DEVELOPMENTAL THYROID TOXICITY STUDY OF ASULAM SODIUM TECHNICAL

Ujawane Deepak G.*1, Hadiya Kishor C.1, Poshiya Mukesh P.1, Rabadia Jay P.1, Parikh Foram K.1, Mistry Jaydip T.1, Shinde Akashrao1, Patel Manish V.1, Olson D.2, Bui Q.2 ¹Jai Research Foundation, India, www.jrfglobal.com; ²United Phosphorus, Inc., 630 Freedom Business Centre, King of Prussia, PA 19406



ABSTRACT

There are no validated and approved guidelines yet to investigate developmental thyroid toxicity. Asulam sodium technical (ASU) was administered through gavage from PND-8 to 21. Water (vehicle control) and 6-propyl-2-thiouracil (PTU) were administered to parent and pups on FND-4. Blood was collected from dams on GD-20 and LD-22. Analysis of T3, T4, and TSH was carried out by ELISA. Microscopic examination of thyroid (with parathyroid) was conducted for GD-20 and LD-22 dams. No treatment related groups in body weight, organ weight, serum hormone levels and microscopic examination in fetuses, pups and dams. Whereas in the PTU treated group, a significant decrease in TSH were noted in parental animals, in foetuses of GD-20, and in pups of PND-4 and PND-22. PTU treatment also caused significant decrease in body weight and increase in thyroid (with parathyroid) weight in dams, foetuses, pups and produced histopathological changes. Based on the results, it is concluded that the ASU is not a developmental thyroid toxicant. Results, observed in the positive control group (PTU), confirm that procedures used in this study met the criteria of a developmental thyroid study.

INTRODUCTION

Normal thyroid hormone blood levels are essential for growth and development of tissues and postnatal development in humans and animals, with major effects on growth, hearing, mental acuity, and development and function of reproductive system in the offspring.

Thyroid hormones can affect prenatal and postnatal development. Screening for thyroid disruption does not occur primarily because there is no definitive, easily measurable biologic end point apart from triiodothyronine (T3), tetraiodothyronine (T4), and thyroid stimulating hormone (TSH) serum concentrations and thyroid gland weight and histology.

There are no firm guidelines to draw conclusion, whether the chemical is a thyroid toxicant or not, so there is a need to establish specific thyroid developmental toxicity study or it can be included in various reproductive/developmental studies.

OBJECTIVE

This study was performed to evaluate the potential of asulam sodium technical to disrupt thyroid development and function in, foetus and postnatal offspring, post-administration of the test item, every day.

Chemicals: Asulam sodium technical (Manufacturer: Sigma-Aldrich), Sodium Chloride (Manufacturer: Fischer Scientific Pvt. Ltd.) were purchased.

Method: Study was divided into, below mentioned, two phases based on various end-points:

Sacrificed on Gestation Day-20: On gestation day 20, animals were subjected to blood collection through retro-orbital plexus under anaesthesia followed by sacrifice. Gravid uterus was excised and foetuses were subjected to blood collection through retro-orbital plexus under anaesthesia followed by sacrifice. for blood collection through decapitation. Remaining foetuses were subjected from all dams and 1 foetus/litter to observe microscopic changes. Blood samples collected from dam and foetuses were subjected to thyroid hormone evaluation.

Sacrificed on Lactation Day-22: All pups were weighed on PND-0, 4, 7-22. On lactation day 4, the size of each litter. Standardised pups were subjected to blood collection through decapitation for thyroid hormone evaluation. Selected pups were dosed with vehicle, ASU, and PTU, from PND-8 to 21, through gavage. On lactation day 22, surviving dams and pups were subjected to blood collection, through retro-orbital plexus, under anaesthesia, followed by sacrifice. Thyroid (with parathyroid) were collected from all dams and 1 male & 1 female pups/litter and evaluated for microscopic changes.

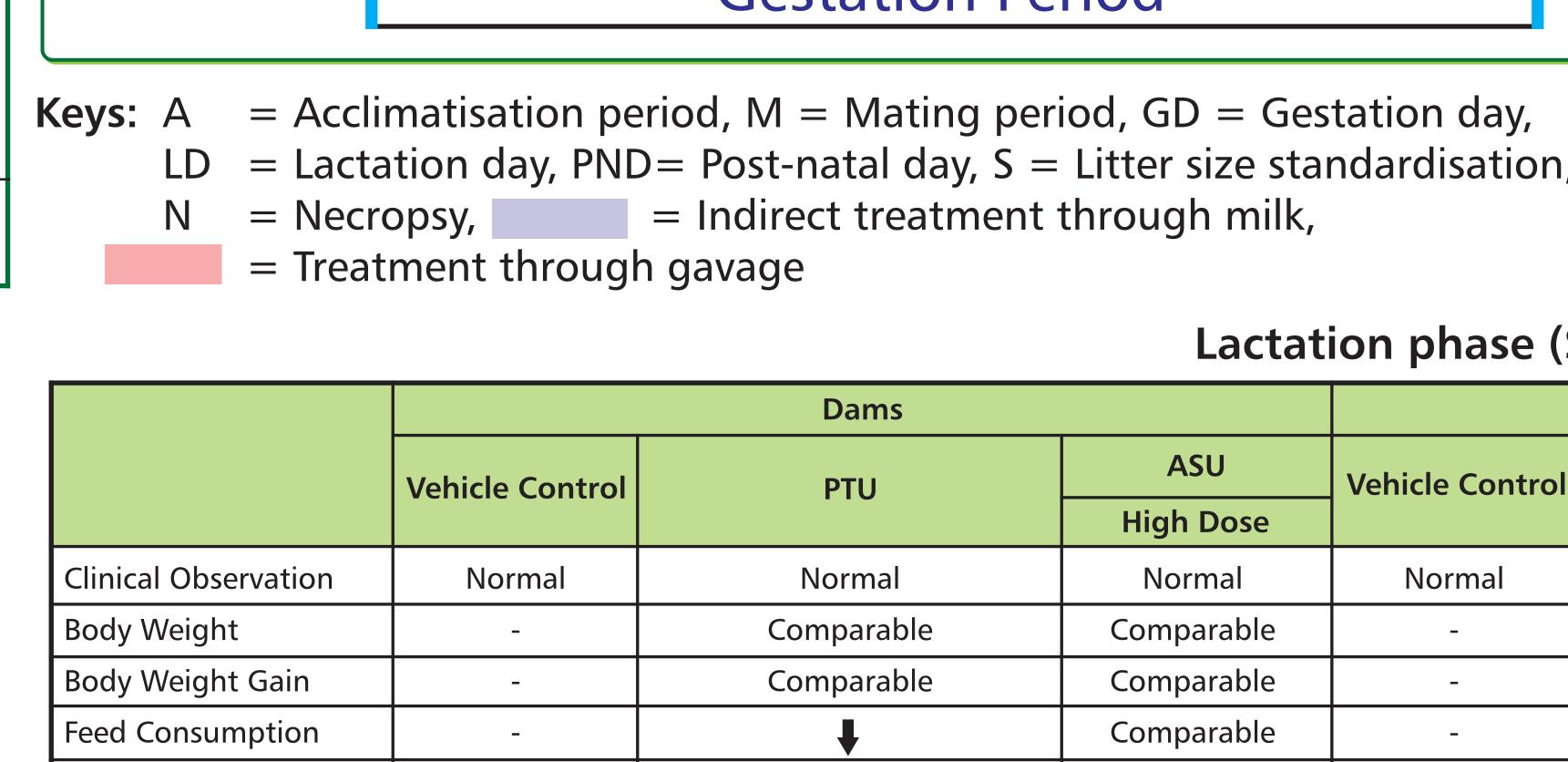
STUDY DESIGN

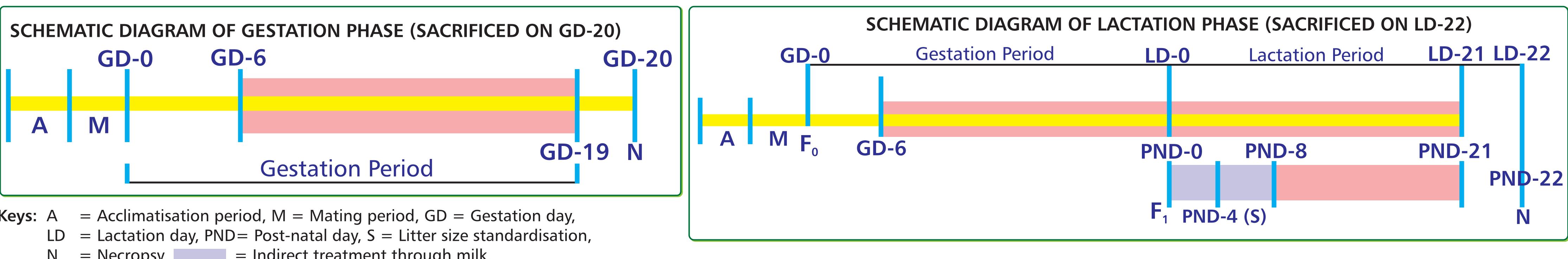
Phase	Dams Sacrificed	on GD-20	Dams Sacrificed	on LD-22	Pups Sacrificed on PND-22		
Group N°	Dose (mg/kg b. wt./day)	Treatment Period	Dose (mg/kg b. wt./day)	Treatment Period	Dose (mg/kg b. wt./day)	Treatment Period	
G1	0		NA	NA	NA	NA	
G2	1.0		NA	NA	NA	NA	
G3	30.0	GD-6 to 19	NA	NA	NA	NA	
G4	45.0		NA	NA	NA	NA	
G5	67.5		NA	NA	NA	NA	
G6	NA	NA	NA 0		0		
G7	NA	NA	1.0		1.0	PND-8 to 21	
G8	NA	NA	30.0	GD-6 to LD-21	30.0		
G9	NA	NA	45.0		45.0		
G10	NA	NA	67.5		67.5		
N° of Animals	25/group		30/group		8 pups/litter/group (4 male & 4 female pups)#		



RESULTS Gestation Phase: Sacrificed on Gestation day-20

	Observations								
Dawa sa at awa		P	ASU						
Parameters	Vehicle Control			Dam	Fetus				
		Dam	Fetus	High Dose	High Dose				
Mortality	Nil	Nil	Nil	Nil	Nil				
Clinical observation	Normal	Normal	-	Normal	Normal				
Body Weight	_	Comparable	Comparable	Comparable	Comparable				
Body Weight Gain	-	Comparable	-	Comparable	-				
Feed Consumption	-	•	-	Comparable	-				
Prenatal Data	Data - Cor		-	Comparable	-				
External Observation	-	-	NAD	-	NAD				
Visceral Observation	ral Observation		NAD	-	NAD				
T3 and T4 Level	nd T4 Level -		↓ (T4), ↑ (T3)	Comparable	Comparable				
TSH Level	-		1	Comparable	Comparable				
Organ Weight									
Liver	-	Comparable	-	Comparable	-				
Thyroid & parathyroid (absolute & relative)	-		-	- Comparable					
Gross Pathology	y NAD NAD		-	NAD					
Histopathology	NAD	Follicular cell hypertrophy and depletion of colloid in follicular lumen in both parental animals	Follicular cell hypertrophy and depletion of colloid in follicular lumen in both parental animals	NAD	NAD				





Lactation phase (Sacrificed on LD-22)

			Lacta	tion phase (s	defined on LD ZZ)					
	Dams			Pups Sacrificed on PND-4			Pups Sacrificed on PND-22		CONCLUSION	
	Vehicle Control	Control PTU	ASU	Vehicle Control	PTU	ASU	ASU High Dose Vehicle Control	PTU	ASU	
	Vernere control		High Dose			High Dose			High Dose	No adverse effect on the thyroid
Clinical Observation	Normal	Normal	Normal	Normal	-	_	Normal	Normal	Normal	structure, weight, and function was
Body Weight	_	Comparable	Comparable	-	_	-	-	↓ (M, F, M+F)	Comparable	noted in foetuses and pups in the present
Body Weight Gain	_	Comparable	Comparable	-	-	-	-	↓ (M, F, M+F)	Comparable	study using a procedure that maximises
Feed Consumption	_	•	Comparable	_	-	_	-	-	-	
Live birth Index	_	-	-	-	-	-	-	Comparable	Comparable	exposure to the test item (from in utero
Mortality Index	_	-	-	_	↓ (M+F)	Comparable	-	↑ (M, F, M+F)	Comparable	throughout lactation and postnatal
Survival Index	_	-	-	_	↑ (M+F)	Comparable	-	↓ (M, F, M+F)	Comparable	period). Hence, it can be concluded that
Lactation Index	_	_	_	_	_	_	-	↓ (M, F, M+F)	Comparable	
Litter size and Sex Rati	io -	-	-	_	Comparable	Comparable	-	Comparable	Comparable	asulam sodium technical has not
T3 and T4 Level	_	↓ (T4), ↑ (T3)	Comparable	_	↓ (T4)	Comparable	-	•	Comparable	produced any toxicity in the development
TSH Level	-	1	Comparable	-		Comparable	-	1	Comparable	(weight, morphology, and microscopic
Organ Weight	an Weight								structure) and function (serum T4, T3, and	
Liver	-	Comparable	Comparable	-	-	-	-	•	Comparable	
Thyroid & parathyroid (absolute & relative)	_	1	Comparable	-	-	_	-		Comparable	TSH) of the thyroid in young animals.
Gross Pathology	NAD	NAD	NAD	NAD	NAD	NAD	-	NAD	NAD	
Histopathology	NAD	Follicular cell hypertrophy, depletion of colloid in follicular lumen, decreased follicular lumen size, and hyperplasia of follicular cells	NAD	NAD	Follicular cell hypertrophy, depletion of colloid in follicular lumen, decreased follicular lumen size	NAD	NAD	Follicular cell hypertrophy, depletion of colloid in follicular lumen, decreased follicular lumen size, and hyperplasia of follicular cells	NAD	

Key: 🛊 = Significantly higher than control, 🎝 = Significantly lower than control, NAD = No abnormalities detected, M = Male, F = female, - = Not applicable Note: No effect were seen in low and mid dose when treated with ASU