

iii) 90-Day Oral Toxicity Study in Beagle Dogs:

Test substance: Thiodiethylene bis (3,5-di-tert-butyl-4-hydroxyhydro cinnamate)
CAS No. 41484-35-9
Batch 68/3/0024/0 Assumed Purity >98%

Method: In this study, the selected animals were housed in kennels equipped with outside runs, four dogs of the same sex and group being accommodated in a single kennel. Test material was incorporated into a stock diet and fed to the dogs seven days a week at 10000, 20000, and 30000 ppm dietary levels. Diets were prepared fresh weekly. Initially, the body weight of each dog in every group was determined and recorded. Thereafter, weekly for the duration of the test. At the end of the investigation, the dogs were exsanguinated and all major tissues and organs were examined grossly. The weights of the following organs were obtained: liver, kidneys, heart, brain, spleen, gonads, adrenal glands, thyroid gland, and pituitary gland. The following tissues and organs were examined histologically: adrenal glands, bone marrow (sternum), brain (cerebrum, cerebellum, pons), caecum, colon, esophagus, gall bladder, gonads, heart, kidneys, liver, lungs, lymph nodes (cervical, mesenteric), muscle (skeletal), pancreas, pituitary gland, prostate gland, salivary gland (submaxillary), small intestine (duodenum, jejunum, ileum), spleen, stomach (cardia, fundus, pylorus), thyroid gland, uterus, urinary bladder.¹

Species/strain: Pure-bred Beagle dogs

Initial age of the animals: 6 months old

No.of animals per group: 4 males and 4 females of 4 groups

Route of administration: Dietary

Exposure period: 90-days

Dose: 0, 10000, 20000, and 30000 ppm

GLP: No

Year: 1973

Results: There were no mortalities. No clinical signs of toxicity were seen. No adverse effects were seen related to body weight gains, food consumption, behavioural reactions, urine analysis, gross pathologic studies, and histopathologic studies.

Hematologic studies revealed no significant differences between treated and untreated animals. Reticulocyte counts for dogs receiving 20000 or 30000 ppm averaged slightly higher than the untreated control values at the 84-day(final) determination. However, all values fall within normal ranges as seen in untreated dogs (0-40 reticulocytes per 1000 RBC) and these variations are not considered to be physiologically significant.

Blood chemistry studies (BUN, serum glucose, SGOT, SGPT) revealed no significant differences between treated and untreated animals.

Organ Weight Data:

Slightly elevated liver weights and liver to body and brain weight ratios were noted at all test levels in both male and female dogs. These changes were not dose related nor were any test material related liver histopathologic alterations. All remaining organ weights and ratios for test animals fall within normal limits.

Organ Weight Data
Liver - Males

Dietary Level (ppm)	Organ Weight (g)	Organ/ Body Weight Ratio (g / 1000 g)	Organ / Brain Weight Ratio (g/ g)
Control	330.00	33.33	4.06
	322.50	31.31	4.26
	400.50	30.80	4.87
	449.70	38.11	5.62
10000	507.20	49.24	7.18
	739.50	50.65	9.43
	507.20	47.40	7.14
	620.80	42.23	7.47
20000	529.10	46.41	6.21
	490.10	48.52	5.76
	616.00	51.76	7.46
	484.40	38.44	5.56
30000	456.90	41.53	5.27
	502.60	49.27	5.94
	406.80	43.27	5.40
	744.70	55.16	9.35

Organ Weight Data
Liver- Females

Dietary Level (ppm)	Organ Weight (g)	Organ/ Body Weight Ratio (g / 1000 g)	Organ / Brain Weight Ratio (g/ g)
Control	317.30	30.21	4.73
	345.70	34.56	4.16
	283.00	32.90	3.72
	293.50	34.12	3.74
10000	409.70	45.52	5.87
	399.20	38.01	5.54
	455.80	45.58	5.39
	482.10	42.66	5.85
20000	364.10	47.28	4.72
	471.10	55.42	6.64
	532.60	49.77	7.64
	479.00	40.94	5.91
30000	466.10	44.39	5.80
	510.70	53.75	6.15
	509.90	44.33	7.23
	498.20	48.84	6.83

Gross and histologic evaluation of a series of tissues from dogs of test groups were comparable to that of the control group. The changes in all tissues are compatible with those of naturally occurring diseases. These changes are present among both test and control animals.

The NOEL was found to be 2000 ppm.

Remarks:

This study was assigned a reliability code of 2c² (comparable to guideline study with acceptable restrictions).

Reference:

¹90-Day sub-acute oral toxicity study in Beagle dogs, IBT No. 651-03562, December 1973. Industrial BIO-TEST laboratories, Inc., Illinois.

²Klimisch, H.J., Andreae, M and Tillman, U. A systemic approach for evaluating the quality of experimental toxicological and ecotoxicological data. *Regulatory Toxicology and Pharmacology*. 25:1-5, 1997.