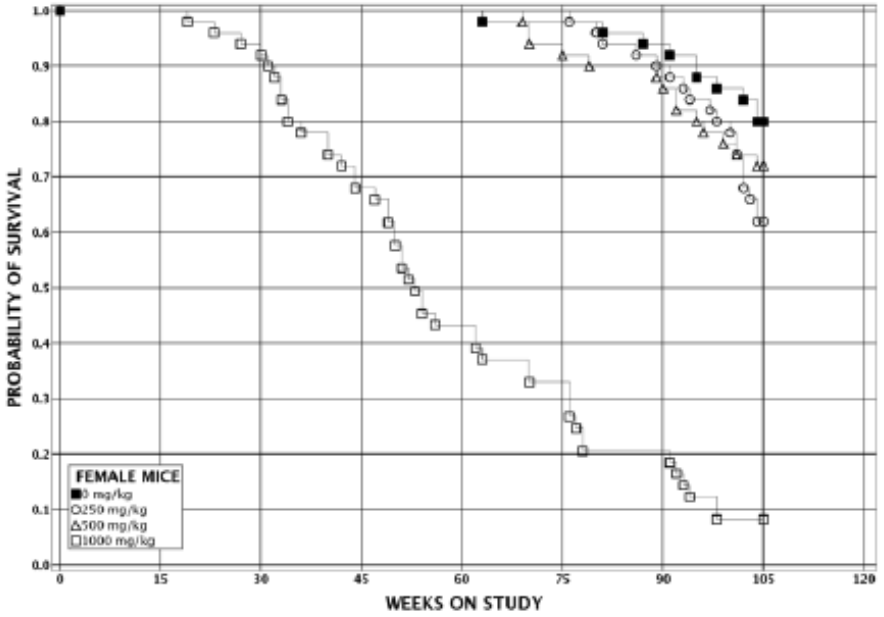
TBBPA 2-year mouse study
Final survival & mean body weight (day 701)

Dose (mg/kg)	0	250	500	1000
Male mice				
Survival	33**	26	39	12**
Mean BW (gm)	51.2	55.5	56.3	53.7
(% control)		(108%)	(110%)	(105%)
Female mice				
Survival	40**	31	36	4**
Mean BW (gm)	63.4	63.7	58.6	54.7
(% control)		(101%)	(92%)	(86%)

N = 50 **p ≤0.01

Survival curves - mice





TBBPA male mouse – Liver

Dose (mg/kg)	0	250	500
Clear cell focus	11	10	25**
Eosinophilic focus	20	33**	40**
Hepatocellular adenoma (multiple)	12	20	28*
Hepatocellular adenoma (includes multiple)	32	33	38
Hepatoblastoma[a]	2	11**	8
Hepatocellular carcinoma	11	15	17
Hepatocellular carcinoma or hepatoblastoma	12	24**	20

N = 50 *p ≤0.05, **p ≤0.01

[a]Historical control incidence (oral gavage): 9/250 (3.6% ± 2.6%), range 0%-6%
[a]Historical control incidence (all routes): 40/949 (4.2% ± 3.5%), range 0%-12%

TBBPA male mouse – Intestine

Dose (mg/kg)	0	250	500
Cecum or colon adenoma or carcinoma[a]	0*	0	3

N = 50 *p ≤0.05

[a]Historical control incidence (oral gavage): 0/250
[a]Historical control incidence (all routes): 4/950 (0.4% ± 0.8%), range 0%-2%

TBBPA male mouse – Hemangioma or hemangiosarcomas (all organs)

Dose (mg/kg)	0	250	500
Hemangioma	2	0	1
Hemangiosarcoma[a]	1*	5	8*
Hemangioma or Hemangiosarcoma	3*	5	9

N = 50 *p ≤0.05

[a]Historical control incidence (oral gavage): 28/250 (11.2% ± 6.4%), range 2%-18%
[a]Historical control incidence (all routes): 92/950 (9.7% ± 4.5%), range 2%-18%

TBBPA male mouse – Kidney

Dose (mg/kg)	0	250	500	1000
Renal tubule, cytoplasmic alteration	0	20**(1.9)[a]	47**(2.4)	46**(2.6)

N = 48-50 **p ≤0.01
[a]Severity grade

TBBPA male mouse – Forestomach

Dose (mg/kg)	0	250	500	1000
Ulcer	9(1.8)[a]	9(2.4)	19*(2.2)	28**(2.4)
Infiltration cellular, mononuclear cell	5(1.6)	8(1.8)	21**(2.1)	27**(2.3)
Inflammation	9(1.3)	10(1.7)	20*(2.2)	26**(2.3)
Epithelium hyperplasia	10(1.7)	13(2.2)	27**(2.8)	28**(2.7)

N = 48-50 **p≤0.01
[a]Severity grade

TBBPA female mouse - Forestomach

Dose (mg/kg)	0	250	500	1000
Ulcer	2(2.0)[a]	15**(2.0)	40**(2.2)	38**(2.1)
Infiltration cellular, mononuclear cell	2(3.0)	13**(2.2)	33**(2.4)	28**(1.8)
Inflammation	2(3.0)	14**(1.4)	41**(2.0)	37**(2.2)
Epithelium hyperplasia	4(2.5)	16**(2.6)	39**(3.0)	39**(2.3)

N = 48-50 **p≤0.01
[a]Severity grade

TBBPA Conclusions

- Male Wistar Han rat [Crl:WI(Han)]
  - Equivocal evidence of carcinogenic activity
    - Testicular interstitial cell adenomas
- Female Wistar Han rat [Crl:WI(Han)]
  - Clear evidence of carcinogenic activity
    - Uterine epithelium tumors (predominantly uterine adenocarcinomas)
- Male B6C3F1/N mice
  - Some evidence of carcinogenic activity
    - Hepatoblastomas
  - Large intestine tumors & hemangiosarcomas (all organs) may have been related to chemical administration
- Female B6C3F1/N mice
  - No evidence of carcinogenic activity

TBBPA Conclusions – treatment-related nonneoplastic lesions

- Female rats – uterus and ovary lesions
- Male mice – liver and kidney lesions
- Male and female mice – forestomach lesions