Abstract Number: **3704** | Poster Board Number: **P396** | www.jrfglobal.com

EVALUATION OF THE ENDOCRINE DISRUPTOR POTENTIAL OF FENBUTATIN OXIDE TECHNICAL IN WISTAR RATS

Patel, Manish V[a].; Poshiya, Mukesh P[a].; Ujawane, Deepak G[a].; Hadiya, Kishor C[a].; Piccirillo, Vincent J.[b]
[a] Department of Toxicology, Jai Research Foundation, Gujarat, India & [b] VJP Consulting, Inc., Ashburn, VA USA

JAI RESEARCH FOUNDATION

ABSTRACT

Potency of fenbutatin oxide technical (FBT) at 10 and 75 mg/kg/day for endocrine disruptor activity following US EPA guidelines was evaluated. For male and female pubertal assay, juvenile wistar rats of PND 23 and 22 were used at the time of initiation of dosing, respectively. For Hershberger and Uterotrophic bioassay, castration and ovariectomy was performed, respectively on PND 42 and 47. Study design (including number of animals/group, number of groups, study specific positive controls, frequency of observation, and duration of treatment) was as per the guideline. Organs, as defined in test guidelines, were excised and weighed from all animals. Significant reduction in body weight, body weight gain and feed consumption was observed in all animals treated with FBT at 75 mg/kg/day. Delay in onset of puberty was observed in male and female animals, treated with FBT at 75 mg/kg/day. In male pubertal assay and androgen antagonist bioassay, statistically significant reduction was observed in androgen dependent organs in animals treated with FBT at 75 mg/kg/day. In female pubertal assay, significant decrease in (pituitary, kidney, adrenal, ovaries, wet uterus, blotted uterus and thyroid) weight was observed in animals treated with FBT at 75 mg/kg/day. Corpus luteum was not present in ovaries of thirteen female animals treated with FBT at 75 mg/kg/day. In pubertal assays, thyroid hormone level in serum was comparable in all the groups whereas testosterone level was significantly reduced in the male animals treated with FBT at 75 mg/kg/day. In androgen agonist bioassay, all androgen dependent organ weights of animals treated with FBT was comparable with control group. In estrogen agonist bioassay, uterus weight (wet and blotted) of FBT treated groups was comparable with control group. In all the assays, endpoints of FBT groups treated at 10 mg/kg/day were comparable with concurrent control group. Hence, it is concluded that fenbutatin oxide technical does not alter pubertal development nor it affects androgen, estrogen and thyroid pathway at 10 mg/kg/day.

INTRODUCTION

Section 408(p) of the Federal Food Drug and Cosmetic Act (FFDCA) requires the EPA to develop a screening program, using appropriate validated test system and other scientifically relevant information, to determine if certain substances may have an effect in humans that is similar to an adverse effect produced by a naturally occurring estrogen, or other such endocrine effect. Subsequent to the passage of the Act, the EPA formed the Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC). In its final report, EDSTAC (1998) made several key recommendations:

- Examine effects of these chemicals on estrogen, androgen, and thyroid hormone-related processes
- Include pesticide and non-pesticide chemicals, contaminants, and (after evaluating single chemicals) mixtures
- Develop a two-tiered screening and testing strategy, now known as the Endocrine Disruptor Screening Program.

The EPA accepted the EDSTAC's recommendations for a two-tier screening program as proposed in a Federal Register Notice in 1998 (US EPA, 1998).

Tier 1 is composed of a screening battery of 6 in vivo {Uterotrophic (rat); Hershberger (rat); Pubertal female (rat); Pubertal male (rat); Amphibian metamorphosis (frog); Fish short-term reproduction} and 5 in vitro {Estrogen receptor (ER) binding – rat uterine cytosol; Estrogen receptor - (hERα) transcriptional activation - Human cell line (HeLa-9903); Androgen receptor (AR) binding - rat prostate cytosol; Steroidogenesis - Human cell line (H295R); Aromatase - Human recombinant microsomes} screening assays.

Fenbutatin oxide technical (FBT) is an organometal compound that is used as an acaricide. FBT was in first list of EDSP programme. FBT is practically non-toxic to bees and mammals during acute studies. It is highly toxic to aquatic organisms. FBT is a severe eye irritant (EPA 1994). It inhibits oxidative phosphorylation at the site of dinitrophenol uncoupling, which affects the production of energy in the form of adenosine triphosphate, ATP.

OBJECTIVE

Fenbutatin oxide technical (FBT) was evaluated for its endocrine disruption potential in several *in vivo* assays {Uterotrophic (rat); Hershberger (rat); Pubertal female (rat); Pubertal male (rat)} following US EPA guidelines.

Solvents and Chemicals

FBT was received from UPL, India. Testosterone propionate, flutamide and 17- α-ehtylene estradiol were purchased from Sigma-Aldrich, USA. Ketamine HCl Injection IP was purchased Toikaa pharmaceuticals Ltd. India. Xylazine was purchased from Indian Immunologicals Ltd., India. Meloxicam Injection was purchased from Intas Pharmaceuticals Ltd., India. Povidone Iodine was purchased from Win-Medicare, India.

STUDY DESIGN:

Tier-I *in vivo* Assay	Pubertal Male	Pubertal Female	Hershberger Agonist	Hershberger Antagonist	Uterotrohic Bioassay
Model	Pubertal Male	Pubertal Female	Castration	Castration	Ovariectomy
N° of animals/ Group	15	15	6	6	6
N° of Groups	1 - vehicle, 1 - positive, 2 - test item	1 - vehicle, 1 - positive, 2 - test item	1 - negative, 1 - positive, 2 - test item	1 - negative, 1 - positive, 3 - test item	1 - vehicle, 1 - positive, 2 - test item
PND at the Time of Dosing	23	22	50	50	57
Duration of Dosing (days)	31	21	10	10	3
Clinical Pathology	As per guidelines	As per guidelines	Not applicable	Not applicable	Not applicable
Hormone	Serum thyroid and testosterone	Serum thyroid	Not applicable	Not applicable	Not applicable
Organ Weight	As per guidelines	As per guidelines	As per guidelines	As per guidelines	As per guidelines
Microscopic Examination	As per guidelines	As per guidelines	Not applicable	Not applicable	Not applicable

RESULTS

Following results were obtained in the test item groups as compared with the vehicle/negative control group:

Tier-I *in vivo* Assay	Pubertal Male		Pubertal Female		Hershberger Agonist		Hershberger Antagonist			Uterotrohic Bioassay	
Dose Levels (mg/kg/day)	10	75	10	75	10	75	10	40	75	10	75
Clinical Sign	-	S	-	S	-	S	-	S	S	-	S
Body Weight	C	↓	C	↓	C	↓	C	↓	↓	C	↓
Body Weight Gain	C	↓	C	↓	C	↓	C	↓	↓	C	↓
Feed Consumption	C	↓	C	↓	C	↓	C	↓	↓	C	↓
Pubertal Development	C	D	C	D	-	-	-	-	-	-	-
Clinical Pathology	C	C	C	C	-	-	-	-	-	-	-
Hormone - Thyroid	C	C	C	C	-	-	-	-	-	-	-
Testosterone	-	↓	-	-	-	-	-	-	-	-	-
Androgen Dependent Organs	C	↓	-	-	C	C	↓	↓	↓	-	-
Ovary*, Uterus (Wet & blotted)	-	-	C	↓	-	-	-	-	-	C	C

Key: D = Delayed, C = Comparable, S = Salivation, - = Not applicable * = Only in Uterotrohic bioassay

CONCLUSION

Based on the results of studies, it is concluded that fenbutatin oxide technical does not alter pubertal development nor it affects androgen, estrogen and thyroid pathway at 10 mg/kg/day. However, it affects sexual maturation of animals at 75 mg/kg/day.

REFERENCE

United States Environmental Protection Agency. "Reregistration Eligibility Decision for Fenbutatin Oxide". September 1994. Accessed 05-11-11.