Scientific Poster Session

n-lorem FOUNDATION

Nano-rare Patient Colloquium 2025

Monday, October 20 | 5:15 – 6:15 pm EST

From Sequence to Safety: Preclinical Assessment of Tolerability and Toxicology Profiles of ASOs

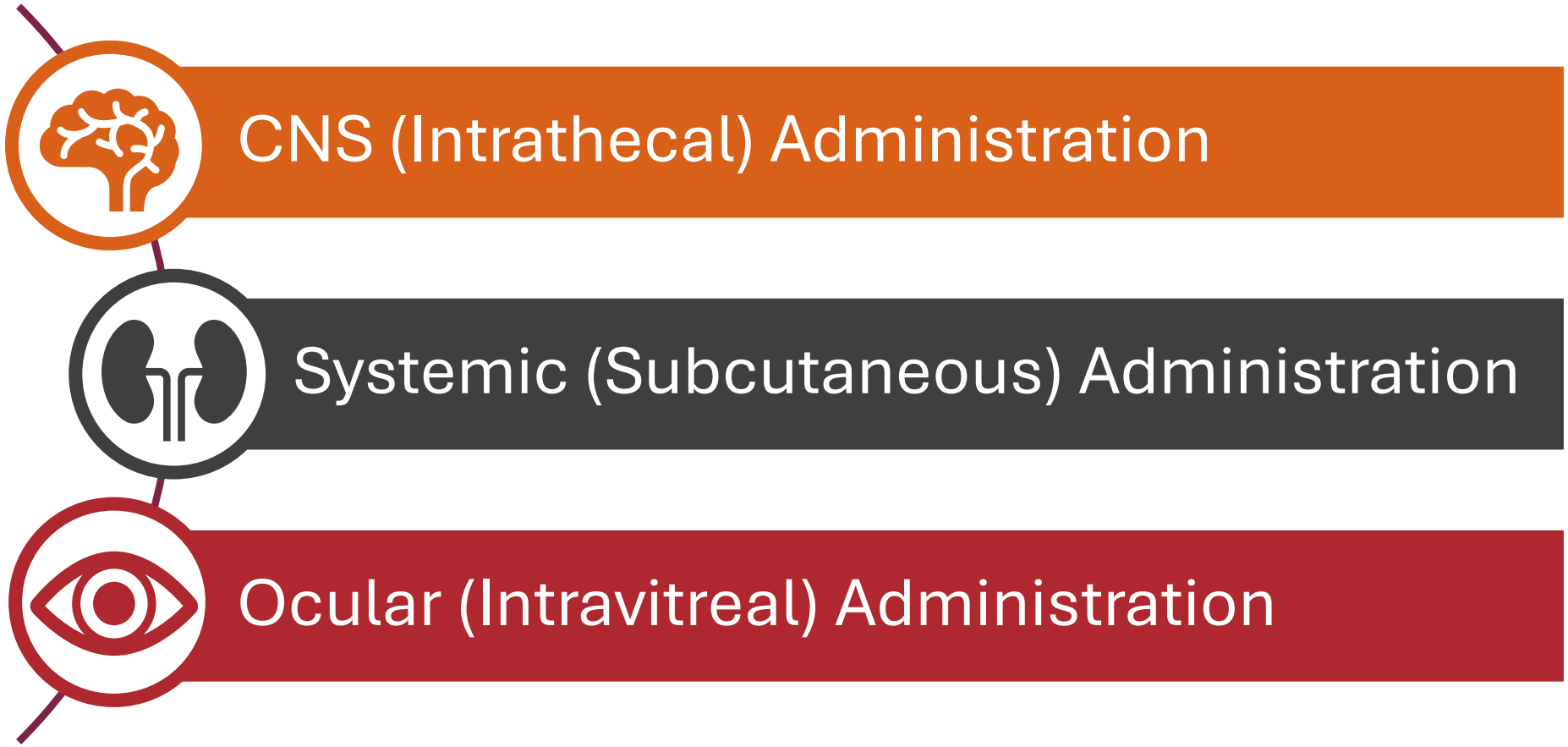
At n-Lorem, patient safety is the foundation of our discovery and development process. The development of optimal experimental ASOs depends on achieving excellent safety profiles, guided by rigorous tolerability and toxicology assessments throughout. This poster outlines n-Lorem's comprehensive preclinical development approach for creating well-tolerated ASOs with favorable toxicological profiles. It will detail standard class-related in vivo findings, the primary types of toxicities encountered, and key safety considerations at each stage of preclinical development.

Catherine Parisien, MSc

Senior Scientist/Toxicologist, Preclinical Development, n-Lorem

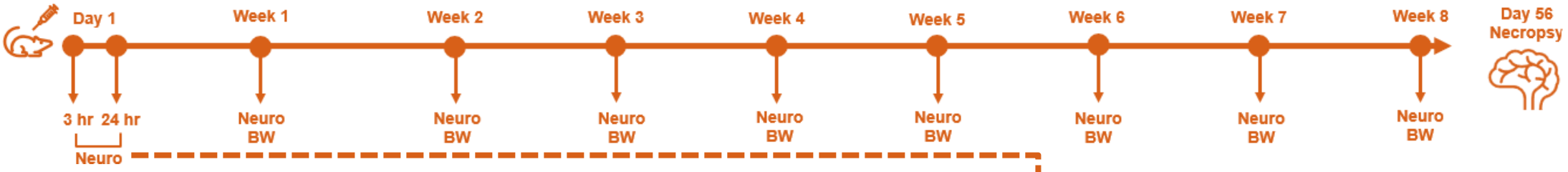
Introduction

Preclinical Assessment of Tolerability and Toxicology Profiles of ASOs



CNS (Intrathecal) Administration

non-GLP 8-Week Single Intrathecal Dose Tolerability Study of ASOs in Rats

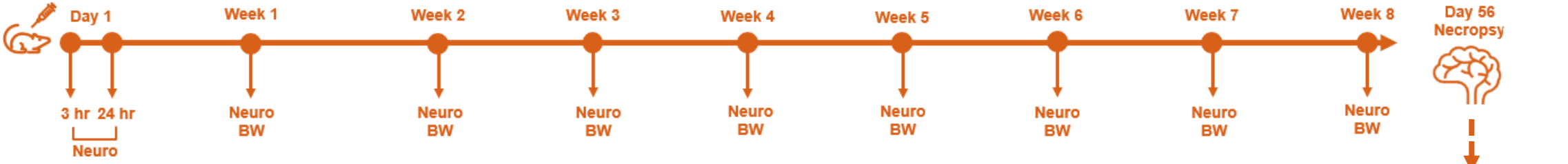


IT Administration - ASO-Related Findings (Single Dose)	
Measure/Marker	**ASO-Related Findings**
3 & 24 hr neurobehavioral evaluation	Acute sedation & paresis
Delayed (weekly) neurobehavioral evaluation	Delayed paresis
Body weight changes	Body weight loss Decreased body weight gain
IBA1 mRNA & IHC	Neuroinflammation - Microgliosis
GFAP mRNA	Neuroinflammation - Astrogliosis
Calbindin IHC	Neurotoxicity – Purkinje cell loss

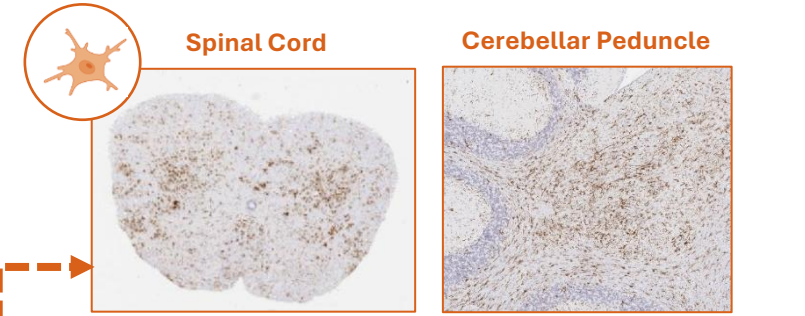
Neurobehavior Scoring System (Neuro)	
Score	**Description**
0	Normal
1	Limp tail OR hind-end weakness
2	Limp tail AND hind-end weakness
3	Unable to support hind-end
4	Complete hind-end paralysis
5	Unable to support body
6	Unresponsive
7	Found dead

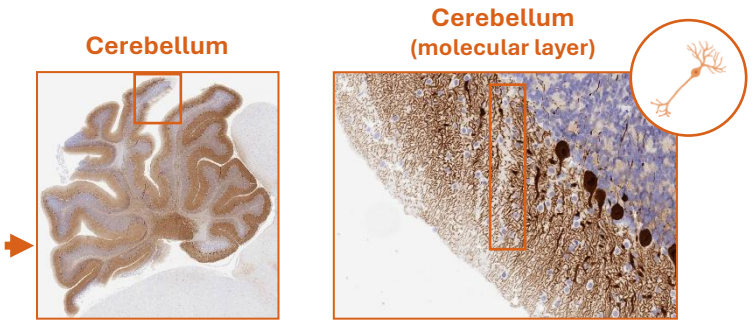
CNS (Intrathecal) Administration

non-GLP 8-Week Single Intrathecal Dose Tolerability Study of ASOs in Rats



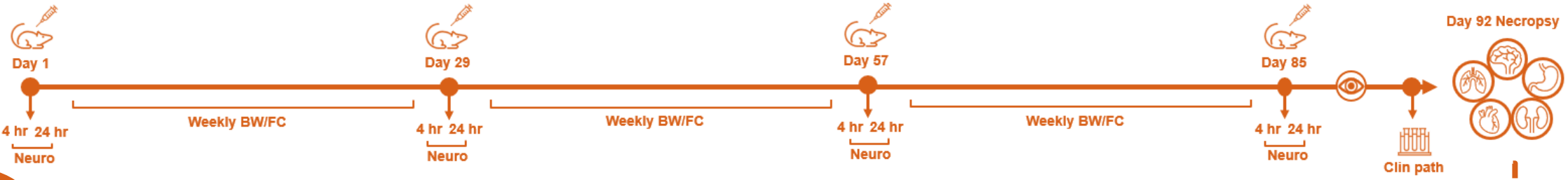
IT Administration - ASO-Related Findings (Single Dose)	
Measure/Marker	**ASO-Related Findings**
3 & 24 hr neurobehavioral evaluation	Acute sedation & paresis
Delayed (weekly) neurobehavioral evaluation	Delayed paresis
Body weight changes	Body weight loss Decreased body weight gain
IBA1 mRNA & IHC	Neuroinflammation - Microgliosis
GFAP mRNA	Neuroinflammation - Astrogliosis
Calbindin IHC	Neurotoxicity – Purkinje cell loss





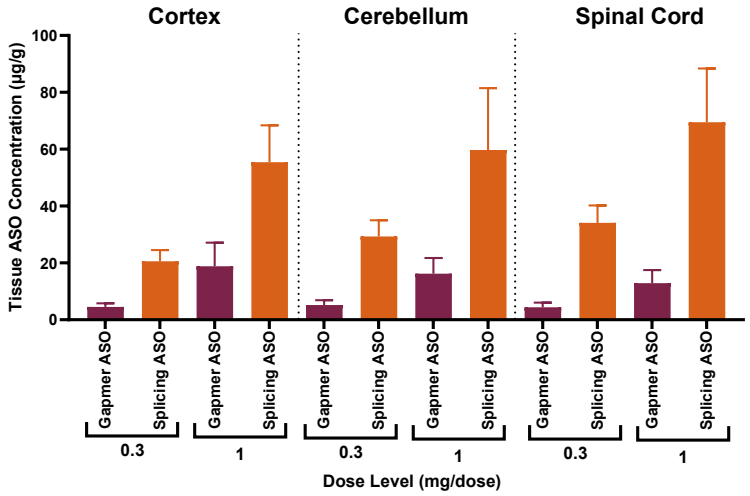
CNS (Intrathecal) Administration

GLP 13-Week Repeat Intrathecal Dose Toxicity Study of ASOs in Rats



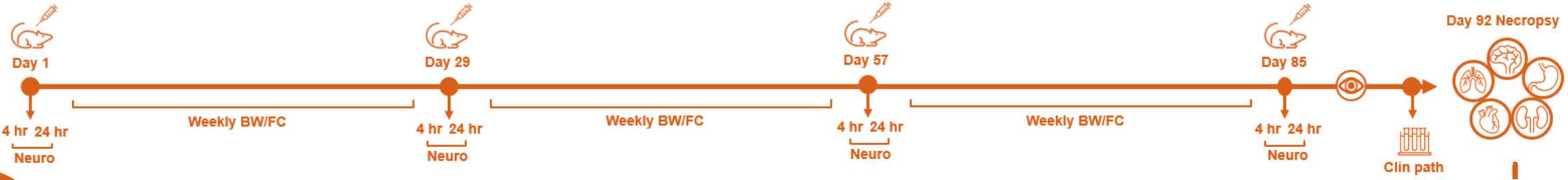
IT Administration - ASO-Related Findings (Repeat Dose)	
Measure	**ASO-Related Findings**
Clinical observations	Transient postdose sedation & paresis
Neurobehavioral evaluations	
Body weight changes	Body weight loss/decreased body weight gain
Food consumption	Decrease – correlating to body weight effect
Ophthalmology	None
Clinical pathology	Rare
Organ weights	None
Macroscopic observations	None
Microscopic observations	Secondary to the uptake and accumulation of ASO

Tissue Bioanalysis



CNS (Intrathecal) Administration

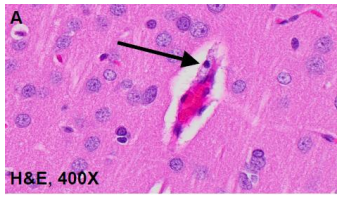
GLP 13-Week Repeat Intrathecal Dose Toxicity Study of ASOs in Rats

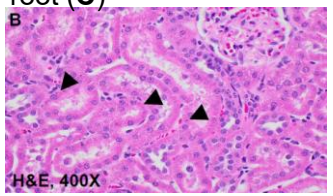


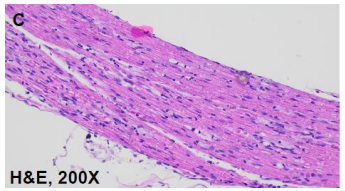
IT Administration - ASO-Related Findings (Repeat Dose)	
Measure	**ASO-Related Findings**
Clinical observations	Transient postdose sedation & paresis
Neurobehavioral evaluations	
Body weight changes	Body weight loss/decreased body weight gain
Food consumption	Decrease – correlating to body weight effect
Ophthalmology	None
Clinical pathology	Rare
Organ weights	None
Macroscopic observations	None
Microscopic observations	Secondary to the uptake and accumulation of ASO

Histopathology

Vacuolated macrophage infiltration in CNS tissues (**A**), basophilic granules within renal tubule epithelial cells (**B**), nerve fiber degeneration, nerve root (**C**)

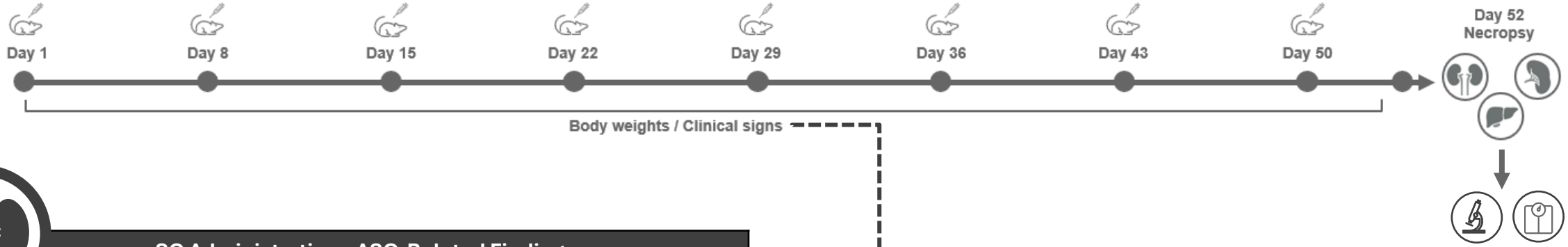






Systemic (Subcutaneous) Administration

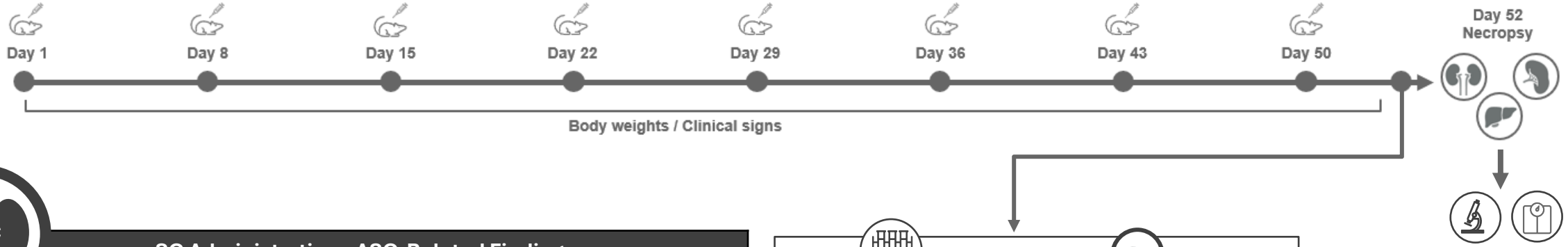
non-GLP 8-Week Repeat Subcutaneous Dose Tolerability Study of ASOs in Mice



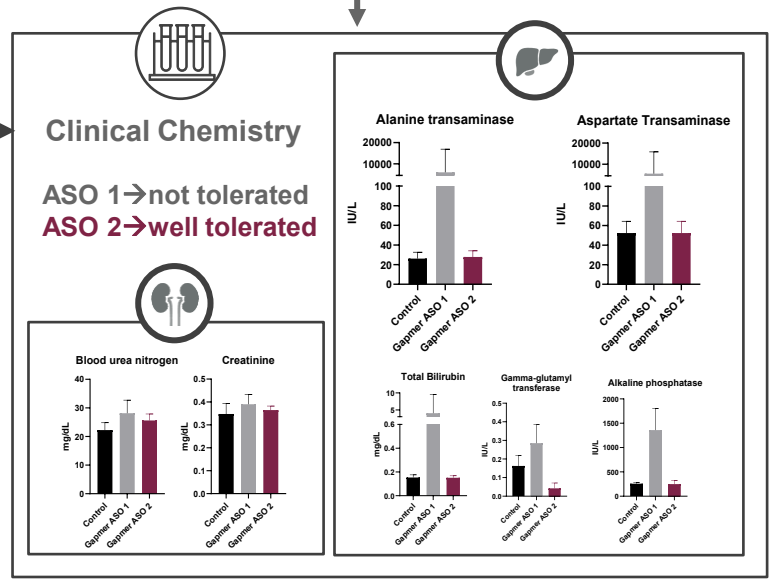
SC Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Clinical observations	Jaundice (correlating to elevated liver enzymes)
Body weight changes	Body weight loss/decreased body weight gain
Clinical chemistry	Increased liver and kidney markers (e.g. ALT)
Organ weights	Increased liver and spleen weights
Macroscopic observations	None
Histopathology	Findings related to tissue accumulation and/or pro-inflammatory effects

Systemic (Subcutaneous) Administration

non-GLP 8-Week Repeat Subcutaneous Dose Tolerability Study of ASOs in Mice

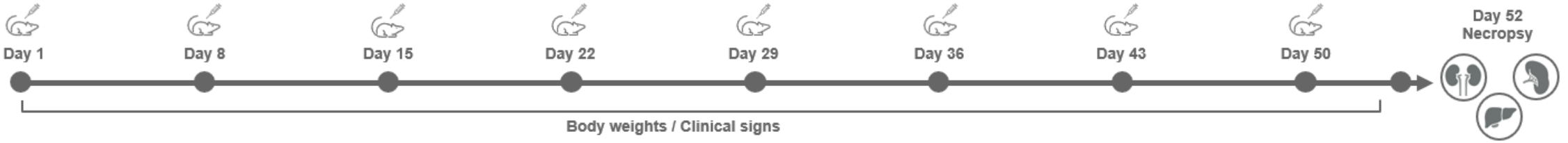


SC Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Clinical observations	Jaundice (correlating to elevated liver enzymes)
Body weight changes	Body weight loss/decreased body weight gain
Clinical chemistry	Increased liver and kidney markers (e.g. ALT)
Organ weights	Increased liver and spleen weights
Macroscopic observations	None
Histopathology	Findings related to tissue accumulation and/or pro-inflammatory effects

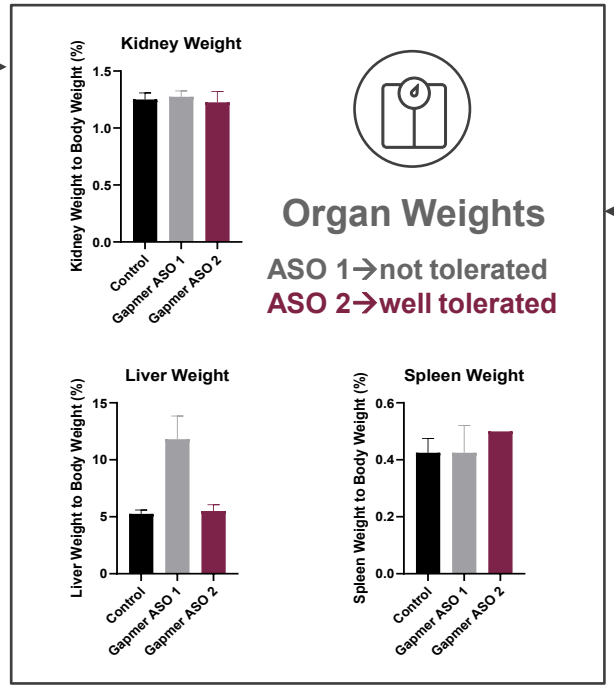


Systemic (Subcutaneous) Administration

non-GLP 8-Week Repeat Subcutaneous Dose Tolerability Study of ASOs in Mice

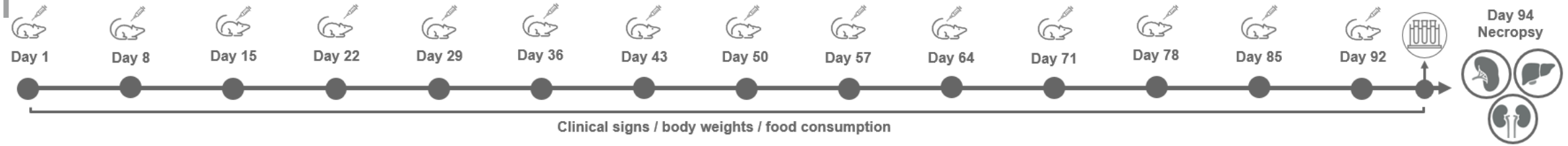


SC Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Clinical observations	Jaundice (correlating to elevated liver enzymes)
Body weight changes	Body weight loss/decreased body weight gain
Clinical chemistry	Increased liver and kidney markers (e.g. ALT)
Organ weights	Increased liver and spleen weights
Macroscopic observations	None
Histopathology	Findings related to tissue accumulation and/or pro-inflammatory effects

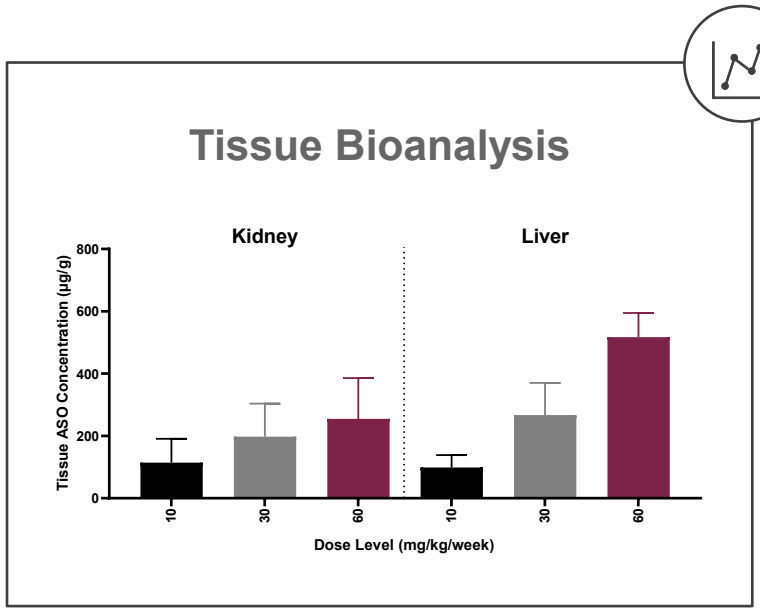


Systemic (Subcutaneous) Administration

GLP 13-Week Repeat Subcutaneous Dose Toxicity Study of ASOs in Mice

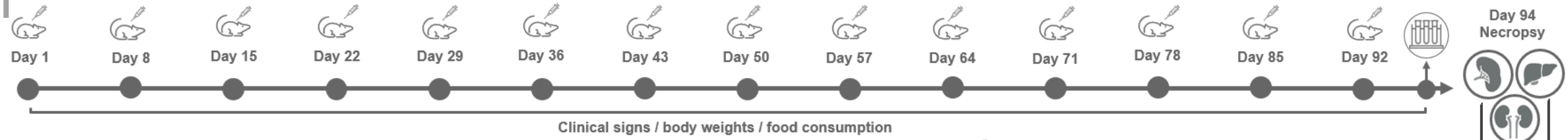


SC Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Clinical observations	Rare
Body weight changes	Rare - Body weight loss/decreased body weight gain
Food consumption	Rare - Decreased – correlating to body weight effect
Clinical chemistry	Increased liver and kidney markers (e.g. ALT)
Hematology	Pro-inflammatory response (e.g. increases in absolute neutrophils, monocytes, and eosinophils)
Organ weights	Rare - increased liver and spleen weights
Macroscopic observations	None
Histopathology	Findings related to tissue accumulation and/or pro-inflammatory effects



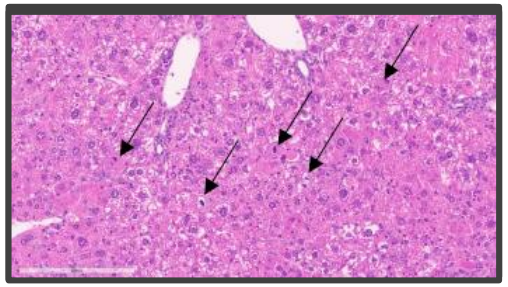
Systemic (Subcutaneous) Administration

GLP 13-Week Repeat Subcutaneous Dose Toxicity Study of ASOs in Mice



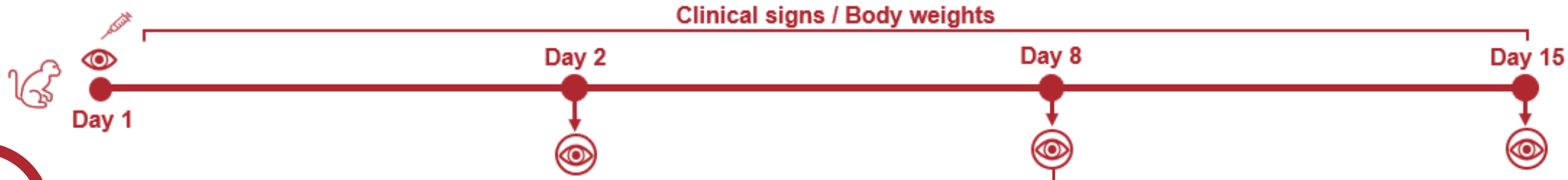
SC Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Clinical observations	Rare
Body weight changes	Rare - Body weight loss/decreased body weight gain
Food consumption	Rare - Decreased – correlating to body weight effect
Clinical chemistry	Increased liver and kidney markers (e.g. ALT)
Hematology	Pro-inflammatory response (e.g. increases in absolute neutrophils, monocytes, and eosinophils)
Organ weights	Rare - increased liver and spleen weights
Macroscopic observations	None
Histopathology	Findings related to tissue accumulation and/or pro-inflammatory effects

ASO-Related Histopathology Findings
Increased cellularity
Hepatocyte karyomegaly ★
Increased hepatocyte mitotic activity ★

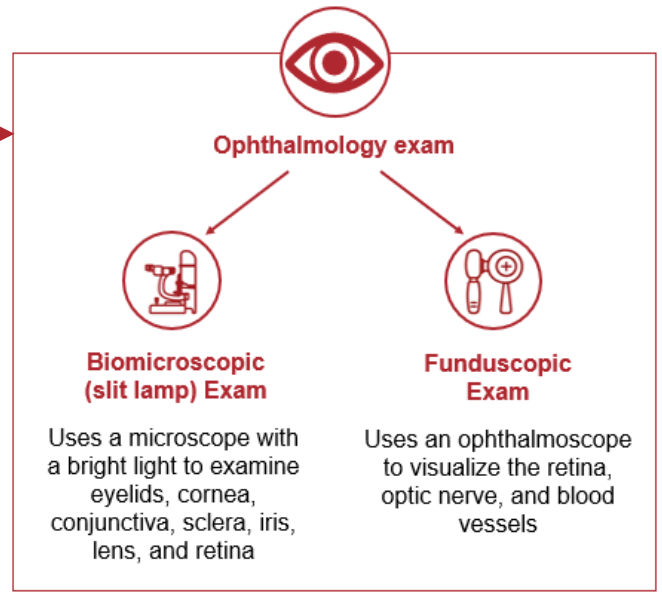


Ocular (Intravitreal) Administration

2-Week Single Intravitreal (IVT) Dose Tolerability Study of ASOs in Monkeys

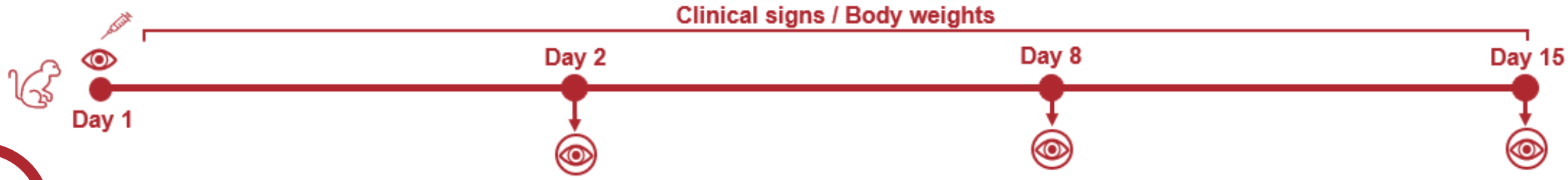


IVT Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Ophthalmology	Ocular inflammation
Body weight changes	None
Clinical observations	Rare (e.g. cloudy eyes)



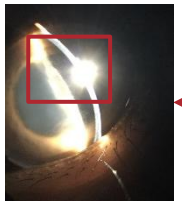
Ocular (Intravitreal) Administration

2-Week Single Intravitreal (IVT) Dose Tolerability Study of ASOs in Monkeys



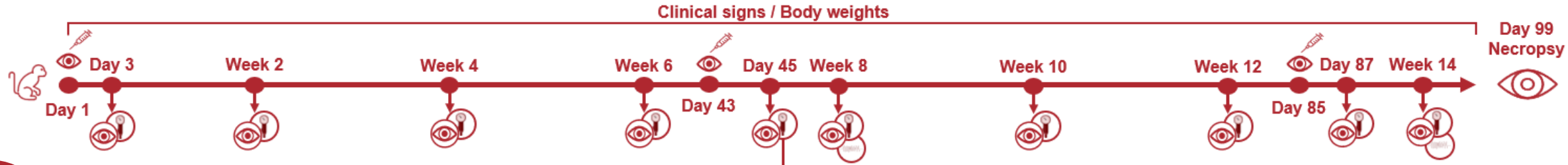
IVT Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Ophthalmology	Ocular inflammation
Body weight changes	None
Clinical observations	Rare (e.g. cloudy eyes)

Common ASO-related Ophthalmology Findings	
Anterior chamber inflammation	Anterior chamber cells
	Fundus, hazy view
	Fibrin
	Keratic precipitates
	Decreased pupillary reaction
	Flare
Vitreous inflammation	Vitreous cell-like opacities
	Vitreous haze
Iris	Miosis
	Irregular margin

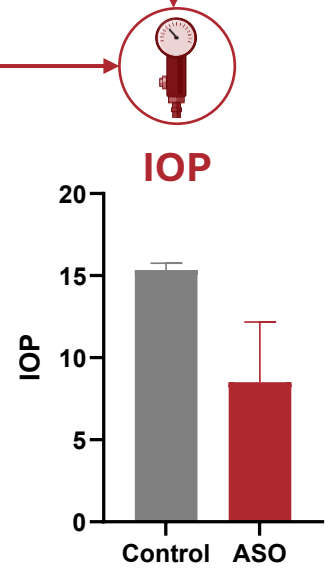


Ocular (Intravitreal) Administration

14-Week Repeat Intravitreal Dose Toxicity Study of ASOs in Monkeys

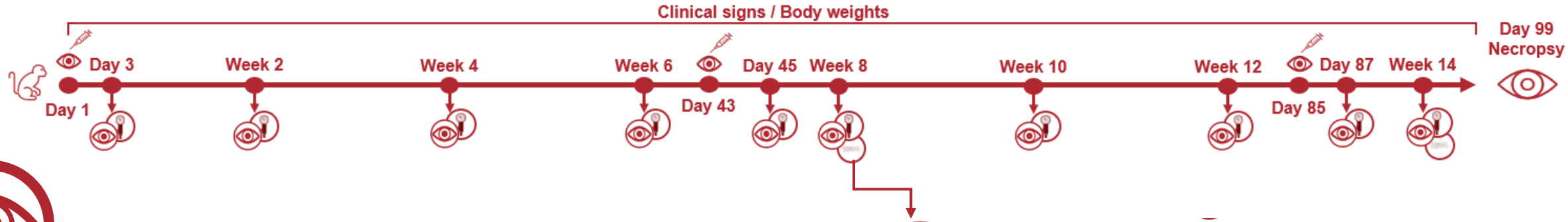


IVT Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Ophthalmology	Ocular inflammation
Body weight changes	None
Clinical observations	Rare (e.g. cloudy eyes)
Electroretinogram (ERG)	Abnormal findings correlating to inflammation
Intraocular pressure (IOP)	Decreases – correlating to inflammation
Macroscopic findings	None
Microscopic findings	Changes related to proinflammatory effects



Ocular (Intravitreal) Administration

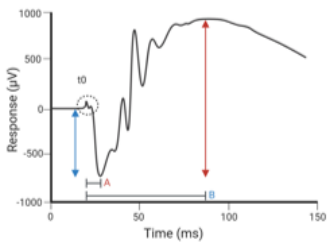
14-Week Repeat Intravitreal Dose Toxicity Study of ASOs in Monkeys

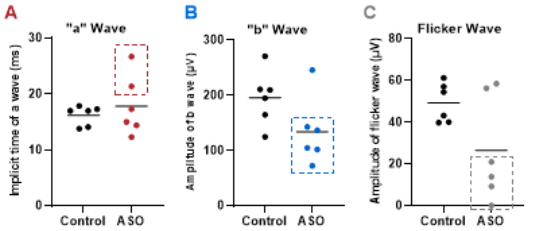


IVT Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Ophthalmology	Ocular inflammation
Body weight changes	None
Clinical observations	Rare (e.g. cloudy eyes)
Electroretinogram (ERG)	Abnormal findings correlating to inflammation
Intraocular pressure (IOP)	Decreases – correlating to inflammation
Macroscopic findings	None
Microscopic findings	Changes related to proinflammatory effects

Electroretinography

Measures the electrical activity of the retina in response to a light stimulus and is an objective measure of retinal function





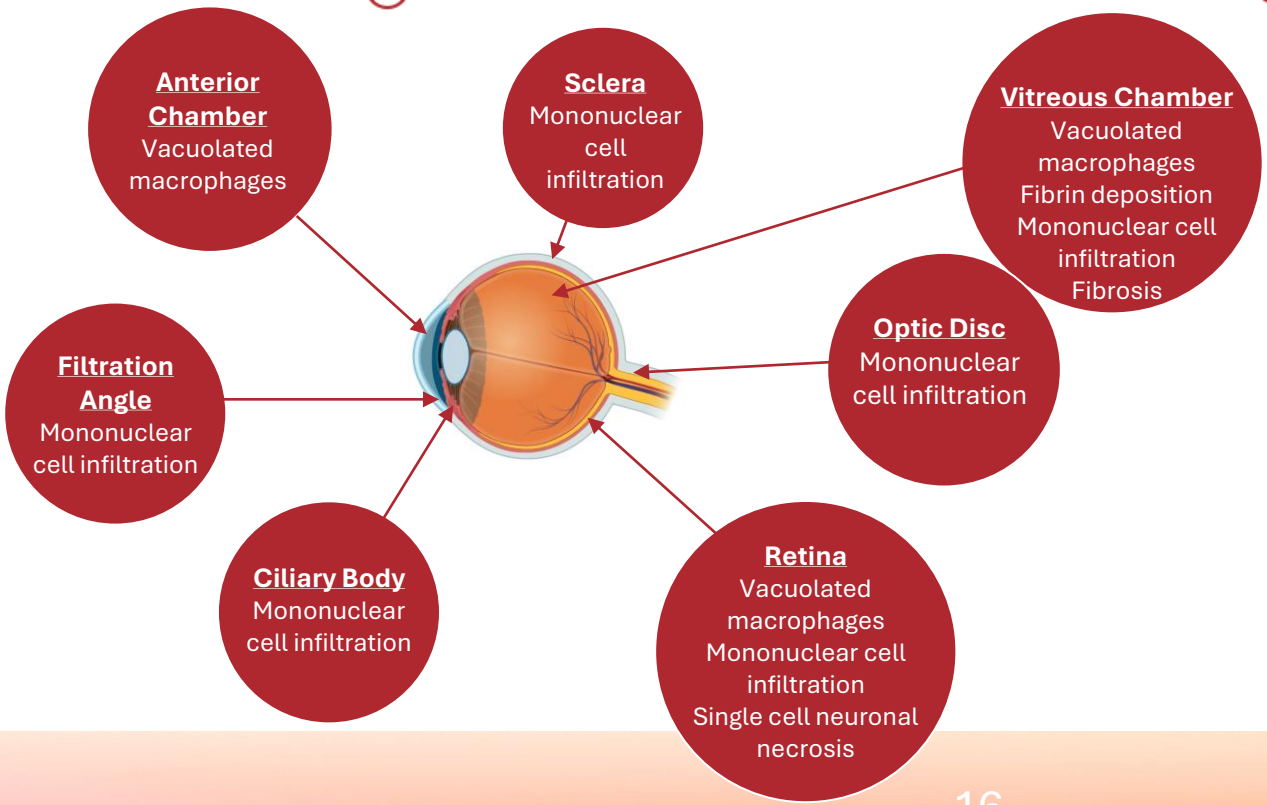
Abnormal findings in ERG parameters correlate with ocular inflammation:

A. Implicit time of "a" wave delayed
B. Amplitude of "b" wave decreased
C. Amplitude of flicker wave decreased

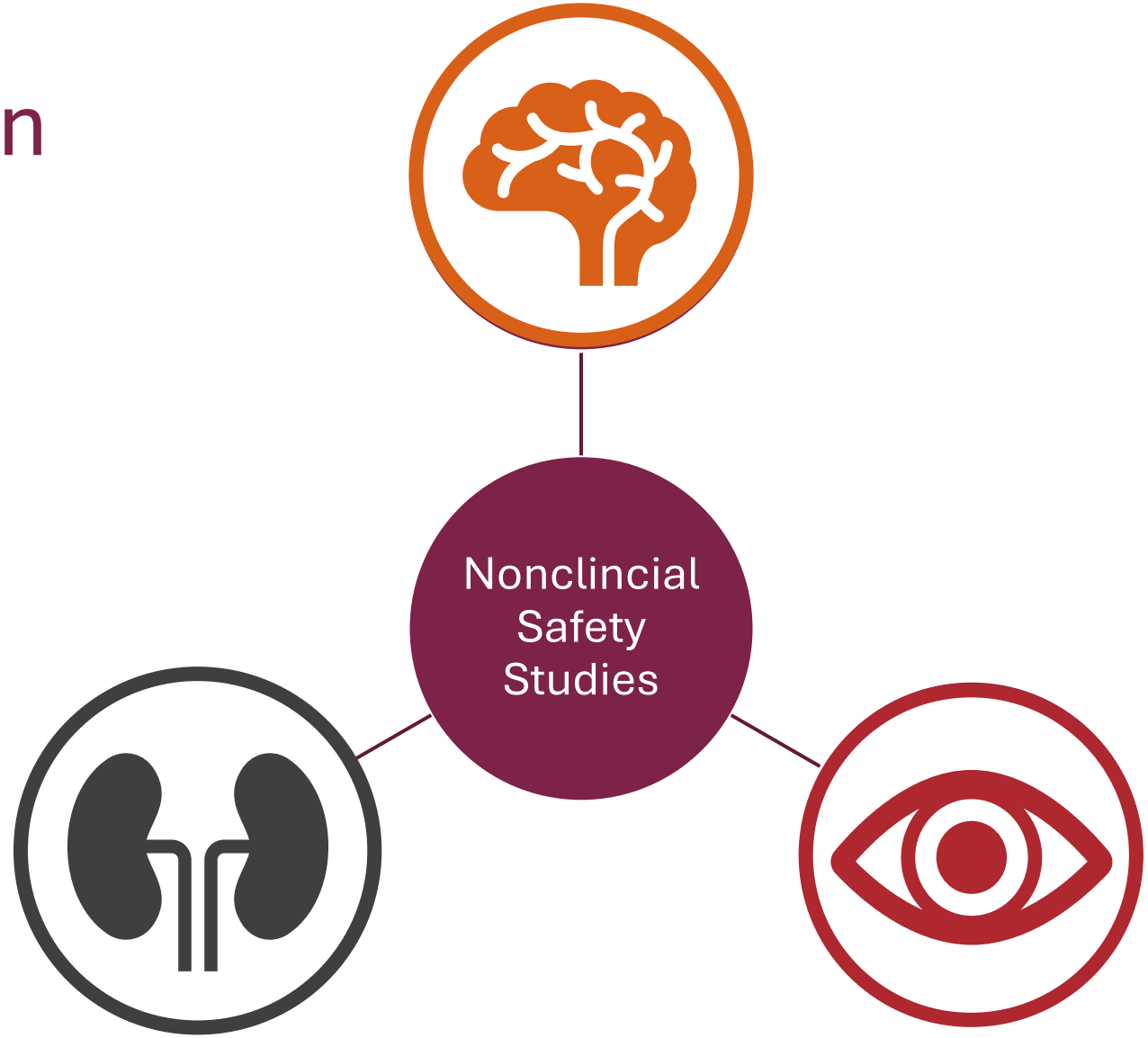
Ocular (Intravitreal) Administration

14-Week Repeat Intravitreal Dose Toxicity Study of ASOs in Monkeys

IVT Administration - ASO-Related Findings	
Measure/Marker	**ASO-Related Findings**
Ophthalmology	Ocular inflammation
Body weight changes	None
Clinical observations	Rare (e.g. cloudy eyes)
Electroretinogram (ERG)	Abnormal findings correlating to inflammation
Intraocular pressure (IOP)	Decreases – correlating to inflammation
Macroscopic findings	None
Microscopic findings	Changes related to proinflammatory effects



Conclusion



Acknowledgements

We would like to thank all of our CROs, partners, and the whole n-Lorem team, who are making this work possible.

Thank you

n-lorem FOUNDATION