

I concur with this review. A. Shearin. 2/21/25

I concur with this review memo. A. Wensky 2/24/2025

FOOD AND DRUG ADMINISTRATION
Center for Biologics Evaluation and Research
Office of Therapeutic Products
Office of Pharmacology/Toxicology

BLA NUMBER:	STN #125798.000
DATE RECEIVED BY CBER:	04/18/2024
DATE REVIEW COMPLETED:	02/21/2025
PRODUCT:	ENCELTO (revakinagene taroretcel)
APPLICANT:	Neurotech Pharmaceuticals, Inc.
PROPOSED INDICATION:	Idiopathic Macular Telangiectasia Type 2 (MacTel)
PHARM/TOX REVIEWER:	Ernesto Moreira
PHARM/TOX TEAM LEADER:	Margaret Benny Klimek
PHARM/TOX BRANCH CHIEF:	Abigail Shearin
PRODUCT (CMC) REVIEWERS:	Carolina Panico; Kyung Sung (Device); Shirin Marfatia; Edhriz Siraliev-Perez
CLINICAL REVIEWERS:	Ekaterina Tsilou
PROJECT MANAGER:	Crystal Melendez
DIVISION DIRECTOR:	Allen Wensky
OFFICE DIRECTOR:	Nicole Verdun

EXECUTIVE SUMMARY:

ENCELTO (revakinagene taroretcel) is a biologic-device combination product consisting of a semi-permeable capsule loaded with recombinant human ciliary neurotrophic factor (rhCNTF) secreting NTC-201-6A cells. ENCELTO is intended for surgical implantation by intravitreal (ITV) insertion via a pars plana sclerotomy. ENCELTO is intended to treat adult patients with idiopathic macular telangiectasia type 2 (MacTel) by providing rhCNTF. Following ITV implantation of ENCELTO, rhCNTF arrests the death of photoreceptors characteristic of MacTel. Due to the extremely short half-life of rhCNTF when administered intravenously¹, the applicant has developed ENCELTO, to secrete rhCNTF. The recommended dose of ENCELTO is 1 implant per eye, each containing 200,000-440,000 allogeneic NTC-201-6A cells expressing rhCNTF.

¹ Dittrich F, Thoenen H, Sendtner M. Ciliary neurotrophic factor: pharmacokinetics and acute phase response in rat. Ann Neurol. 1994;35(2):151-63.

The nonclinical development program evaluated a product representative of ENCELTO, NT-501. An early pharmacology study showed that the outer nuclear layer (ONL) in dogs with retinitis pigmentosa caused by rod-cone dysplasia type 1 (rcd1) was protected from photoreceptor loss following implantation of NT-501. A pharmacology study in healthy (b) (4) rabbits showed that secretion of 5 ng/day of rhCNTF following NT-501 implantation had no deleterious effects on photoreceptors, but that high doses of 22 ng/day of rhCNTF could have negative effects on cones.

The nonclinical pharmacokinetic (PK) studies measured systemic exposure of rhCNTF in two distribution studies in rabbits and two toxicology studies in minipigs and pigs. There was no evidence of systemic exposure to rhCNTF after ITV implantation of NT-501 in minipigs for up to 6 months or in rabbits for up to 9 months. In rabbits, NT-501 implants established CNTF levels averaging about 7-9 ng/day for up to 9 months.

The effect of long-term ITV implantation of NT-501 was evaluated in a 3-part toxicology study in 4–8-month-old (b) (4) minipigs, 4–10-year-old (b) (4) pigs, and catastrophic device failure of NT-501 was evaluated by ITV injection of unencapsulated NTC-201-6A cells into 8-9 month old (b) (4) minipigs. Overall, data from this study suggests that the effects of NT-501 implantation were minimal: lens changes, focal refractive changes in the vitreous, and minimal to mild amounts of inflammatory cells and/or inflammation associated with the vitreous, the aqueous chamber, iris/ciliary body, and the corneoscleral junction. Although many of these appear to be triggered by the implantation of the NT-501 capsule itself or the implantation procedure, a dose-response to rhCNTF was present, with eyes implanted with empty capsules and capsules with a low CNTF output cell line exhibiting less intense changes than eyes implanted with NT-501. Microscopic analysis of ocular tissues corroborated these in-life findings. Intraocular pressures (IOP), pupillary response, cornea, and other clinical ophthalmic parameters were normal. ITV implantation of NT-501 did not cause any effects on systemic clinical signs, gross pathology, or histopathology of non-ocular organs.

Four Developmental and Reproductive Toxicity (DART) studies were conducted in rats and rabbits to establish the risks to fertility and teratogenicity associated with the subcutaneous (SC) administration of high doses of rhCNTF. In male rats administered rhCNTF at dose levels of 0, 10, 100, or 300 µg/kg/day subcutaneously (SC) for 62 days, there were no adverse effects on mating performance, fertility, or the postnatal development of the offspring. In female rats administered rhCNTF at dose levels of 0, 10, 100, or 300 µg/kg/day SC for 2 weeks prior to mating to postpartum Day 21, mating performance, fertility and gestational parameters were normal. No adverse effects on fetuses or pup postnatal development were reported. In pregnant rats administered rhCNTF at 10, 100, 300, or 1000 µg/kg/day SC on gestational Days 7-21, clinical changes were present in pregnant rats administered the highest dose level and decreased body weight gain was present at dose levels ≥ 100 µg/kg/day of rhCNTF. There were no rhCNTF-related teratologic changes reported in the fetuses. In pregnant rabbits administered SC rhCNTF at 2, 5, or 10 µg/kg/day SC on gestational Days 7-29, anorexia, abortion, and body weight loss occurred at 10 µg/kg/day. There were no rhCNTF-related teratologic changes reported in the fetuses.

A battery of genotoxicity studies was conducted to support licensure of NT-501 in keeping with ISO-10993, guidance for biological evaluation of medical devices. These studies included a (b) (4) study, (b) (4) study in mammalian cells, mouse bone marrow micronucleus study, and a Kligman maximization test. No genotoxicity was observed in these studies.

Studies to evaluate the carcinogenicity/tumorigenicity of ENCELTO were not conducted. These studies are not warranted based on the safety profile described in the provided toxicological risk assessments (TRAs).

PHARMACOLOGY/TOXICOLOGY RECOMMENDATION:

There are no nonclinical deficiencies related to the animal studies identified in this submission. There are no outstanding requests for additional nonclinical animal data or major deficiencies identified for evaluation of ENCELTO. The nonclinical information provided in the BLA submission supports approval of the licensure application.

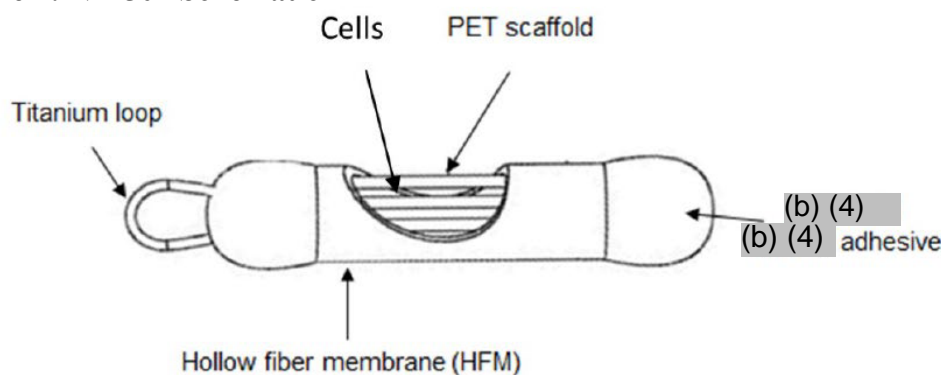
Formulation and Chemistry:

ENCELTO is a biologic-device combination product comprised of a semi-permeable capsule loaded with human ciliary neurotrophic factor (rhCNTF) secreting NTC-201-6A cells.

The ENCELTO dose of rhCNTF derives from an implant containing 200,000-440,000 allogeneic retinal pigment epithelial (RPE) cells expressing rhCNTF (NTC-201-6A cell line). NT-501 is intended for surgical implantation by ITV insertion (via a pars plana sclerotomy) providing rhCNTF in patients with idiopathic macular telangiectasia (MacTel) type 2.

ENCELTO is manufactured as sterile, non-pyrogenic, and retrievable, measuring 6.5 mm long with an external diameter of 1.3 mm. The semi-permeable capsule is the device constituent of each ENCELTO and consists of a titanium anchor loop (fixation loop, to facilitate placement and retrieval, if medically necessary), polyethylene terephthalate (PET) scaffolding within a semi-permeable hollow fiber membrane (HFM), and (b) (4) adhesive (seal). The active region membrane component of the capsule is approximately (b) (4) mm long, with an internal diameter of approximately 0.87 mm and a wall thickness of approximately (b) (4) mm. Each end of the semi-permeable capsule is sealed with methacrylate adhesive (b) (4) (Figure 1).

Figure 1: NT-501 Schematic



Composition of ENCELTO DP

Component	Quantity	Function	Compendial Status
Semi-Permeable Capsule	1 Each	Encapsulates (b) (4) cells in Endo-SFM)	Neurotech Batch Production Record (In House)
NTC-201-6A (b) (4)	(b) (4) 200,000-440,000 cells per NT-501 (b) (4)	Active - (b) (4) rhCNTF protein	Neurotech Batch Production Record (In House)
(b) (4) Adhesive	Thin layer over semi-permeable capsule opening	Seals cells within semi-permeable capsule after injection	Neurotech specification (b) (4)

Endo-SFM = endothelial-serum free medium

Reviewer note: See the Chemistry, Manufacturing, and Controls memo for details.**Abbreviations**

(b) (4)	
(b) (4)	
(b) (4)	
ELISA	Enzyme-Linked Immunosorbent Assay
ENDO	Human endothelial-SFM medium
g	gram
rhCNTF	Human ciliary neurotrophic factor
H&E	Hematoxylin and Eosin
HFM	Hollow fiber membrane
ID	Intradermal
IOP	Intraocular pressure
IP	Intraperitoneal
IS/OS	Photoreceptor inner segment/outer segment
ITV	Intravitreal
IV	Intravenous
LLOQ	Lower limit of quantification
MacTel	Idiopathic macular telangiectasia type 2
mm	millimeter
(b) (4)	
ONL	Outer nuclear layer
PCE	Polychromatic erythrocyte

PET	Polyethylene terephthalate
PK	Pharmacokinetic
rdcl	Rod-cone dysplasia Type 1
SC	Subcutaneous
SD-OCT	Spectral domain-optical coherence tomography
SFM	Serum free media
TRA	Toxicological risk assessment

Related File(s)

IND 10931- Neurotech Pharmaceuticals, Inc.; Allogeneic Retinal Pigment Epithelial Cells Transfected with DNA Plasmid Vector (b) (4), CNTF, Encapsulated in a Hollow Fiber Membrane (HFM); Treatment of idiopathic macular telangiectasia (MacTel) type 2; Status: ACTIVE

Table of Contents

INTRODUCTION	5
NONCLINICAL STUDIES.....	6
PHARMACOLOGY STUDIES.....	6
Summary List of Pharmacology Studies	6
Overview of In Vivo Pharmacology Studies	7
SAFETY PHARMACOLOGY STUDIES.....	11
PHARMACOKINETIC / DISTRIBUTION STUDIES	11
Summary List of Pharmacokinetics Studies.....	11
TOXICOLOGY STUDIES	17
Summary List of Toxicology Studies.....	17
APPLICANT’S PROPOSED LABEL.....	51
CONCLUSION OF NONCLINICAL STUDIES	51
KEY WORDS/TERMS	51

INTRODUCTION

Idiopathic macular telangiectasia type 2 (MacTel) is a bilateral neuro-vascular and glial degenerative condition, of unknown etiology, with characteristic neurosensory atrophy, perifoveal telangiectatic vessels, and diffuse hyper-fluorescence observed in the late phase of fluorescein angiography¹. Other characteristic findings include loss of retinal transparency, crystalline deposits, a decrease or absence of macular pigment, and hyperplasia of the retinal

pigment epithelium in the macular area. The natural course of MacTel is gradual bilateral macular photoreceptor loss and consequent loss of vision, occasionally accompanied by the development of neovascularization and severe vision loss¹.

There is significant evidence to support the use of rhCNTF as a potential therapy for retinal degenerative diseases^{2,3}. Histopathologic studies in naturally occurring and genetically engineered animal models of photoreceptor dysfunction that phenotypically model retinitis pigmentosa support a role for rhCNTF as a therapeutic agent for reducing photoreceptor loss associated with outer retinal degeneration. Specifically, rhCNTF is one of several neurotrophic factors that are produced endogenously by neurons and Müller cells, and rhCNTF has been shown to be effective in slowing photoreceptor neuron loss in various animal models of retinal degeneration⁴.

Although rhCNTF is a candidate for treatment of retinal neurodegenerative diseases, it is limited by its extremely short half-life when administered IV⁵. Therefore, Neurotech Pharmaceuticals has developed a biologic-device combination product, NT-501, implanted ITV to treat retinal degenerative diseases and overcome the limitations associated with IV administration of rhCNTF.

NONCLINICAL STUDIES

PHARMACOLOGY STUDIES

Summary List of Pharmacology Studies

The following pharmacology studies were conducted to support the rationale for the administration of NT-501 in the proposed clinical population with MacTel.

In Vitro Studies

No *in vitro* pharmacology studies were conducted for NT-501.

In Vivo Studies

Pharmacology studies

Study Number	Study Title / Publication Citation	Report Number
1	Effect of Encapsulated Cell-Based Delivery of CNTF in a Mutant Dog Model of Retinitis Pigmentosa	P0003-NT501-0003-INT
2	Electroretinographic (ERG) Assessments for Intra-Ocular Implantation of NT-501 Devices	P0017
3	Tao et al. (2002) Effect of encapsulated cell-based delivery of CNTF on photoreceptor degeneration. Vol.43, 3292-3298.	Publication
4	Li Y, et al. (2010) CNTF Induces Regeneration of Cone Outer Segments in a Rat Model of Retinal Degeneration. PLoS ONE 5(3): e9495.	Publication

Overview of In Vivo Pharmacology Studies

Study No. 1

Report Number		P0003-NT501-0003-INT
Date Report Signed		30-July-2003
Title		Effect of Encapsulated Cell-Based Delivery of CNTF in a Mutant Dog Model of Retinitis Pigmentosa
GLP Status		No
Testing Facility		(b) (4)
Objective(s)		<ul style="list-style-type: none"> To evaluate whether intra-ocular implantation of NT-501 (encapsulated rhCNTF secreting NTC-201 cells) would affect the normal loss of cells in the ONL in dogs with rod-cone dysplasia type 1 (<i>rcd1</i>). To collect clinical observations of the eye after device implantation.
Study Animals	Strain/Breed	<i>rcd1</i> dogs
	Species	<i>Canis familiaris</i>
	Age	7 weeks old at implantation
	Body Weight	10 kg
	#/sex/group	Sex not provided
	Total #	41 (estimated)
Test Article(s)		NT-501 units prepared at the Applicant's manufacturing facility (see Reviewer comment). Lot numbers not provided.
Control Article(s)		None
Route of Administration		IVT implantation
Description of the Disease/Injury Model and Implant Procedure		<i>Rcd1</i> dogs are a well-established animal model for retinitis pigmentosa. These dogs have progressive retinal atrophy where at 7 weeks of age they typically have only 5-7 rows in the ONL which normally decreases to 2-3 rows by 14 weeks of age.
Study Groups and Dose Levels		The left eye was implanted with NT-501 with 400,000 cells/device. The right eye served as a control (no implant).
Dosing Regimen		Single implantation
Randomization		No
Description of Masking		None
Scheduled Sacrifice Time Points		Weeks 7 and 14

➤ **Reviewer comment:**

- *Per the applicant (Section 5 of the study), “The NT-501 units that were used in this study possessed varying rhCNTF output levels. They were manufactured by research and development staff under non-GMP conditions by loading devices with different levels of a variety of NTC-201 cell lines that had inherently different rhCNTF expression levels.”*
- *Several versions of the device were used in this study (NT-501; NT-501-6A; NT-501-10; NT-501-10-E; NT-501M).*
- *The device used in this study is an early prototype that is 11 mm in length (the proposed clinical device is 6.5 mm). Thus, this study is preliminary regarding assessing the overall pharmacology of the product, NT-501.*

- *The cell lines and rhCNTF expression levels were:*
 - NTC-201-(b) (4) (aka (b) (4)) <100 ng/lx10⁶ cells/day
 - NTC-201-10: (b) (4) cells/day
 - NTC-201-6A (the cell line included in the final clinical product): (b) (4) cells/day.

Summary:

Key evaluations and assessments:

- Clinical status based on ophthalmic examination: Vitreous, lens, and cornea assessments.
- rhCNTF output: rhCNTF protein levels (ng/day) were measured prior to implant and post-explant by enzyme-linked immunosorbent assay (ELISA).
- Histopathology: Cell viability and effect of NT-501 implantation on retention of the rows of nuclei in the ONL were analyzed by Hematoxylin and Eosin (H&E) staining.

Key results:

- Clinical status based on ophthalmic examination:
 - Vitreous: Change in the refractive index and/or clarity of the vitreous; milky/filmy appearance on exam - association to NT-501 unclear. This finding did not resolve by the Week 14 time point.
 - Lens: Transient, dose-dependent appearance of one or more cystic, bubble-like features in the lens which were translucent at the Week 7 time point. These findings were resolved by the Week 14 time point.
 - Cornea: Transient vascular engorgement or reddening present at the Week 7 time point and resolved by the Week 14 time point.
- rhCNTF output: At Weeks 7 and 14, results showed correlation between pre-implant and post-explant rhCNTF output levels, with an expected decrease in rhCNTF output in post-explant samples. The post-explant rhCNTF output level was a minimum of 1.18 ng/day.
- Histopathology:
 - Histological examination of explanted devices confirmed the presence of viable cells in all cases.
 - ITV implantation of the NT-501 had a significant, dose-dependent preserving effect in the number of rows of nuclei in the ONL in the *rcd1* dog retinitis pigmentosa model compared to the ONL in the contralateral eye that did not receive the NT-501 implant.

Reviewer conclusion:

The data from this preliminary study show that ITV implantation of the NT-501 device may provide a protective effect against loss of photoreceptors; and may produce a transient, dose-dependent lens change.

Study No. 2

Report Number		P0017
Date Report Signed		Not signed
Title		Electroretinographic (ERG) Assessments for Intra-Ocular Implantation of NT-501 Devices
GLP Status		No
Testing Facility		Neurotech USA Inc. (b) (4)
Objective(s)		1. To evaluate the ERG responses to ITV implant of NT-501 devices in healthy rabbits. 2. Evaluate photoreceptors and the outer segment of the retina histologically following NT-501 device implantation.
Study Animals	Strain/Breed	(b) (4)
	Species	<i>Oryctolagus cuniculus</i>
	Age	Not reported
	Body Weight	Not reported
	#/sex/group	Sex not reported. N = 5 in Group 1 N = 11 in Groups 2-3
	Total #	16
Test Article(s)		NT-501-10 (low rhCNTF output, 400,000 cells/device) NT-501-6A (high rhCNTF output, 400,000 cells/device) Lot numbers not provided.
Control Article(s)		Empty device
Route of Administration		ITV implantation
Description of the Disease/Injury Model and Implant Procedure		Healthy (b) (4) rabbits Implant procedure: Sclerotomy The left eye received an implant, and the right eye was naive
Study Groups and Dose Levels		Group 1 – Control; empty device implanted. Group 2 – NT-501-10 device (low output) implanted. Group 3 – NT-501-6A device (high output) implanted.
Dosing Regimen		Single implantation
Randomization		No
Description of Masking		None
Scheduled Sacrifice Time Point		Day 25

Summary:

Key evaluations and assessments:

- NT-501 device performance:
 - rhCNTF output: Assessed by measuring pre-implant rhCNTF output levels (ng/day) and a post-explant rhCNTF output (ng/day) via ELISA.
 - Cell viability: Determined in explanted devices processed for histological evaluation by H&E staining.

- Ophthalmic examinations: Conducted with slit lamp and indirect ophthalmoscope.
- ERG: Recordings were performed under dark-adapted and light-adapted conditions. a- and b-wave ERG responses to different intensities of light were recorded at time 0 (prior to implantation), and at Day 5, 15, and 25 post implantation. At each time point, ERG measurements were recorded for both eyes).

Key results:

- NT-501 device performance:
 - rhCNTF output:
 - For *NTC-201-10* the pre-implant levels ranged from 23.7 through 32.0 ng/day of rhCNTF, while the post-implant levels ranged from 4.0 and 5.8 ng/day of rhCNTF.
 - For *NTC-201-6A* the pre-implant levels ranged from 118.0 and 130.3 ng/day of rhCNTF, while the post-implant levels ranged from 19.0 and 26.0 ng/day of rhCNTF.
 - Cell viability: Explanted devices had a high density of viable cells, particularly along the membrane wall. A low density of viable cells was found towards the center of the device.
- Ophthalmic examination: minor vitreous opacity, bleeding, iris neovascularization, and moderate vitreous membrane (fibrosis).
- ERG:
 - No change in ERG a-wave amplitudes for dark-adapted ERG, which represents the function of the photoreceptors.
 - No change was observed in light-adapted ERG a-wave, which represents the function of photoreceptors, for day 5 and day 15. At day 25, rabbits implanted with either high dose rhCNTF or empty devices showed a decrease in ERG a-wave amplitude, and therefore this finding was not attributed to a rhCNTF effect.
 - An increase in ERG b-wave amplitude under low light intensities (filters 6.0~4.5), which represents the input from the proximal part of retina.
 - No change in ERG b-wave responses under middle range light intensities (filters 4.0~3.0), which represents the bipolar cell activated Rod system.
 - No change in ERG b-wave responses under bright light intensities (filter 2.5~0.5), which represents the cone activities.
 - In high dose rhCNTF treated animals, at Days 15 and 25 post implantation, there was a decrease in ERG b-wave amplitude, which represents function of secondary neuron: bipolar cells and Müller cells).

Reviewer conclusions:

- *rhCNTF delivered at 5 ng/day did not affect the ERG of the healthy rabbit retina.*
- *rhCNTF delivered at 22 ng/day did not affect the rod function but decreased cone ERG b-wave by 30%, indicating the cone and the rod systems respond to rhCNTF differently.*
- *Peri-implant tissue (fibrosis) was present around the implant near its attachment site.*
- *There were no histopathology abnormalities in the implanted eyes.*

Studies Nos. 3 and 4

Reviewer note: The applicant submitted two publications, Tao, et al. (2002) Encapsulated cell-based delivery of rhCNTF reduces photoreceptor degeneration in animal models of retinitis pigmentosa. Invest Ophthalmol Vis Sci. 2002;43(10):3292-8⁴ and Li, Y, et al. (2010) rhCNTF induces regeneration of cone outer segments in a rat model of retinal degeneration (PLoS ONE 5(3): e9495)² as pharmacology studies. These publications did not use the clinical product and the resulting data are therefore not reviewed in detail this memo.; These data provide preliminary support that precursor NT-501 implants and rhCNTF secretion improve the disease phenotypes in animal models.

SAFETY PHARMACOLOGY STUDIES:

No safety pharmacology studies with NT-501 were conducted.

Study Number	Study Title / Publication Citation	Report Number
5	Effect of Daily Administration of Recombinant Human Ciliary Neurotrophic Factor on Gross Behavior and Symptomatology in Male Mice	504-M-94-92997-007-SC-SIP

Reviewer note: This safety pharmacology study for rhCNTF did not evaluate NT-501 and was therefore not reviewed in this memo.

PHARMACOKINETIC / DISTRIBUTION STUDIES**Summary List of Pharmacokinetics Studies****In Vivo Studies**

The pharmacokinetic study described in Study Report No. R491: “12-Week Ocular Toxicity and Pharmacokinetic Study in Rabbits Evaluating NT-501 Manufactured with (b) (4)

² Li, Y, Tao, W, Luo, L, et al. (2010) CNTF induces regeneration of cone outer segments in a rate model of retinal degeneration. PLoS ONE 5(3): e9495.

(b) (4) ” was conducted in tandem with Toxicology Study No. BLK20-01 “Evaluation of NT-501.6A Intraocular Implants in Rabbits.” These studies are reviewed in the Toxicology Study Section in this review memo.

Study Number	Study Title / Publication Citation	Report Number
6	Evaluation of Intraocular CNTF Dosing in (b) (4) Rabbits	R331
7	Pre-Clinical Evaluation of the NT-501 in the Rabbit Eye: CNTF Vitreous Levels and Pre-Implant and Post-Explant CNTF Device Output Rates	NT-501-0027-INT
8	Pre-Clinical Evaluation of NT-501-X.02 in the Rabbit Eye: CNTF Vitreous Levels and Pre-Implant and Post-Explant CNTF Device Output Rates.	P0068
9	Pre-Clinical Evaluation of NT-501 in the Rabbit Eye: Pharmacokinetics and Safety Profile of CNTF using (b) (4) and Scaffolding Material.	P0143
10	Evaluation of a 4 and 8 Week Preimplant Hold for 6 mm vs. 11 mm Devices in the Rabbit Eye.	P0053
11	Investigation of NT-501-6A.02 In Vitro and In Vivo Device Performance; Initial Assessment of Impact of Lot Size (b) (4) -	P-0101
12	Sub-Group Analysis for Protocol Study P01 13.00: Evaluation of (b) (4) Polyethylene Terephthalate (PET) for Use in the NT-501 Clinical Program -	P0113
13	Evaluation of performance in vitro and in vivo of NT-501-6A.02 produced with (b) (4) HFM	P0117
14	Optimization of NT-501-6A.02 Loading Parameters: Impact of Injection Volume, Cell Density and Fill Rate on Device Performance In Vivo.	R453
15	Evaluation of NT-502 Intraocular CNTF delivery and Device Anchoring Techniques in the Rabbit A Collaboration with (b) (4)	P0051
16	Comparison of NT-501 Device Performance Following Hold in Endothelial- SFM With and Without (b) (4)	P0089
17	The Effects of Temperature Shifts on Encapsulated NTC-201 Cells	P0094
18	Qualification of a (b) (4) Hollow Fiber Membrane and a Polyethylene terephthalate Alternative (PET) Scaffolding for Use in the NT-501 Clinical Program	P0095
19	Assessment of In Vivo CNTF Levels for (b) (4) Devices	P0097
20	Comparison of NT-501.02 Performance in Hold Media Supplemented with (b) (4)	P0110
21	Periocular Triamcinolone Acetonide (TA) in the Rabbit	P0052
22	Drug-Drug Interactions of Common Antibiotics on NT-501	R580-2

Study No. 6

Report Number	R331
Date Report Signed	September 17, 2019
Title	Evaluation of Intraocular CNTF dosing in (b) (4) Rabbits
GLP Status	No

Testing Facility		(b) (4) : Neurotech Pharmaceuticals 900 Highland Corporate Dr. St 101 Cumberland, RI 02864 (b) (4)																																			
Objective(s)		To evaluate PK in vitreous humor, aqueous humor, retina, and serum following ITV implant of two rhCNTF-producing devices in (b) (4) rabbits.																																			
Study Animals	Strain/Breed	(b) (4)																																			
	Species	<i>Oryctolagus cuniculus</i>																																			
	Age	Not specified																																			
	Body Weight	2-4 kgs																																			
	#/sex/group	Sex not specified. Group 1: n=6 Group 2: n=9 Group 3: n=12																																			
	Total #	27 (21 rabbits unilaterally implanted test article, 6 naïve rabbits)																																			
Test Article(s)		(b) (4) HFM + 203,000 NTC-201-6A cells/device) (b) (4) HFM 406,000 NTC-201-6A cells/device) (b) (4) <i>From: Module 4.2.2.3, Study Report No. R331, page 21</i>																																			
Control Article(s)		N/A																																			
Route of Administration		ITV																																			
Description of the Disease/Injury Model and Implant Procedure		Healthy (b) (4) rabbits Implant procedure: Sclerotomy Unilateral ITV implant in the left eye																																			
Study Groups and Dose Levels		<p>Table 1. Group Designation and Dose Levels</p> <table border="1"> <thead> <tr> <th rowspan="2">Group No.</th> <th rowspan="2">Left Eye Treatment^a</th> <th rowspan="2">Dose Level/ Left Eye</th> <th rowspan="2">Right Eye Treatment^a</th> <th rowspan="2">Total No. of Animals^a (M or F)</th> <th colspan="3">No. of Animals for Scheduled Sacrifice</th> </tr> <tr> <th>1m</th> <th>3m</th> <th>9m</th> </tr> </thead> <tbody> <tr> <td>1</td> <td>Untreated Naïve</td> <td>NA</td> <td>Untreated</td> <td>6</td> <td>2</td> <td>2</td> <td>2</td> </tr> <tr> <td>2</td> <td>(b) (4)</td> <td>1 device</td> <td>Untreated</td> <td>9</td> <td>3</td> <td>3</td> <td>3</td> </tr> <tr> <td>3</td> <td>(b) (4)</td> <td>1 device</td> <td>Untreated</td> <td>12</td> <td>4</td> <td>4</td> <td>4</td> </tr> </tbody> </table> <p>(b) (4) = 10.5 mm, F = Female; M = Male; ^a Animals will be grouped into three cohorts. Day 1 of the dosing phase will be designated as the first day of dosing. Animals will be dosed once on Day 1 of the dosing phase. <i>From: Module 4.2.2.3, Study Report No. R331, page 11</i></p>	Group No.	Left Eye Treatment ^a	Dose Level/ Left Eye	Right Eye Treatment ^a	Total No. of Animals ^a (M or F)	No. of Animals for Scheduled Sacrifice			1m	3m	9m	1	Untreated Naïve	NA	Untreated	6	2	2	2	2	(b) (4)	1 device	Untreated	9	3	3	3	3	(b) (4)	1 device	Untreated	12	4	4	4
Group No.	Left Eye Treatment ^a	Dose Level/ Left Eye						Right Eye Treatment ^a	Total No. of Animals ^a (M or F)	No. of Animals for Scheduled Sacrifice																											
			1m	3m	9m																																
1	Untreated Naïve	NA	Untreated	6	2	2	2																														
2	(b) (4)	1 device	Untreated	9	3	3	3																														
3	(b) (4)	1 device	Untreated	12	4	4	4																														
Dosing Regimen		Single implantation																																			
Randomization		No																																			
Description of Masking		Not provided																																			
Scheduled Sacrifice Time Point		Months 1, 3, and 9																																			

Summary:

Key evaluations and assessments:

- In vitro assessment of rhCNTF concentration: Determined at 2 weeks post-manufacture by (b) (4).
- Clinical observations: Conducted weekly for the duration of the study.
- Ophthalmic observations: Conducted by indirect ophthalmoscope at pre-implantation and at Months 1, 3, and 9 for all active animals. Assessments included: lens changes, anterior cell infiltrate, vitreous haze/flare, vitreous cells, retinal detachment.
- rhCNTF concentration and rhCNTF PK analysis: Measured by (b) (4) at explant (Months 1, 3, and 9) in the vitreous, aqueous, retina, and serum. Bioanalytical analysis to determine Cmax.
- Device histology: Upon explant cell 'health' and distribution were determined by (b) (4) staining.

Key results:

- In vitro assessment of rhCNTF concentration:
 - (b) (4) devices expressed 26.2 ± 5.9 rhCNTF ng/mL/day.
 - (b) (4) devices expressed 77.2 ± 12.7 ng/mL/day.
- Clinical observations: No gross clinical observations were noted for the duration of the study.
- Ophthalmic observations:
 - Vitreous haze was observed in 4/12 Group 3 animals at Month 1, 2/8 Group 3 animals at Month 3 and had resolved (0/4 Group 3 animals) at Month 9.
 - Possible retinal detachment in 1/12 Group 3 animals at Month 1.
 - No other (b) (4)-related findings were reported in any animals.
- rhCNTF concentration and rhCNTF PK analysis:
 - Group 2: Cmax rhCNTF concentration in the vitreous, aqueous, and retina samples peaked at Month 1 followed by steady-state levels for the duration of the study.
 - Group 3: Cmax rhCNTF concentrations in the vitreous occurred at Month 3 and maximized in the retina and aqueous at Month 9.
 - All serum rhCNTF concentrations were below the lower limit of quantitation (LLOQ).
- Device histology: (b) (4) devices had a highly distributed and viable cells at all time points.

Reviewer conclusions:

- (b) (4) devices were well-tolerated.
- (b) (4) consistently produced greater rhCNTF levels with the ocular samples during the study (up to 9-month post-implantation).
- rhCNTF was not detected in the serum of any animals, indicating a lack of systemic exposure and localized secretion from the device.

Study No. 7

Report Number		NT501-0027-INT
Date Report Signed		July 30, 2003
Title		Pre-Clinical Evaluation of the NT-501 in the Rabbit Eye: CNTF Vitreous Levels and Pre-Implant and Post-Explant CNTF Device Output Rates
GLP Status		No
Testing Facility		<p>(b) (4)</p> <p>NT-501 Preparation and (b) (4) Assay: Neurotech USA, Inc. (b) (4)</p> <p>Histopathology: (b) (4)</p>
Objective(s)		To evaluate the relationship between pre-implant and explant NT-501 device rhCNTF output, rhCNTF levels in vitreous, after various durations of ITV implantation.
Study Animals	Strain/Breed	(b) (4)
	Species	<i>Oryctolagus cuniculus</i>
	Age	5-6 months old (adult)
	Body Weight	3-5 kgs
	#/sex/group	Sex not specified. 9/group
	Total #	18
Test Article(s)		NT-501-10 (lower rhCNTF output, 400,000-(b) (4) NTC-201-10 cells/device) NT-501-6A (higher rhCNTF output, 400,000-(b) (4) NTC-201-6A cells/device) Lot numbers not provided.
Control Article(s)		N/A
Route of Administration		Bilateral ITV
Description of the Disease/Injury Model and Implant Procedure		Healthy (b) (4) rabbits Implant procedure: Sclerotomy
Study Groups and Dose Levels		Group 1: NTC-201-10 Group 2: NTC-201-6A
Dosing Regimen		Single implantation

Randomization	No
Description of Masking	None
Scheduled Sacrifice Time Point	Day 180/2 or 365

Summary:*Key evaluations and assessments:*

- rhCNTF output:
 - rhCNTF levels were measured by ELISA from pre-implant, throughout the study in the vitreous, and at explant. Vitreous was collected from both eyes under sedation on Days 1, 3, 7, 14, 30, and 90 for both groups. Vitreous was collected from Group 1 animals on Day 180 and from Group 2 animals on Days 135, 182, and 365.
- Ocular histology:
 - Histological evaluation was conducted on three eyes from three rabbits implanted with NT-501-6A.
 - Samples were fixed, processed, sectioned, and stained with H&E and (b) (4) staining.
- Device histology at explant.
Reviewer note: Procedure not specified.

Key results:

- rhCNTF output:
 - NT-501 devices continued to produce rhCNTF *in vivo* for the maximum implant duration—180 days for Group 1 and 365 days for Group 2.
 - Half-life for rhCNTF output was 71 days for Group 1 and 198 days for Group 2.
 - The rhCNTF concentration in the vitreous remained at a relatively constant proportion to device output measured at explant.
- Ocular histology:
 - All eyes had protein in the aqueous and vitreous chambers.
 - 2/3 eyes exhibited mild degeneration on the side near the incision.
 - 1/3 eyes had disruption of the lens capsule.
 - 2/3 eyes had retinal detachment.
 - No fibrosis was observed.
- Device histology at explant: Histological evaluation of explanted devices indicated that all devices contained healthy, viable cells.

Reviewer conclusions:

- *The vitreous rhCNTF levels and device rhCNTF output at explant are reasonably constant for up to 365 days for NT-501-6A devices.*
- *The rate of decrease of rhCNTF output is greater for NT-501-10 devices than NT-501-6A devices (71 days and 198 days, respectively).*
- *These data suggest that NT-501-6A will function in vivo for up to one year.*

Studies Nos. 8-22:

These studies are evaluations of changes made to NT-501 during the clinical development program (e.g., including sources of the HFM, PET membrane, scaffolding, etc.) incorporate versions of the product that differ from the clinical product (Study #7), or exploratory studies regarding drug interactions (Studies #21 and 22). For each change made to the product while under investigation, or drug interaction, differences in NT-501 function were evaluated clinically. Therefore, these studies are not reviewed in this memo.

TOXICOLOGY STUDIES

Summary List of Toxicology Studies

The following toxicology studies were conducted to evaluate the safety of revakinagene tarorelcel (NT-501).

Toxicology Studies:

Study Number	Study Title / Publication Citation	Report Number
23	12-Week Ocular Toxicity and Pharmacokinetic Study in Rabbits Evaluating NT-501 Manufactured with (b) (4) and Evaluation of NT-501.6A Intraocular Implants in Rabbits	R491, inclusive of BLK20-01
24	Toxicology Assessments for Intraocular Implantation of NT-501 in Pigs	P0008
25	Toxicology Assessments for Intraocular Implantation of Encapsulated NTC-200 Cells	P0042
26	Exploratory Assessment of Toxicity of Intraocular Implantation of NT-501 Devices in Mini-Pigs	P0007
27	The Effect of Reimplanting the NT-501 Device on Wound Healing in Rabbit Eyes	P0093

Study No. 23

Report Number	R491, inclusive of BLK20-01
Date Report Signed	R491: March 14, 2021; BLK20-01: March 9, 2021
Title	12-Week Ocular Toxicity and Pharmacokinetic Study in Rabbits Evaluating NT-501 Manufactured with (b) (4) and Evaluation of NT-501.6A Intraocular Implants in Rabbits
GLP Status	No

Testing Facility		Bioanalytical Test Site and Analysis Information: Neurotech Pharmaceuticals Inc. 900 Highland Corporate Drive Building 1, Suite 101 Cumberland, RI 02864 (b) (4)		
Objective(s)		To evaluate the stability of NT-501 devices containing NTC-201-6A cells engineered to produce rhCNTF, manufactured with the following investigational membranes: <ul style="list-style-type: none"> • (b) (4) membrane used in the final clinical product.		
Study Animals	Strain/Breed	(b) (4)		
	Species	<i>Oryctolagus cuniculus</i>		
	Age	Not provided		
	Body Weight	Not provided		
	#/sex/group	5-8 males/group		
	Total #	35 males		
Test Article(s)		NT-501.6A (Lot# DEV-06OCT2020, 203,000 NTC-201-6A cells/device)		
Control Article(s)		N/A		
Route of Administration		ITV implantation		
Description of the Disease/Injury Model and Implant Procedure		Healthy (b) (4) rabbits Implant procedure: Sclerotomy Unilateral implant to the left eye		
Study Groups and Dose Levels		Group No.	No. of An.	Test Article
		1 ⁺	6	Control
		2 ⁺	8	NT-501_(b) (4)
		3 ⁺	8	NT-501_(b) (4)
		4 ⁺	8	NT501_(b) (4)
		5 ⁺	5	NT-501_(b) (4)
		Dose Regimen ROA NA Unilateral intraocular surgical implantation (Sponsor provided surgeon)		
		From: Module 4.2.2.3, Study Report R491, page 11		
Dosing Regimen		Single implantation		
Randomization		Yes		
Description of Masking		Not provided		
Scheduled Sacrifice Time Points		Weeks 4 and 12		

Summary:*Key evaluations and assessments:*

- Clinical observations: Conducted at Day -1 and at least once daily for the duration of the study. Special attention was paid to the eyes for signs of ocular pain, discharge, redness, abnormal pupil dilation/constriction as compared to the contralateral eye.

- Surgical site observation: Conducted at Days 1, 2, 3, and 7 post-surgery.
- Body weights: Recorded prior to implantation and once weekly post-implantation.
- Ophthalmic examinations: Conducted on Day -3 and Weeks 4 and 12 post-implantation. These included gross examination (hyperemia, discharge, squinting, lid swelling), and dilated exams using a slit-lamp and an indirect ophthalmoscope.
- IOP: Measured at Day -3 and Weeks 4 and 12 post-implantation.
- Samples were taken from serum, vitreous and aqueous humor, and retinal tissue and evaluated for:
 - rhCNTF levels: Determined by a (b) (4) using (b) (4) human (b) (4) cell line which proliferates in a dose-dependent manner in response to rhCNTF.
- Cell Viability:
 - (b) (4) Assay.
Reviewers note: This assay uses (b) (4) and allows quantitative evaluation of cell viability.
 - Cell number based on DNA recovery and measurement.
 - Histopathology evaluation of device using sections from the device and H&E staining.
- In vitro analyses were conducted on devices at (b) (4) days post-manufacture and following a (b) (4):
 - rhCNTF expression levels
 - Cell viability determined as described above.

Key results:

- Clinical observations: Instances of mild squinting or redness were reported in 3 animals. These animals were closely monitored, and no additional interventions were required.
- Surgical site observation: Hyperemia and chemosis were reported in all groups at Days 1, 2, and 3, but resolved by Day 7.
- Body weights: There were no significant changes in body weights throughout the study.
- Ophthalmic examinations:

- There were no significant findings throughout the study. Occasional instances of conjunctival and/or iridial redness were reported at the Week 12 time point in all groups.
- There were fibrin deposits in the anterior lens reported sporadically at Weeks 4 and 12 indicating an inflammatory response.
- IOP: There were no significant changes comparing the eye that received NT-501 to the contralateral eye throughout the study.
- rhCNTF levels: There were no differences in rhCNTF levels between Groups 2-5 in the vitreous humor, aqueous humor, and retina at each time point. Serum samples were below the limit of detection for rhCNTF.
- Cell viability: Explanted NT-501 devices from all groups showed a decrease in viability between 1 and 3 months based on (b) (4) Assay, cell DNA recovery and measurement, and explanted device histology.

Reviewer conclusions:

- (b) (4) rabbits tolerated the various compositions of the NT-501 implant well.
- rhCNTF levels in the vitreous humor, aqueous humor, and retinal samples were consistent for both membranes evaluated.
- rhCNTF was below the limit of detection in the serum for all samples.

Study No. 24

Report Number	P0008 – Parts A, B, and C
Date Report Signed	July 14, 2013
Title	Toxicology Assessments for Intraocular Implantation of NT-501 in Pigs
GLP Status	No

Testing Facility		<p>Animal Care & Surgery: (b) (4)</p> <p>NT-501 Preparation and (b) (4) Assay: Neurotech USA, Inc. (b) (4)</p> <p>Necropsy conducted at (b) (4)</p> <p>Histopathology: (b) (4)</p> <p>Blood Chemistry: (b) (4)</p>
Objective(s)		<p>Parts A and C: To evaluate the effect of ITV implantation of NT-501-10 (low output) and NT-501-6A (high output) devices secreting rhCNTF on the eye of (b) (4) mini-pigs and mature (b) (4) pigs.</p> <p>Part B: To investigate the effect of potential catastrophic failure of the NT-501 device by injecting NT-201-6A cells directly into the eye of (b) (4) mini-pigs.</p>
Study Animals	Strain/Breed	(b) (4) mini-pigs and (b) (4) pigs
	Species	<i>Sus scrofa domesticus</i>
	Age	<p>Part A: (b) (4) mini-pigs - 4 to 8 months old</p> <p>Part B: (b) (4) mini-pigs - 4 to 8 months old</p> <p>Part C: (b) (4) pigs - mature 4 to 10 years old</p>
	Body Weight	<p>(b) (4) mini-pigs 10-12 kg</p> <p>(b) (4) pigs - 61-95 kg</p>
	#/sex/group	<p>Part A: (b) (4) mini-pigs 16 female, 16 male</p> <p>Part B: (b) (4) mini-pigs 6 female, 6 male</p> <p>Part C: (b) (4) pigs - 14 female</p>
	Total #	<p>(b) (4) Mini-Pigs - 44</p> <p>(b) (4) pigs - 14</p>
Test Article(s)		<p>Parts A and C: NT-501-10 (Lot No. 100401, low rhCNTF output, 400,000 NTC-201-10 cells/device) and NT-501-6A (Lot No. 091901, high rhCNTF output, 400,000 NTC-201-6A cells/device)</p> <p>Part B: NT-201-6A cells</p>
Control Article(s)		<p>Part A: Empty device (Lot No. 103001)</p> <p>Part B: (b) (4)</p> <p>Part C: None</p>
Route of Administration		ITV implantation to the left eye
Description of the Disease/Injury Model and Implant Procedure		<p>Healthy pigs</p> <p>Implant procedure: Sclerotomy</p>

Study Groups and Dose Levels

Part A:

Group #	Number of pigs/Group	Treatment	NT-501 Device Lot #	Duration of Observation [Animal No. (Sex)]	
				3 Months	6 Months
1	6	No treatment	N/A	84329 (M) 84291 (F) 84324 (F)	84272 (M) 84367 (M) 84401 (F)
2	10	NT-501-6A (high output device)	091901	84319 (M) 84374 (M) 84377 (M) 84297 (F) 84308 (F)	84341 (M) 84398 (M) 84286 (M) 84305 (F) 84315 (F)
3	10	NT-501-10 (low output device)	100401	84437 (M) 84516 (M) 84309 (F) 84487 (F) 84511 (F)	84429 (M) 84438 (M) 84317 (F) 84443 (F) 84518 (F)
4	6	Empty device (contains no NTC-201 cells)	103001	84337 (M) 84473 (F) 84486 (F)	84322 (M) 84440 (M) 84499 (M)

From: Module 4.2.3.1, Study Report P0008, page 15

Part B:

Group #	Pigs No./Group	Treatment	Duration of Observation [Animal No. (Sex)]		
			1 Week	4 Weeks	12 Weeks
5	3	No treatment	84525 (F)	84445 (M)	84498 (F)
6	9	Left Eye: NT-201-6A cells Right eye: Saline ^a	84378 (M) 84528 (M) 84451 (F)	84332 (F) 84495 (F) 84530 (M)	84464 (M) 84475 (M) 84338 (F)

a: Animal. No. 84451 inadvertently received cell injection in the right eye and saline in the left eye.

From: Module 4.2.3.1, Study Report P0008, page 43

Part C:

Group #	Number of pigs/Group	Treatment	NT-501 Device Lot #*	Animal No.
7	5	NT-501-6A (High Output Device)	021202-01	2627 2710 2665 2747 2705
8	5	NT-501-10 (Low Output Device)	021202-02	1796 2462 2505 2596 2669
9	4	No treatment (control group)	N/A	2714 2728 2759 2908

From: Module 4.2.3.1, Study Report P0008, page 65

Dosing Regimen

Single

Randomization

No

Description of Masking

N/A

Scheduled Sacrifice Time Points

Part A: Months 3 and 6

Part B: Weeks 1, 4, and 12

Part C: Month 3

➤ **Reviewer Comment:**

This study consisted of three experiments (Parts A, B, and C) entitled:

- *Part A (pages 6-34): The Effect on the Eye of 3-Month and 6-Month Implantation of NT-501-10 and NT-501-6A Devices in 4 to 8 Months Old (b) (4) Mini-Pigs.*
- *Part B (pages 35-54): The Effect on the Eye of Intra-ocular Injection of Un-encapsulated NTC-201-6A Cells at 1 Week, 4 Weeks, and 12 Weeks After Injection in 8 to 9 Months Old (b) (4) Mini-Pigs.*
- *Part C (pages 55-80): Effect on the Eye of 3-Month Implantation of NT-501-10 and NT-501-6A Devices in Mature (b) (4) Pigs.*

Summary for Part A, B, and C:

Key evaluations and assessments for Parts A, B, and C:

- Clinical examinations: Animals were examined at pre-implantation and Weeks 2, 4, 8, 12, 16, 20, and prior to sacrifice for general condition, body weight, temperature, oral stomatitis, gross motor deficiencies, and cough.
- Ophthalmic examinations: Animals were examined at pre-implantation and Weeks 2, 4, 8, 12, 16, 20, and prior to sacrifice for pupillary response, conjunctiva, anterior chamber, device-associated complications, IOP, cornea, retina, device location, lens, and vitreous.
- Blood was collected at the time of implantation and at Weeks 2, 4, 8, 12, and prior to sacrifice for the following evaluations:
 - Clinical pathology: Hematology and serum chemistries.
 - rhCNTF levels: Measured by (b) (4) in serum samples.
 - Antibody analysis: Anti-CNTF antibodies and anti-NTC-201 cell antibodies from serum were measured by ELISA.
- Ocular histopathology: Eyes were enucleated, fixed, mounted, and sections stained with H&E for histopathology evaluation.
- Gross pathology: Complete gross pathology was conducted at the time of necropsy.
- Non-ocular histopathology: The following organs were collected, fixed, mounted, and sections stained with H&E for histopathology evaluations: adrenals, brain, heart, kidneys, liver, lungs, peri-aortic and peri-ocular lymph nodes, ovaries/testes, pancreas, pituitary gland, spleen, stomach, thymus, and any gross lesions.
- Explanted device rhCNTF output: rhCNTF levels were measured *in vitro* in ng/day.
Reviewer note: *The method for quantification of rhCNTF levels was not described.*

Key results for Part A:

- Clinical examinations: No NT-501-related effects were reported.
- Ophthalmic examinations:
 - Cystic lens changes were seen in all groups that received a device with or without cells. Incidence increased with time and with the presence of cells, but no dose-response was observed between the rhCNTF low output and high output cell lines.
 - Group 4 (empty device) animals: 2/6 at Month 1, 1/6 at Month 2, 1/6 at Month 3, and 1/3 at Month 6.
 - Group 3 (NT-501-10) animals: 2/10 at Month 1, 6/10 at Month 2, 7/10 at Month 3 and 5/5 at Month 6.
 - Group 2 (NT-501-6A) animals: 2/10 at Month 1, 6/10 at Month 2, 8/10 at Month 3, and 5/5 at Month 6.
 - Vitreous refractive changes were seen in all groups that received rhCNTF-producing cells. There was no clear dose response between the rhCNTF low output and high output cell lines.
 - Group 3 (NT-501-10) animals: 3/10 at Month 2, 6/10 at Month 3, 4/5 at Month 6.
 - Group 2 (NT-501-6A) animals: 5/10 at Month 2, 9/10 at Month 3, and 5/5 at Month 6.
- Clinical pathology: No NT-501-related effects were reported on hematology or serum chemistries.
- rhCNTF levels: All post-implantation levels of serum rhCNTF were below the LLOQ.
- Antibody analyses:
 - Anti-CNTF antibodies: Present in serum samples from all animals at pre-implantation ranging from 25-625 with no consistent increases in anti-CNTF antibodies observed.
 - Anti-NTC-201 cell antibodies: Present in serum samples from all animals at pre-implantation ranging from 5-25 with no consistent increases in anti-NTC-201 cell antibodies observed.
- Gross pathology: No significant gross pathology findings were reported.
- Ocular histopathology:
 - Changes at Month 3:
 - Bladder cells (swollen posterior epithelial cells) and posterior migration of the epithelium were seen on the lens of all animals in Groups 2 and 3.
 - Minimal to moderate posterior lens degeneration was observed in 5/5 animals in Group 3 and 3/5 animals in Group 2.

- Minimal inflammatory cells were present in the vitreous in all animals in Groups 2 and 3.
- Changes at Month 6
 - Minimal to moderate lens changes including the presence of bladder cells, posterior degeneration, and posterior migration of epithelium were observed in all animals in Groups 2 and 3.
 - A higher mean severity of changes was present in Group 2 compared to Group 3, suggesting a rhCNTF-related dose response.
 - Fibrosis associated with the posterior capsule of the lens occurred in 4/5 animals in Group 2.
 - Minimal inflammatory cells in the vitreous was present in 4/5 animals in Group 2 and 5/5 animals in Group 3.
 - Minimal inflammation at the corneoscleral junction was seen in 1/5 animals in Group 3.
 - In Group 2, minimal inflammation in the stroma of the cornea (3/5), minimal neovascularization of the cornea (1/5), minimal to mild inflammation at the corneoscleral junction (5/5), minimal inflammatory cells in the aqueous chamber (2/5), minimal protein in the aqueous chamber (3/5), and synechiae of the iris/ciliary body (3/5) were also observed.
- Non-ocular histopathology: No significant histopathology findings of major organs were reported.
- Explanted device rhCNTF output: 15/20 devices demonstrated rhCNTF levels > 0.5 ng/day at the time of explantation.

Key results for Part B:

- Clinical examinations: No NT-501-related effects were reported.
- Ophthalmic examinations:
 - Minimal lens changes were observed in eyes administered NTC-201-6A cells at Weeks 2 (1/16), 8 (3/3) and 12 (3/3).
 - No other NTC-201-6A cell-related changes were observed.
- Clinical pathology: No NT-501-related effects were reported on hematology or serum chemistries.
- rhCNTF levels: All post-injection levels of serum rhCNTF were below the LLOQ.
- Antibody analyses:
 - Anti-CNTF antibodies: No consistent increases in anti-CNTF antibodies in serum were reported.

- Anti-NTC-201 cell antibodies: No consistent increases in anti-NTC-201 cell antibodies in serum were reported.
- Gross pathology: No significant gross pathology findings were reported.
- Ocular histopathology:
 - Lens changes in Group 6 (NTC-201-6A cells) included minimal posterior epithelial migration at Weeks 4 and 12, the presence of bladder cells and posterior degeneration at Week 12.
 - Vitreous changes in Group 6 were inflammatory cells at Weeks 1 and 4.
 - Inflammation in animals Group 6 was observed around central retinal blood vessels at Week 4.
- Non-ocular histopathology: No significant histopathology findings of major organs were reported.

Key results for Part C:

- Clinical examinations:
 - No NT-501-related effects were reported.
 - One animal in Group 7 (An. No. 2705, NT-501-6A implant) died after examination at the Week 2 time point because of respiratory obstruction related to self-inflicted trauma to the tongue during sedation.

Reviewer note: *The study report states this is a Group 7 animal. For ophthalmic examinations and ocular histopathology, some parameters indicate 4 surviving animals in Group 7 (e.g., lens changes), some indicate 4 surviving animals in Group 8 (e.g., vitreous changes), and some parameters indicate 5 surviving animals in Groups 7 and 8 (e.g., lens and vitreous histology). This reviewer included the numbers given in the study report.*
- Ophthalmic examinations:
 - Lens changes were observed in Groups 7 (NT-501-6A) and 8 (NT-501-10). There was no dose response observed.
 - Vitreous changes were observed in Groups 7 and 8. There was no dose response observed.
- Clinical pathology: No NT-501-related effects were reported on hematology or serum chemistries.
- rhCNTF levels: All post-implantation levels of serum rhCNTF were below the LLOQ.
- Antibody analyses:
 - Anti-CNTF antibody analysis: No consistent increases in anti-CNTF antibodies in serum were reported.

- Anti-NTC-201 cell antibodies: No consistent increases in anti-NTC-201 cell antibodies in serum were reported.
- Gross pathology: No significant gross pathology findings were reported.
- Ocular histopathology:
 - Lens changes were observed in 4/5 animals in Groups 7 and 8 at Week 12, including minimal or mild bladder cells, posterior degeneration, and posterior migration of epithelium. There was no dose-response observed.
 - Minimal to mild fibrosis of the lens was present in 1/5 animals in Group 7 and 2/5 Group 8 animals.
 - Minimal to mild inflammatory cells were present in the vitreous in 4/5 animals in Groups 7 and 8. 1/5 and 2/5 eyes in each group, respectively, had associated protein effusion.
 - Minimal inflammatory cells, protein, and adhesions were present in the corneo-scleral junction, aqueous chamber, and iris/ciliary body in 1/5 Group 7 animals.
 - Minimal iris inflammation was present in 1/5 Group 7 animals.
 - Proteinaceous fluid and inflammation of central vessels was present in the retina of 1/5 Group 8 animals.
 - The retina was adhered to the lens in 1/5 Group 7 animals.
- Non-ocular histopathology: No significant histopathology findings of major organs were reported.

Reviewer conclusions:

- *Implantation of NT-501-10 and NT-501-6A devices into the eye were associated with the occurrence of minimal to moderate ophthalmologic and histologic changes in the lens of the eye, and inflammation and fibrosis in the tissues surrounding the implanted device.*
- *The lens changes were slightly more severe in pigs implanted with the NT-501-6A (high rhCNTF output) devices, and the inflammation changes in the tissues surrounding the implant occurred mostly in animals received NT-501-6A, suggesting a rhCNTF dose-related effect.*
- *There was no evidence of 1) rhCNTF in the serum, 2) serum antibodies to rhCNTF or NTC-201 cells, nor 3) non-ocular toxicities following implantation of NT-501 or ITV administration of NT-201-6A cells.*

Study No. 25

Report Number	P0042
Date Report Signed	June 12, 2003
Title	Toxicology Assessment of Intraocular Implantation of Encapsulated NTC-200 Cells
GLP Status	No

Testing Facility		Animal Care & Surgery: (b) (4) NT-501 Preparation and (b) (4) Assay: Neurotech USA, Inc. (b) (4) Histopathology: (b) (4) Antibody Analysis of Serum: Neurotech USA, Inc. (b) (4)	
Objective(s)		To assess the ocular safety and tolerability of non-rhCNTF secreting, encapsulated parent cells (NTC-200 cells) in devices compared to NT-501-6A (high rhCNTF output) and NT-501-10 (low rhCNTF output) devices.	
Study Animals	Strain/Breed	(b) (4) mini pig	
	Species	<i>Sus scrofa domesticus</i>	
	Age	4-8 months old	
	Body Weight	9-18 kgs	
	#/sex/group	9 females/NT-501-200 group 6 females/NT-501-6A group 1 female/NT-501-10 group	
	Total #	16 females	
Test Article(s)		NT-501 (Lot Nos. DL043002-01, DL052102-01, 400,000 NTC-201-6A cells/device) NT-501 (Lot Nos. not provided, 400,000 NTC-201-10 cells/device)	
Control Article(s)		NT-501 (Lot Nos. DL043002-02, DL052102-02, 400,000 NTC-200 cells/device)	
Route of Administration		ITV implantation	
Description of the Disease/Injury Model and Implant Procedure		Healthy mini pigs Implant procedure: Sclerotomy Unilateral for NT-501-200 and NT-501-6A groups Bilateral for NT-501-10.	
Study Groups and Dose Levels		Test Article	(b) (4) Mini-Swine F 4 - 8 Mos. Age
		NT-501-200	3 Months / N = 9
		NT-501-6A	3 Months / N = 6
		NT-501-10	3 Months / N = 2 (bilateral)
Dosing Regimen		Single implantation	
Randomization		Yes	
Description of Masking		Not provided	
Scheduled Sacrifice Time Points		Weeks 12, 14, and 26	

Summary:

Key evaluations and assessments:

- Clinical observations: Conducted daily for 7 days post-implantation.
- Clinical examinations: Animals were examined at Weeks 4, 8, and 12 post-implantation for general condition, body weight, temperature, oral stomatitis, gross motor deficiencies, and cough.
- Ophthalmic examination: All surviving animals were examined at Weeks 5/6, 12, 14 (NT-501-10 animal only), 16, 20, and 26 weeks for pupillary response, IOP, device location, conjunctiva, cornea, lens, anterior chamber, retina, vitreous, device-associated complications, and limbus diameter.
- Antibody analyses: Blood was collected at the time of implantation and Week 12 for assessment in serum.
 - Anti-CNTF antibodies
 - Anti-NTC-201 cell antibodies

Reviewer note: *The study report did not specify the methods for antibody detection.*
- Ocular histopathology: Eyes were enucleated, fixed, mounted, and sections stained with H&E for histopathology evaluation.

Key results:

- Clinical observations: No NT-501-related effects were reported.
- Clinical examinations: No NT-501-related effects were reported.
- Ophthalmic examination:
 - Minimal lens changes occurred in:
 - 1/9 Group 1 animals at Weeks 5/6, 16, 20, and 26, 2/9 animals administered NT-501-200 at Week 12.
 - 1/6 animals administered NT-501-201-6A at Week 5/6 and in 5/6 animals at Weeks 12-26.
 - 2/2 eyes (1 animal) administered NT-501-201-10 at Weeks 8 and 14.
 - Focal vitreous cloudiness occurred in:
 - 2/6 animals administered NT-501-6A at Week 5/6 and 3/6 animals at Week 12. This finding resolved by Week 16.
 - 1/9 animals administered NT-501-200 at Week 5/6, 2/9 animals at Week 12, and 1/9 animals at Week 16. This finding resolved by Week 20.
 - Tissue formation was observed around the device in:
 - 1/6 animals administered NT-501-6A at Week 5/6 and 6/6 animals at Weeks 12, and in all surviving animals at Weeks 16, 20, and 26.
 - 5/9 animals administered NT-501-200 at Week 5/6 and 5/9 animals at Week 12.

- Antibody analyses:
 - Anti-CNTF antibodies: No consistent increases in anti-CNTF antibodies in serum were reported.
 - Anti-NTC-201 cell antibodies: No consistent increases in anti-CNTF antibodies in serum were reported.
- Ocular histopathology:
 - Findings in all groups at Week 12 included:
 - Lens changes including posterior epithelial migration with formation of bladder cells.
 - Choroid/scleral inflammation
 - Fibrous peri-implant tissue in animals where the anchor loop was not secured with in the sclera and the sclera wound healed poorly.
 - Minimal to moderate vitreous inflammation with increased protein
 - Retinal detachment in 1/9 animals administered NT-501-200.
 - There was no difference in incidence or severity between groups, suggesting that these findings are not related to the expression of rhCNTF.

Reviewer conclusions:

- *Implantation of NT-501 can lead to peri-implant fibrous tissue formation. This is not affected by rhCNTF expression.*
- *Anchor loop positioning affects the formation of peri-implant fibrous tissue. Careful attention to securing the anchor loop well within the sclera and tight closure of the implant wound minimizes or eliminates tissue formation.*
- *Lens changes were more consistently observed in groups with rhCNTF-expressing cells at later time points, suggesting that these changes may be related to rhCNTF expression.*

Studies Nos. 26 and 27

Reviewer note: *These studies did not evaluate a product representative of the intended clinical version of NT-501. Devices with NTC-201-6A cells were not evaluated. Therefore, these studies are not reviewed in this memo.*

Developmental and Reproductive Toxicology Studies¹:

Study Number	Study Title	Report Number
28	Subcutaneous male fertility and reproduction study in rats with recombinant human ciliary neurotrophic factor (RS-92997-007)	44-R-94-92997-007-SC-MR
29	Subcutaneous female fertility and reproduction study in rats with recombinant human ciliary neurotrophic factor (RS-92997-007)	45-R-94-92997-007-SC-RP

Study Number	Study Title	Report Number
30	Subcutaneous teratology study in rats with recombinant human ciliary neurotrophic factor	46-R-94-92997-007-SC-TT
31	Subcutaneous teratology study in rabbits with recombinant human ciliary neurotrophic factor	47-B-94-92997-007-SC-TT
32	DART Exemption Evaluation of NT-501	Neu-10

➤ **Reviewer Comment:**

- *It is unclear if the rhCNTF tested in Studies 28-31 is representative of the rhCNTF expressed by the cells in NT-501.*
- *The dose levels of rhCNTF administered and route of administration were not representative of the proposed clinical dose level and route of administration of NT-501.*

Study No. 28

Report Number		44-R-94-92997-007-SC-MR
Date Report Signed		December 7, 1994
Title		Subcutaneous male fertility and reproduction study in rats with recombinant human ciliary neurotrophic factor (RS-92997-007)
GLP Status		No
Testing Facility		(b) (4)
Objective(s)		To evaluate the possible effects of subcutaneously administered rhCNTF on fertility and reproductive performance of the male rat.
Study Animals	Strain/Breed	(b) (4)
	Species	<i>Rattus norvegicus</i>
	Age	Males: 2 months old Females: 3.5 months old
	Body Weight	Not provided
	#/sex/group	20 M/group 40 F/group
Total #		240
Test Article(s)		rhCNTF (Lot No. 01093-06DE)
Control Article(s)		Vehicle (b) (4)
Route of Administration		SC
Description of the Disease		Healthy rats

Study Groups and Dose Levels	Group	Animal Number Series	Concentration of rhCNTF (µg/ml)	Dose Volume (ml/kg)	Dose of rhCNTF (µg/kg/day)
	1	100	(b) (4)	1	0
	2	200		1	10
	3	300		1	100
	4	400		0.3	300
<i>From: Module 4.2.3.5.1, Study Report 44-r94-92997-007-SC-MR, page 5</i>					
Dosing Regimen	Daily (males only)				
Randomization	Yes				
Description of Masking	Not provided				
Scheduled Sacrifice Time Points	Midgestation evaluation: Half of the pregnant females were sacrificed on gestational Day 14 ± 1. Litter evaluation: Half of the pregnant females were allowed to litter and sacrificed following weaning. Males were sacrificed following weaning of the pups.				

Summary:*Mating design:*

- After 62 days of SC rhCNTF administration to male rats, mating groups of 1 male and 2 naïve female rats were established.
- Females were examined daily. Day 1 of pregnancy was defined as the day there was presence of copulatory plug or sperm in vaginal lavage.
- Cohabitation continued until at 32 females in each Study Group had evidence of mating.
- At the end of the mating period, the remaining cohabitation groups were separated, and each rat housed individually.

Key evaluations and assessments:

- Clinical observations/mortality: All animals were observed daily for general condition. Clinical observations were recorded weekly, at sacrifice, and when a change occurred.
- Body weights: Recorded weekly and at termination. For females with evidence of mating, body weights were recorded on Day 1 of pregnancy, weekly thereafter, and at sacrifice.
- Mating and fertility performance of males: Males were evaluated based on mating with at least 1 female, at least 1 female pregnant, males with at least 1 female pregnant among those with evidence of mating.
- Limited necropsy of females sacrificed on gestational Day 14: The reproductive tract, including the uterus, corpora lutea, live fetuses, resorptions, and mammary chain were grossly evaluated.

- Gestation/neonatal data: Gestation period, number of pups born alive, gestation index, or pup survival indices on postpartum Days 4, 7, 14, and 21 were recorded.

Reviewer notes:

- *A gestation index is the number of litters with live pups divided by the number of pregnancies, multiplied by 100.*
 - *A pup survival index is the number of pups that survived to different life stages divided by the number of pups born live, multiplied by 100.*
- Clinical observations of pups: Recorded on Days 4, 7, 14, and 21 postpartum, including general condition, group body weight of pups/sex/litter, number of pups/sex/litter, and number of live pups/litter.

Key results:

- Clinical observations/mortality:
 - All male rats survived until scheduled termination of the study.
 - Some male rats in Groups 3 and 4 (100 and 300 µg/kg/day, respectively) were inactive and had labored respiration during Weeks 1 and/or 2.
 - One female rat, which littered but had no surviving pups, and was euthanized and removed from the study.
 - Female rats were clinically normal except for incidental conditions.
- Body weights:
 - Males in Groups 3 and 4 gained less weight than the vehicle controls during the first 3 weeks of treatment.
 - Males in Group 4 continued to gain less weight through week 10.
 - There were no differences in body weight gain of pregnant females.
- Mating and fertility performance: There were no statistically significant differences across groups.
- Limited necropsy of females sacrificed on gestational Day 14: The number of corpora lutea, implantations, live litter size, and total resorptions were similar among all groups.
- Gestational/neonatal data: There were no differences in gestation period, number of pups born alive, gestation index, or pup survival indices across groups.
- Clinical observations of pups:
 - All pups were clinically normal until weaning.
 - There were no differences in body weights of pups.
 - There were no differences across groups of pups with unscheduled deaths.

Reviewer conclusions:

- *There were no adverse effects on mating performance or fertility of male rats administered rhCNTF SC.*

- *There were no adverse effects on the gestation of pregnant female rats mated with male rats administered rhCNTF SC.*
- *There were no adverse effects on the postnatal development of the offspring from male rats administered rhCNTF SC.*

Study No. 29

Report Number	45-R-94-92997-007-SC-RP																									
Date Report Signed	May 3, 1995																									
Title	Subcutaneous female fertility and reproduction study in rats with recombinant human ciliary neurotrophic factor (RS-92997-007)																									
GLP Status	No																									
Testing Facility	(b) (4)																									
Objective(s)	To evaluate the effects of SC administration of rhCNTF on fertility and reproduction in female rats.																									
Study Animals	Strain/Breed (b) (4)																									
	Species <i>Rattus norvegicus</i>																									
	Age Female: 16 weeks old Male: 9 months old																									
	Body Weight Not specified																									
	#/sex/group 20 M/sex/group 40 F/sex/group																									
	Total # 240																									
Test Article(s)	rhCNTF (Lot No. (b) (4))																									
Control Article(s)	Vehicle (b) (4)																									
Route of Administration	SC																									
Description of the Disease	Healthy rats																									
Study Groups and Dose Levels	<table border="1"> <thead> <tr> <th>Group</th><th>Animal Number Series</th><th>Concentration of rhCNTF (µg/ml)</th><th>Dose Volume (ml/kg)</th><th>Dose of rhCNTF (µg/kg/day)</th></tr> </thead> <tbody> <tr> <td>1</td><td>100</td><td>(b) (4)</td><td>1</td><td>0</td></tr> <tr> <td>2</td><td>200</td><td>(b) (4)</td><td>1</td><td>10</td></tr> <tr> <td>3</td><td>300</td><td>(b) (4)</td><td>1</td><td>100</td></tr> <tr> <td>4</td><td>400</td><td>(b) (4)</td><td>0.3</td><td>300</td></tr> </tbody> </table> <p><i>From: Module 4.2.3.5.1, Study Report 45-r-94-92997-007-SC-RP, page 5</i></p>	Group	Animal Number Series	Concentration of rhCNTF (µg/ml)	Dose Volume (ml/kg)	Dose of rhCNTF (µg/kg/day)	1	100	(b) (4)	1	0	2	200	(b) (4)	1	10	3	300	(b) (4)	1	100	4	400	(b) (4)	0.3	300
Group	Animal Number Series	Concentration of rhCNTF (µg/ml)	Dose Volume (ml/kg)	Dose of rhCNTF (µg/kg/day)																						
1	100	(b) (4)	1	0																						
2	200	(b) (4)	1	10																						
3	300	(b) (4)	1	100																						
4	400	(b) (4)	0.3	300																						
Dosing Regimen	Daily (females only)																									
Randomization	Yes																									
Description of Masking	Not provided																									
Scheduled Sacrifice Time Points	Midgestation evaluation: Half of the pregnant females were sacrificed halfway through gestation (Day not specified). Litter evaluation: Half of the pregnant females were allowed to litter and sacrificed following weaning. Males were sacrificed following weaning of the pups.																									

Summary:*Mating design:*

- Female rats were administered rhCNTF once daily beginning at Week -2 and continuing until either midgestation necropsy or postpartum Day 21.
- Females were housed with a male rat until at least 26 females in each group had evidence of mating.

Key evaluations and assessments:

- Clinical observations/mortality: Recorded weekly.
- Body weights: Recorded weekly.
- Mating and fertility performance of females: Females were evaluated based on the proportion of females that mated compared to the vehicle control group, and the proportion of females with evidence of mating that became pregnant.
- Limited necropsy of females sacrificed on gestational Day 14: The reproductive tract, including the uterus, corpora lutea, live fetuses, and resorptions were grossly evaluated.
- Gestational/neonatal data: Gestation period, number of pups born alive, gestation index, or pup survival indices on postpartum Days 4, 7, 14, and 21 were recorded.
- Clinical observations of pups:
 - Pups were evaluated for clinical condition, body weight, and a battery of physical development and behavior parameters including pinna detachment, incisor eruption, eye opening, surface righting reflex, negative geotaxis, auditory startle response, pupillary reflex, and fur development.
 - At weaning, 1 male and 1 female pup from each litter were randomly selected for assessment of testes descent and vaginal opening.

Key results:

- Clinical observations/mortality:
 - No rhCNTF-related clinical changes were reported in females during pre- and post-mating periods.
 - All females survived to scheduled termination.
- Body weights:
 - During the pre-mating interval, Group 3 animals had decreased body weight gain and Group 4 animals had body weight loss.
 - Post-mating, body weight gain was similar in all groups.

- Mating and fertility performance:
 - The mating performance in Group 4 females was decreased. This was considered incidental since an adequate number of females with evidence of mating were observed in each group.

***Reviewer comment:** No evidence was provided to support the claim that the decreased mating performance was incidental in females administered 300 ug/kg/day rhCNTF. This dose level is dramatically higher than the anticipated level of systemic rhCNTF exposure with ITV implantation of NT-501. Therefore, this reviewer does not consider this finding clinically relevant.*

 - Mating performance was comparable across all groups.
 - Of the females with evidence of mating, fertility as was similar across all groups.
- Limited necropsy of females sacrificed on gestational Day 14:
 - The number of corpora lutea was slightly lower in Groups 3 and 4 females than the Group 1.
 - The number of implantations was lower in Group 4 females compared to Group 1.
 - There were no changes in resorption index or implantation index between groups.
 - There were no gross anatomic changes in any groups.
- Gestational/neonatal data:
 - There were no rhCNTF-related differences in gestation period or pup survival indices in any groups.
 - The number of pups born alive was lower in Group 3 females compared to Group 1.
- Clinical observations of pups:
 - No rhCNTF-related differences were reported in the clinical condition, body weights, or in the physical development or behavioral assessments of pups across groups.
 - Unscheduled death numbers were:
 - 2 pups born to females in Group 1.
 - 4 pups born to females in Group 2.
 - 4 pups born to females in Group 3.
 - 5 pups born to females in Group 4.

- No rhCNTF-related differences in abnormalities of the pups that experienced unscheduled deaths were reported. Anophthalmia/microphthalmia and hydrocephaly were present in pups born to groups that received rhCNTF but were considered sporadic malformation and not rhCNTF-related. The incidences of these findings out of the pups with unscheduled deaths were:
 - 0/2 pups with anophthalmia/microphthalmia and hydrocephaly born to females in Group 1.
 - 2/4 pups with anophthalmia/microphthalmia and hydrocephaly born to females in Group 2.
 - 2/4 pups with anophthalmia/microphthalmia and hydrocephaly born to females in Group 3.
 - 3/5 pups with anophthalmia/microphthalmia and hydrocephaly born to females in Group 4.

Reviewer Comment: *As there is no dose-response observed with administration of increasing dose levels of rhCNTF, this reviewer agrees that these findings are unlikely to be related to rhCNTF administration.*

Reviewer conclusions:

- *A decrease in corpora lutea was observed in female rats administered >100 ug/kg/day of rhCNTF.*
- *No other clinically relevant findings were reported for mating performance, fertility, gestational parameters of female rats, or clinical observations of pups born to female rats, following SC administration of rhCNTF daily from Week -2 to postpartum Day 21.*

Study No. 30

Report Number		46-R-94-92997-007-SC-TT
Date Report Signed		October 11, 1994
Title		Subcutaneous teratology study in rats with recombinant human ciliary neurotrophic factor
GLP Status		No
Testing Facility		(b) (4)
Objective(s)		To determine the potential effects of rhCNTF on developmental parameters after maternal/embryonic drug exposure during organogenesis.
Study Animals	Strain/Breed	(b) (4)
	Species	<i>Rattus norvegicus</i>
	Age	Females: 15 weeks old Males: 3-12 months old
	Body Weight	Not specified
	#/sex/group	25-27 F/group Males not specified, but approximately 13 M/group
Total #		154
Test Article(s)		rhCNTF (Lot No. 9208D)

Control Article(s)	Vehicle (b) (4)			
Route of Administration	SC			
Description of the Disease/Injury Model	Healthy rats			
Study Groups and Dose Levels	Group	Animal Number Series	Concentration of rhCNTF (µg/ml)	Dose of rhCNTF (µg/kg/day)
	1	100	(b) (4)	0
	2	200		10
	3	300		100
	4	400		1000
	From: Module 4.2.3.5.1, Study Report #46-R-94-92997-007-SC-TT, page 5			
Dosing Regimen	Daily on gestational Days 7-16			
Randomization	Yes			
Description of Masking	Not provided			
Scheduled Sacrifice Time Points	Day 21 of gestation			

Summary:*Key evaluations and assessments:*

- Clinical observations/mortality: General condition was recorded daily and on gestational Days 1, 7, 14, 16, and 21.
- Body weights: Recorded on gestational Days 1, 7, 14, 17, and 21.
- Limited necropsy of females sacrificed on gestational Day 21: The reproductive tract, including the mammary chain, uterus, corpora lutea, live fetuses, resorptions, and any abnormal uterine conditions were grossly evaluated.
- Fetal observations:
 - At sacrifice, fetuses were weighed, examined externally, and sex was determined.
 - One-third of fetuses were preserved and examined for visceral abnormalities.
 - Two-thirds were eviscerated and processed for skeletal examination.

Key results:

- Clinical observations/mortality:
 - All rats survived the duration of the study.
 - Rats in Groups 1-3 were clinically normal throughout the study.
 - Rats in Group 4 had the following clinical changes on post-mating Days 15 and 16: inactivity, labored respiration, pallor, swollen face, and/or rough coat.
 - Rats in Group 4 were generally clinically normal for the remainder of the study.

- Body weights: A dose-responsive decrease in weight gain was present in Groups 3 and 4. Groups 1 and 2 had an average weight gain of 50g, Group 3 gained 30g, and no weight gain was observed in Group 4.
- Limited necropsy of females sacrificed on Day 14 of pregnancy:
 - No gross pathology changes were observed.
 - Pregnancy rates: Group 1-21/25, Group 2-25/27, Group 3-18/25, and Group 4-19/25.
Reviewer note: These pregnancy rates were not reported as abnormal.
 - The number of corpora lutea, total implantations, live fetuses, and resorptions were similar across groups.
- Fetal observations:
 - No treatment-related changes in fetal body weights.
 - The number of fetuses: Group 1-295 from 21 litters, Group 2-342 from 24 litters, Group 3-253 from 18 litters, and Group 4-221 from 19 litters.
Reviewer note: These litter sizes were reported as similar across groups.
 - No treatment-related external, visceral, or skeletal changes were present in fetuses.

Reviewer conclusions:

- *Decreased body weight gain was observed in a dose-responsive manner in pregnant rats administered ≥ 100 ug/kg/day rhCNTF SC.*
- *There were no rhCNTF-related teratologic changes.*

Study No. 31

Report Number		47-B-94-92997-007-SC-TT
Date Report Signed		October 28, 1994
Title		Subcutaneous teratology study in rabbits with recombinant human ciliary neurotrophic factor
GLP Status		No
Testing Facility		(b) (4)
Objective(s)		To evaluate the effects of hCNTF on developmental parameters in rabbits after maternal/embryonic drug exposure during organogenesis.
Study Animals	Strain/Breed	(b) (4)
	Species	Rabbit
	Age	Not specified

	Body Weight	Not specified		
	#/sex/group	20 F/group		
	Total #	80		
Test Article(s)		rhCNTF (Lot No. 01093-06DE)		
Control Article(s)		Vehicle (b) (4)		
Route of Administration		SC		
Description of the Disease/Injury Model		Healthy (b) (4) rabbits		
Study Groups and Dose Levels		<u>Group</u>	<u>Animal Number Series</u>	<u>Concentration of rhCNTF (µg/ml)</u>
		1	100	(b) (4)
		2	200	
		3	300	
		4	400	
		<u>Dose of rhCNTF (µg/kg/day)</u>		
		0		
		2		
		5		
		10		
		<i>From: Module 4.2.3.5.1, Study Report #47-b-94-92997-007-SC-TT, page 5</i>		
Dosing Regimen		Daily on gestational Days 7-19		
Randomization		Yes		
Description of Masking		Not provided		
Scheduled Sacrifice Time Points		Gestational Day 29		

Reviewer note: The study report uses ‘gestational’ and ‘post-mating’ Day interchangeably. This reviewer uses ‘gestational’ only.

Summary:

Key evaluations and assessments:

- Clinical observations/mortality: General condition was recorded daily and on gestational Days 1, 7, 13, 19, 22, and 29 and whenever a change in clinical condition occurred.
- Body weights: Recorded on gestational Days 1, 7, 13, 19, and 22, and 29.
- Limited necropsy of females sacrificed on gestational Day 29: The reproductive tract, including the mammary chain, uterus, corpora lutea, live fetuses, resorptions, and any abnormal uterine conditions were grossly evaluated.
- Fetal observations:
 - At sacrifice, fetuses were weighed, examined externally, and sex was determined.
 - Fetuses were euthanized and viscera examined.

Key results:

- Clinical observations/mortality:
 - Rabbits in Groups 1 and 2 were clinically normal throughout the study.
 - One rabbit in Group 3 was anorexic gestational Days 20-22.

- Most rabbits in Group 4 were anorexic and/or wasting beginning on gestational Day 13.
- Three rabbits in Group 4 aborted between gestational Days 22 and 27 and were euthanized and necropsied.
- Body weights:
 - Groups 1-3 had similar body weight gain.
 - Group 4 rabbits lost body weight compared to Group 1 during the dosing period (gestational Days 7-19) and gained weight after cessation of dosing.
- Limited necropsy of females sacrificed on gestational Day 29:
 - Gross changes were observed in 2/3 dams in Group 4 that aborted and were euthanized early.
 - 1 dam had red tracheal discoloration and no body fat.
 - 1 dam had liver and kidney pallor, and gas in the stomach and large intestine.
 - Pregnancy rates: Group 1-16/20, Group 2-18/20, Group 3-16/20, and Group 4 17/20.
 - The number of total implantations and live fetuses were lower in Group 4 and Group 1.
 - There were no changes in the number of corpora lutea or resorptions across groups.
- Fetal observations:
 - There were no rhCNTF-related changes in fetal body weights.
 - The number of fetuses: Group 1-107 from 16 litters, Group 2-126 from 18 litters, Group 3-103 from 16 litters, and Group 4-44 from 9 litters.
 - No rhCNTF-related external, visceral, or skeletal changes were reported.

Reviewer conclusions:

- *Anorexia, abortion, and body weight loss were present in rabbits administered 10 µg/kg/day rhCNTF SC.*
- *There were no rhCNTF-related teratologic changes.*

Study No. 32:

Study Report #NEU-10

Title: DART Exemption Evaluation of NT-50147-B-94-92997-007-SC-TT

Date Signed: Not signed.

The applicant submitted a Toxicological Risk Assessment (TRA) to support an exemption from Developmental and Reproductive Toxicity (DART) Evaluation of NT-501 (Study Report #NEU-10). This evaluation was completed in April 2022 by (b) (4), to support an exemption from conducting a DART study with NT-501. This study reviewed the data in Studies Nos. 28-31 in this memo and the PK data reviewed in Study No. 6, as well as data available in the literature from Dittrich, F, et al. (1994)¹. In addition, the applicant provided in silico modeling of systemic rhCNTF accumulation arising from intraocular NT-501 implantation to predict systemic exposure from intraocular implantation of devices releasing 20 ng/day rhCNTF. (b) (4) noted that not all stages of development and reproduction were assessed. Particularly, male and female fertility were not fully evaluated and estrous cyclicity and sperm parameters, development and maturation of gametes, and time to conception were not assessed. In addition, not assessments were conducted regarding post-weaning development and growth, adaptation, puberty, etc. of pups born in the completed studies.

(b) (4) compared the predicted maximum systemic rhCNTF blood concentration identified in the rat and rabbit PK studies (0.00017 µg/mL in rats and 0.00004 µg/mL in rabbits) to the no-observed-adverse-effect-level (NOAELs) identified in Studies #9-12 to calculate the margins of exposure (MOE).

Reviewer note: MOE is commonly used in human health risk assessment to determine whether anticipated human exposure is likely to be associated with adverse health effects and is calculated as a ratio between the NOAEL and the human exposure level. Generally, an MOE of at least 100 is considered protective of human health.

(b) (4) provided the below table with MOE calculations for each species:

Study	NOAEL	Equivalent Blood Concentration of NOAEL	MOE Calculation	MOE
Male (b) (4) rat fertility study (#44-R-94-92997-007-SC-MR)	100 µg/kg/day	100 µg/kg/day * 0.2048 kg / 25 mL = 0.82 µg/mL/day	0.82 µg/mL/day ÷ 0.00017 µg/mL	4,823
Female (b) (4) rat fertility study (#45-R-94-92997-007-SC-RP)	10 µg/kg/day	10 µg/kg/day * 0.2653 kg / 25 mL = 0.11 µg/kg/day	0.11 µg/mL/day ÷ 0.00017 µg/mL	647
Female (b) (4) rat teratology study (#46-R-94-92997-007-SC-TT)	10 µg/kg/day	10 µg/kg/day * 0.2793 kg / 25 mL = 0.11 µg/kg/day	0.11 µg/mL/day ÷ 0.00017 µg/mL	647
Female (b) (4) rabbit teratology study (#47-B-94-92997-007-SC-TT)	5 µg/kg/day	5 µg/kg/day * 3.1243 kg / 120 mL = 0.13 µg/mL/day	0.13 µg/mL/day ÷ 0.00004 µg/mL	3,250

From: Module 4.2.3.7.7, Study Report #NEU-10, page 23

(b) (4) concluded that the estimated blood rhCNTF levels in the Studies #9-12 are 3-4 order of magnitude higher than the modeled maximum blood rhCNTF levels in rats and rabbits because of intraocular dosing. As shown in the above table, MOEs are above 100, indicating adequate protection of human health and a low risk of adverse reproductive and developmental effects.

Reviewer conclusion:

Based on lack of meaningful exposure and the absence of health risks identified by multiple DART studies, additional DART studies are not warranted for rhCNTF.

Genotoxicity Studies:

Study Number	Study Title / Publication Citation	Report Number
33	Genotoxicity: (b) (4) Study	v0023-211
34	Genotoxicity: (b) (4) Study in Mammalian Cells (Extract)	v0002-130
35	Mouse Bone Marrow Micronucleus Study	t0212-500
36	Kligman Maximization Test –ISO	22-00760-g3

Reviewer note: *These tests were conducted to satisfy, in part, the requirements of the International Organization for Standardization (ISO) 10993, Part 3: Tests for Genotoxicity, Carcinogenicity and Reproductive Toxicity.*

Study No. 33

Report Number	v0023-211
Date Report Signed	October 30, 2001
Title	Genotoxicity: (b) (4) study
GLP Status	Yes
Testing Facility	(b) (4)
Objective(s)	(b) (4)
Test Article(s)	(b) (4)
Control Article(s)	(b) (4)

<p>Description of Test System</p>	<p>(b) (4)</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p>
<p>Study Groups</p>	<p>(b) (4)</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p> <p>[Redacted]</p>

Key results:

- There were no instances of a 2-fold or greater increase in the mean number of revertants of tester strains.
- Each positive control mean exhibited at least a 3-fold increase over the respective mean of the tester strains employed.

Reviewer conclusions:

- Under the conditions of this assay, the (b) (4) test article extract was non-mutagenic to (b) (4)
[Redacted]
- The negative and positive controls performed as anticipated.

Study No. 34

Report Number	v0003-130
Date Report Signed	November 29, 2001
Title	Genotoxicity: (b) (4) study in mammalian cells (extract)
GLP Status	Yes
Testing Facility	(b) (4)
Objective(s)	To determine whether an extract of the test article would cause clastogenic changes in (b) (4) in the presence and absence of (b) (4) metabolic activation.
Test Article(s)	(b) (4) of NT-501 prepared at (b) (4).
Control Article(s)	Negative: (b) (4) subjected to the extraction conditions without NT-501 Positive: Known genotoxic compounds: (b) (4)
Description of Test System	(b) (4)
Study Groups	<ol style="list-style-type: none"> 1. Negative control (b) (4) with and without (b) (4) activation 2. (b) (4) 3. (b) (4) of NT-501 with and without (b) (4) activation

Key results:

- The percentage of cells with (b) (4) for Group 3 (test article) was similar to that of the (b) (4) control (Group 1) (2.0-3.3% simple aberrations, 0.0-0.3% complex aberrations).
- The positive control groups (Groups 2) had high rates of (b) (4) (54-58% simple aberrations, 23-25% complex aberrations).

Reviewer conclusions:

- *Under the conditions of this assay, the test article extract was not considered genotoxic to (b) (4) in the presence or absence of (b) (4) metabolic activation.*
- *The negative and positive controls performed as expected.*

Study No. 35

Report Number	t0212-500
----------------------	-----------

Date Report Signed		January 21, 2002
Title		Mouse Bone Marrow Micronucleus Study
GLP Status		Yes
Testing Facility		(b) (4)
Objective(s)		To evaluate the test article (NT-501, Lot #DM0023) extracted in (b) (4) for genotoxicity using the Mouse Bone Marrow Micronucleus model.
Study Animals	Strain/Breed	(b) (4)
	Species	<i>Mus musculus</i>
	Age	6-8 weeks
	Body Weight	Not specified
	#/sex/group	5/sex/group
	Total #	30
Test Article(s)		(b) (4) of NT-501 prepared at (b) (4).
Control Article(s)		Negative: (b) (4) subjected to the extraction conditions without NT-501 Positive: Known mutagenic compound (b) (4)
Route of Administration		Intraperitoneal (IP)
Description of the Disease/Injury Model		Healthy (b) (4) mice
Description of Test System		A Mouse Bone Marrow Micronucleus study determines whether a test article extract causes genotoxic changes in chromosomes or the mitotic apparatus of murine polychromatic erythrocytes (PCEs). An increase in frequency of micronucleated polychromatic erythrocytes of treated animals is used as an indication of genetic toxicity. The percentage of polychromatic erythrocytes among total erythrocytes (polychromatic + normochromatic) is used as an index of bone marrow toxicity.
Study Groups		1. Negative control (b) (4) 2. Positive control (b) (4) 3. (b) (4) of NT-501
Dosing Regimen		Groups 1 and 3 received an IP injection for 2 consecutive days (Days 1 and 2). Group 2 received a single IP injection on Day 2.
Randomization		Yes
Description of Masking		Not provided
Scheduled Sacrifice Time Points		Day 3

Key evaluations and assessments:

- Clinical observations: Mice were observed daily for general health. Immediately post-injection they were observed for any adverse reaction.
- Body weights: Measured on Days 1 and 3 (sacrifice).
- Bone marrow evaluation:
 - Bone marrow slides were microscopically examined:
 - PCEs were scored for the presence of micronuclei.
 - A minimum of 2000 PCEs were examined for the presence of micronuclei.
 - The number of normochromatic erythrocytes were observed and the ratio of PCEs to total erythrocytes was calculated as an index of bone marrow toxicity.

Key results:

- Clinical observations: All animals were clinical normal throughout the study.
- Body weights: Weight changes were normal throughout the study.
- Bone marrow evaluation:
 - Groups 1 (negative control) and 3 (test article) had similar percentages of PCEs (37-38% in Group 1, 31.3-39% in Group 3) and numbers of PCEs with micronuclei were similar (1.4-1.5/2000 in Group 1, 1.4-1.6/2000 in Group 3).
 - Group 2 (positive control) had 13-17% PCEs and 10-11.7/2000 PCEs with micronuclei, indicating bone marrow toxicity.

Reviewer conclusions:

- *Under the conditions of this study, the test article extract was not considered to be genotoxic to the mouse.*
- *There was no evidence of cellular toxicity.*
- *The negative and positive controls performed as expected.*

Study No. 36

Report Number		22-00760-G3
Date Report Signed		August 26, 2022
Title		Kligman Maximization Test -- ISO
GLP Status		Yes
Testing Facility		(b) (4)
Objective(s)		To determine the potential allergenic or sensitizing capacity of the test article.
Study Animals	Strain/Breed	(b) (4) guinea pig
	Species	<i>Cavia porcellus</i>
	Age	6-8 weeks
	Body Weight	320-455 g
	#/sex/group	Sex distribution not specified
Total #		35
Test Article(s)		1. (b) (4) of NT-501 titanium clip prepared at (b) (4) 2. (b) (4) of NT-501 titanium clip prepared at (b) (4)
Control Article(s)		Negative: (b) (4) or (b) (4) subjected to the extraction conditions without NT-501 titanium clip Positive: Known mutagenic compound (b) (4)
Route of Administration		Intradermal (ID) injection Topical Application
Description of the Disease/Injury Model		Healthy guinea pigs

Study Groups	<ol style="list-style-type: none"> 1. (b) (4) negative control (5 animals) 2. (b) (4) negative control (5 animals) 3. Positive control (b) (4) (5 animals) 4. (b) (4) extract of NT-501 titanium clip (10 animals) 5. (b) (4) of NT-501 titanium clip (10 animals)
Dosing Regimen	<p>Induction phase: 3 pairs of ID injections (6 total) were made, one on each side of the midline.</p> <p>Topical application:</p> <ul style="list-style-type: none"> • On Day 6, animals that showed no signs of irritation or corrosion after the induction phase were pre-treated with (b) (4) • 24 hrs. later, 0.3 mLs of the indicated solution (negative control, positive control, test article) was used to saturate a 2cm x 4cm piece of absorbent material. The patch was placed on the injection sites and secured with an occlusive wrapping or guinea pig jacket and left in place for 48 hrs. <p>Challenge application:</p> <ul style="list-style-type: none"> • On Day 23, 2cm x 2cm pieces of absorbent material were saturated with the appropriate solution and secured to the injection sites with a wrapping described for the topical application.
Randomization	Yes
Description of Masking	Not provided
Scheduled Sacrifice Time Points	End of study

Summary:*Key evaluations and assessments:*

- Clinical observations/mortality: Animals were monitored daily.
- Sensitization scoring: The System of Magnusson and Kligman was used that evaluates sensitization based on a grading scale of observed reactions (Grade 0=no visible change, 1=discrete or patch erythema, 2=moderate and confluent erythema, 3=intense erythema and swelling) and determines the sensitization classification based on the percentage of the group that is positive for a reaction (0=nonsensitizer, <10=weak, 10-30=mild, 31-60=moderate, 61-80=strong, 80-100=extreme).

Key results:

- Clinical observations/mortality: All animals survived for the duration of the study. No systemic signs of toxicity were observed in any groups.
- Sensitization scoring:
 - None of the animals in Groups 1, 2, 4, and 5 exhibited any reaction to the challenge (0% sensitized).

- Group 3 elicited a discrete Grade 1 reaction in four animals and moderate Grade 2 reactions in one animal (100% sensitized).

Reviewer conclusion: As defined by the grading scale of the (b) (4), the test article is classified as a non-sensitizer.

Carcinogenicity/Tumorigenicity Studies:

Study Number	Study Title / Publication Citation	Report Number
37	Two-Year Cancer Study Waiver Recommendation for the (b) (4) NT- 501-6A.02 Device	Neurotech 8
38	Cancer Study Waiver Recommendation	Neurotech 9

Studies No. 37-38

The applicant provided TRAs conducted by (b) (4) to support waivers for carcinogenicity/tumorigenicity studies for rhCNTF and the medium used within the device, human Endothelial-SFM medium (ENDO medium).

Per the TRA, studies were not conducted to evaluate this safety endpoint for rhCNTF for the following reasons:

- Relevant toxicokinetic and toxicodynamic data indicate that rhCNTF has a very short systemic half-life and dose not bioaccumulate.
- Animal studies and human studies consistently demonstrate that continuous local release of rhCNTF from implanted NT-501-6A.02 devices does not result in systemic exposure of rhCNTF proteins.
- A literature search did not identify epidemiological or toxicological studies or human case reports pertaining to exposure to rhCNTF and subsequent carcinogenic effects.
- Two preclinical studies in minipigs or pigs implanted with NT-501-6A.02 device for up to 6 months showed no immune response to rhCNTF or cell producing the protein. No neoplastic or pre-neoplastic lesions were found in implanted eyes. Mild to moderate severity inflammatory changes were observed.

Per the TRA, studies were not conducted to evaluate this safety endpoint for ENDO medium for the following reasons:

- Less than (b) (4) of ENDO medium will be present in one NT-501 device.
- (b) (4) conducted a risk assessment on every known chemical in the ENDO medium, including inorganic salt, amino acids, and vitamins, and derived tolerable intakes, human daily exposure dose, assuming two implanted devices for at least 6

months, and margins of safety (MOS) for each compound. An MOS of >1 indicates a low health concern.

- The MOS for all chemicals in ENDO medium were >5,000.
- (b) (4) concludes that the presence of residual ENDO medium in Neurotech's implanted devices assessed is not expected to pose a significant systemic toxicity risk and that there is adequate justification for waiving two-year cancer studies.

Reviewer Comment:

- *This reviewer agrees with the rationale provided in the TRAs for not conducting carcinogenicity/tumorigenicity studies.*

Other Safety/Toxicology Studies

The studies listed below were conducted to evaluate biocompatibility and are incorporated into the Chemistry, Manufacturing, and Controls review memo.

Study Number	Study Title / Publication Citation	Report Number
39	(b) (4)	
40		
41		
42		
43		
44		
45		
46		
47		
48		
49		
50		
51		
52		
53		
54		

Analytical Methods and Validation Reports

The following studies are not summarized in this review memo; they are primarily assay development reports for analytical testing methods used for serum, plasma, and liver tissue samples from the nonclinical studies.

Study Number	Study Title / Publication Citation	Report Number
55	(b) (4)	
56		
57		
58		
59		
60		
61		
62		
63		

APPLICANT'S PROPOSED LABEL

Subsections 8.1-8.3 ('Use in Specific Populations') should be revised to comply with 21 CFR 201.56(d)(1), 201.57(c)(9), and 201.57(c)(14), as applicable.

Section 12.3 ('Pharmacokinetics - Nonclinical data') should be revised, as applicable, to accurately reflect the available nonclinical data.

Section 13 ('Nonclinical Toxicology') should be revised, as applicable, to accurately reflect the available nonclinical data.

CONCLUSION OF NONCLINICAL STUDIES

Review of the available nonclinical studies did not identify any safety concerns that could not be addressed in the product label. The nonclinical data support approval of the license application.

KEY WORDS/TERMS

ENCELTO, revakinagene tarorectel, NT-501, NTC-201-6A cells, recombinant human ciliary neurotrophic factor, rhCNTF, hCNTF, CNTF, idiopathic macular telangiectasia type 2, MacTel, rod-cone dysplasia type 1 canine model, rcd1 canine model, Good Laboratory Practice, intravitreal, intraocular, sclerotomy, (b) (4) rabbit, gene therapy, cell therapy, device, combination product, genotoxicity, developmental and reproductive toxicity, pharmacokinetics, pharmacology, toxicology
