WAVE LIFE SCIENCES

Phosphoryl guanidine (PN)-containing oligonucleotides support exon skipping in skeletal muscle in mice and boys with DMD

Pachamuthu Kandasamy

Vice President, Medicinal Chemistry

May 17, 2023

26th Annual ASGCT meeting, May 16-20, 2023

Forward-looking statements

This document contains forward-looking statements. All statements other than statements of historical facts contained in this document, including statements regarding possible or assumed future results of operations, preclinical and clinical studies, business strategies, research and development plans, collaborations and partnerships, regulatory activities and timing thereof, competitive position, potential growth opportunities, use of proceeds and the effects of competition are forward-looking statements. These statements involve known and unknown risks, uncertainties and other important factors that may cause the actual results, performance or achievements of Wave Life Sciences Ltd. (the "Company") to be materially different from any future results, performance or achievements expressed or implied by the forward-looking statements. In some cases, you can identify forward-looking statements by terms such as "may," "will," "should," "expect," "plan," "aim," "anticipate," "could," "intend," "target," "project," "contemplate," "believe," "estimate," "predict," "potential" or "continue" or the negative of these terms or other similar expressions. The forward-looking statements in this presentation are only predictions. The Company has based these forward-looking statements largely on its current expectations and projections about future events and financial trends that it believes may affect the Company's business, financial condition and results of operations. These forward-looking statements speak only as of the date of this presentation and are subject to a number of risks, uncertainties and assumptions, including those listed under Risk Factors in the Company's Form 10-K and other filings with the SEC, some of which cannot be predicted or quantified and some of which are beyond the Company's control. The events and circumstances reflected in the Company's forward-looking statements may not be achieved or occur, and actual results could differ materially from those projected in the forward-looking statements. Moreover, the Company operates in a dynamic industry and economy. New risk factors and uncertainties may emerge from time to time, and it is not possible for management to predict all risk factors and uncertainties that the Company may face. Except as required by applicable law, the Company does not plan to publicly update or revise any forward-looking statements contained herein, whether as a result of any new information, future events, changed circumstances or otherwise.

WAVE
LIFE SCIENCES

Disclosures

Pachamuthu Kandasamy[1], Graham McClorey, Laurent Servais, Craig Campbell, Xiao Shelley Hu[1], Andrew Hart[1], Joseph Haegele[1], Kuldeep Singh[1], Jeanette Rheinhardt[1], Anamitra Ghosh[1], Mamoru Shimizu[1], Nayantara Kothari[1], Naoki Iwamoto[1], Michael Byrne[1], Fangjun Liu[1], Chikdu Shivalila[1], Carlo Rinaldi, Hailin Yang[1], Danlin Xu[1], Stephen Lake[1], Michael Panzara[1], Anne-Marie Li-Kwai-Cheung[1], Matthew Wood, Chandra Vargeese[1]

[1]Employees of Wave Life Sciences

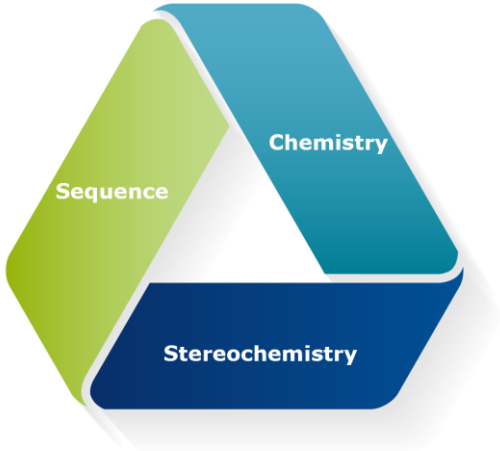
Laurent Servais has provided consultancy and lectures, and attended advisory boards for Sarepta, Dyne, Pfizer, Santhera, RegenxBio, Affinia, and Fibrogen

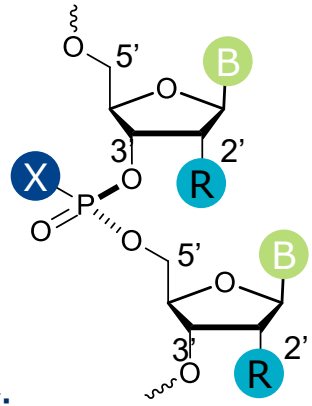
Craig Campbell has served as site investigator for Acceleron, AMO, Biogen, Dyne, Fibrogen, Pfizer, Roche, PTC, Sarepta, Cytokinetics, Ultragenix, and Wave and has acted as a Data Safety Monitoring Board member for Catabasis, Edgewise, and Solid. Additionally, he has received investigator-initiated grants from Genzyme, PTC Therapeutics, and Biogen

Matthew Wood is a cofounder of PepGen

The data in this presentation are from preliminary analyses of an ongoing clinical trial

PRISM™ platform enables rational drug design



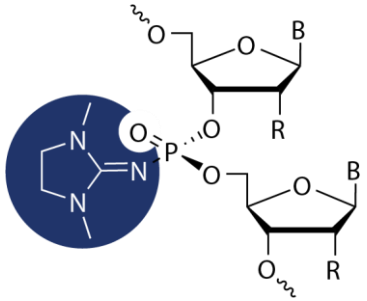


X:
O: Phosphodiester
S: Phosphorothioate
N: Phosphoryl guanidine

B Base

R 2'-ribose modification

X Stereochemistry & backbone modification



Phosphoryl guanidine

WAVE LIFE SCIENCES

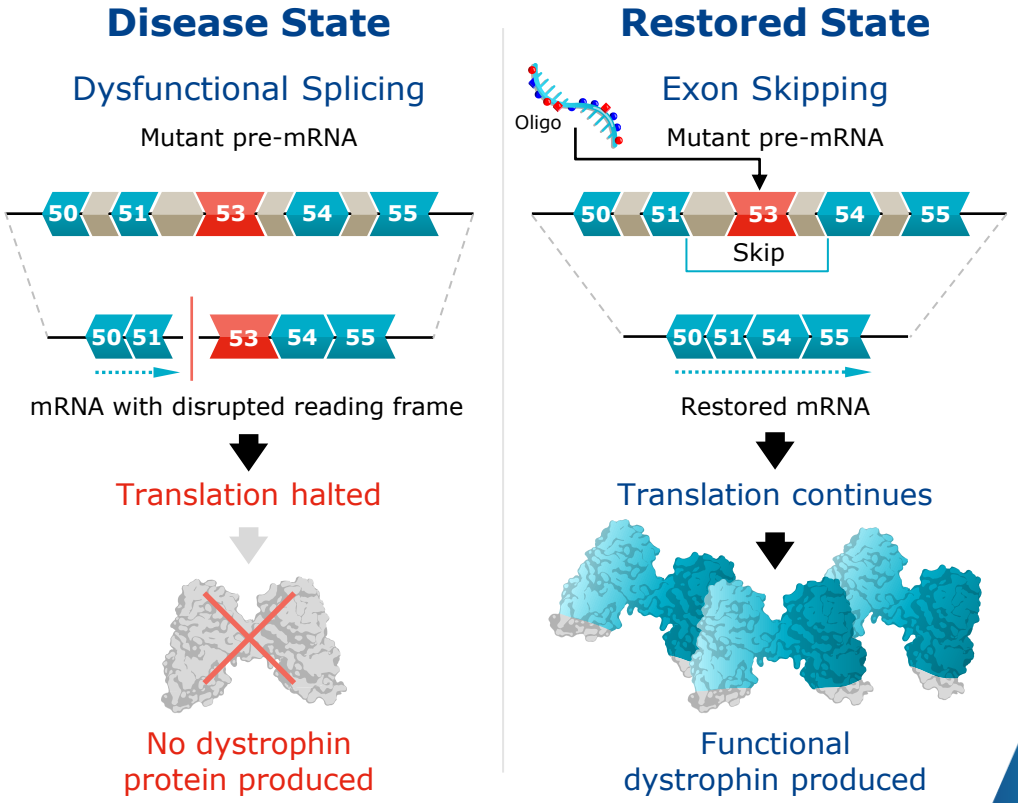
Duchenne muscular dystrophy and WVE-N531

Duchenne Muscular Dystrophy

A rapidly progressive, rare disease characterized by irreversible muscle degeneration and weakness, loss of ambulation and upper body function, and cardiorespiratory complications

WVE-N531

- Investigational exon-skipping oligonucleotide
- Contains PN chemistry
- Designed to skip exon 53
- 8-10% of patients with DMD are amenable to exon 53 skipping[1]



1. Aartsma-Rus *et al.*, 2009 Hum Mut doi: 10.1002/humu.20918

PN chemistry improves pharmacological properties in multiple preclinical model systems

***mdx23* mouse**

	Protein	Pathology
✓	Utrophin	Mild No decrease in survival
×	Dystrophin	

Double knockout mouse (dKO)

	Protein	Pathology
×	Utrophin	Severe muscular dystrophy Premature death
×	Dystrophin	

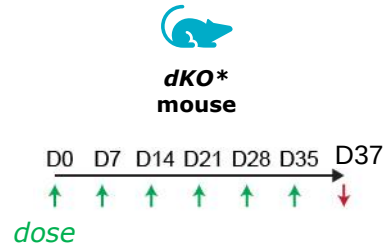
Nonhuman primate (NHP)

	Protein	Pathology
✓	Utrophin	Healthy
✓	Dystrophin	

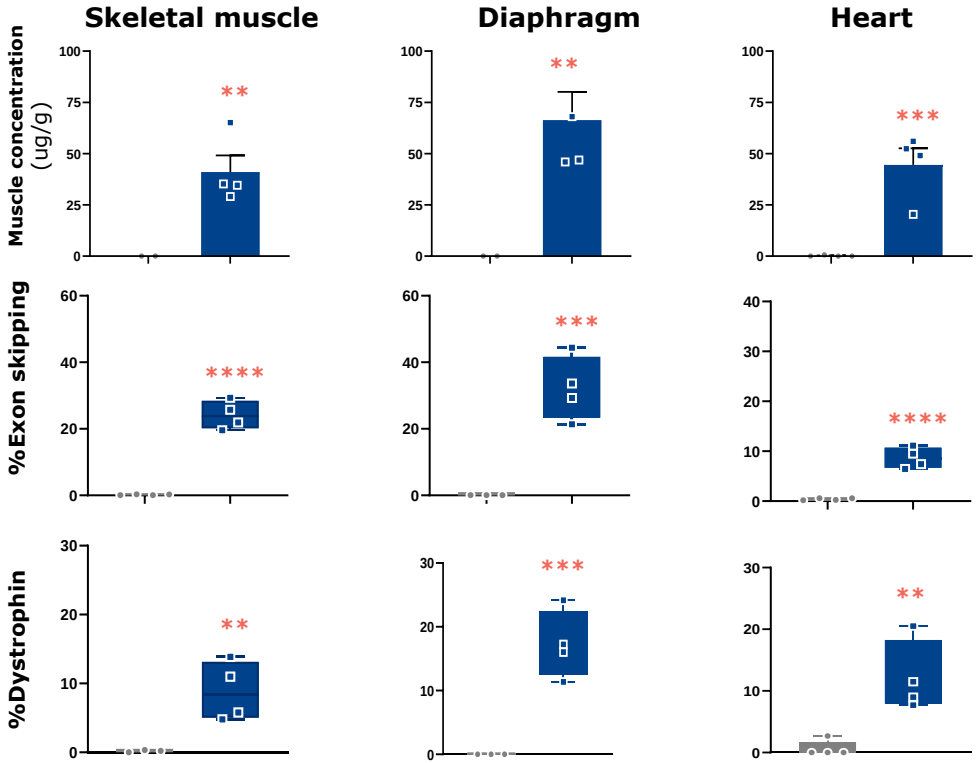
NHP: cynomolgous monkey

WVE-N531 surrogate yields excellent muscle exposure, exon skipping and dystrophin protein expression in *dKO* mouse model

- PBS
- WVE-N531 surrogate for mouse Dmd exon 23



*Similar results observed in *mdx23* mouse



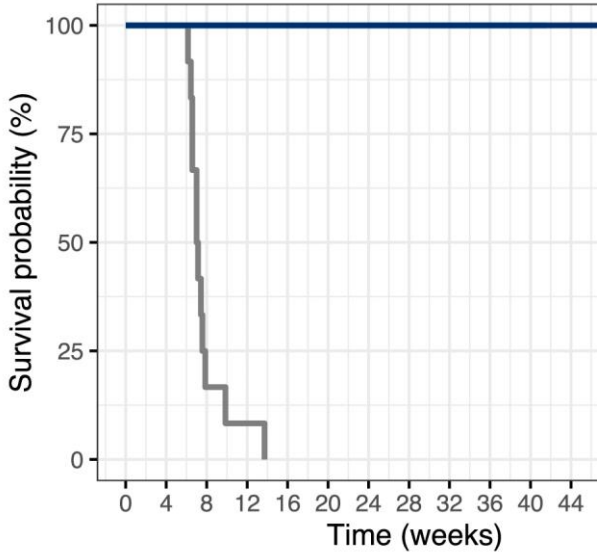
Kandasamy et al., 2022 *Nuc Acids Res* doi: 10.1093/nar/gkac018

Biweekly administration of WVE-N531 surrogate improves survival and muscle function in dKO mouse model

dKO
biweekly
dosing

Median survival	
PBS	49 days
WVE-N531 surrogate	280 days

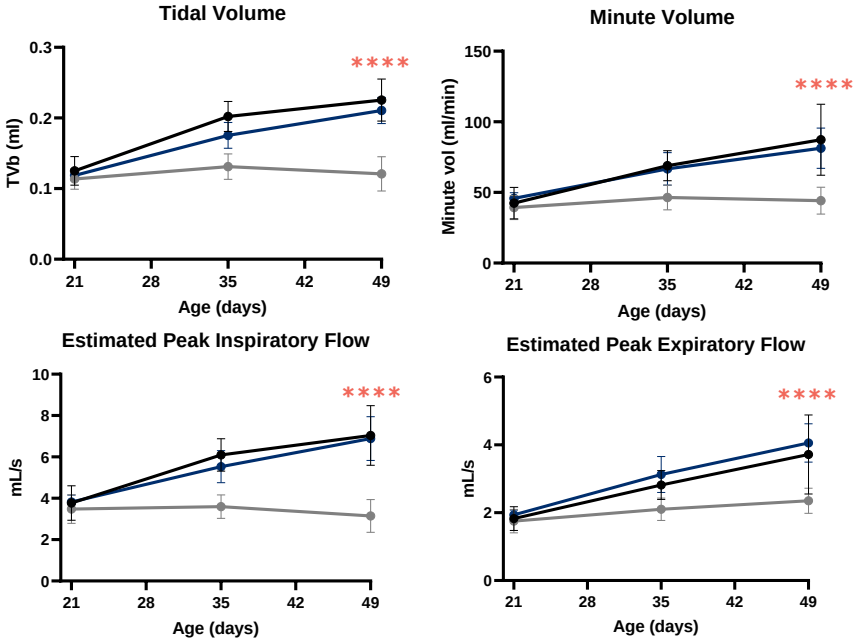
$P=2 \times 10^{-11}$



Respiratory function comparable to age-matched healthy controls

(6 x weekly doses, dKO mouse)

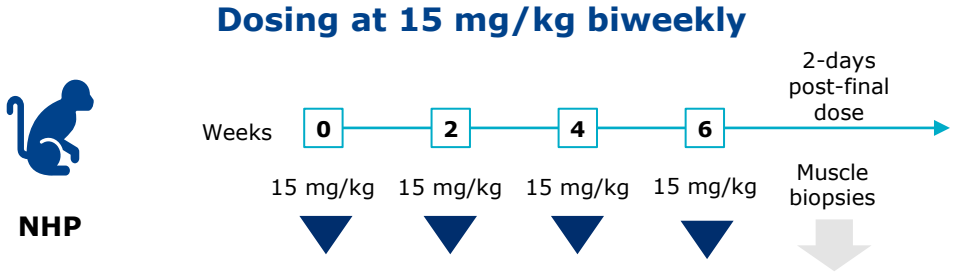
Healthy controls — Treated dKO — PBS



Kandasamy et al., 2022 *Nuc Acids Res* doi: 10.1093/nar/gkac018

WAVE LIFE SCIENCES

WVE-N531 reached high concentrations in heart and diaphragm in NHP



	Mean Tissue Concentration		
	Skeletal muscle	**Diaphragm**	**Heart**
15 mg/kg* IV dose	2.17 ug/g	10.8 ug/g	57.2 ug/g

*approximately equivalent to 10 mg/kg in patients based on plasma AUC values

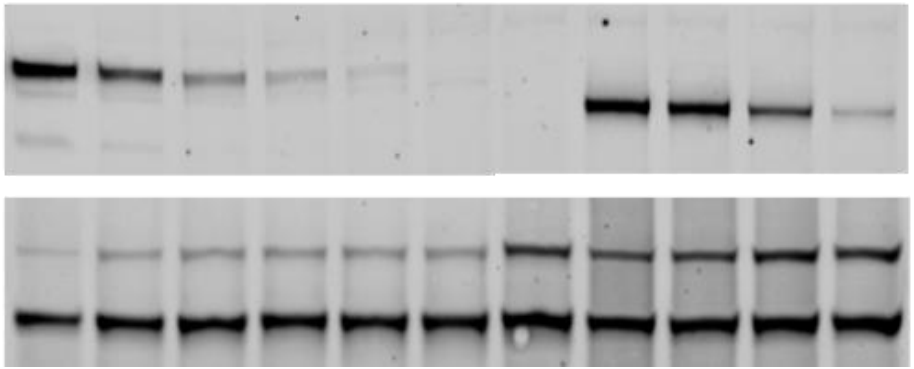
Muscle concentrations of WVE-N531 were measured 2 days post-last dose by HL-ELISA.
AUC: area under the plasma concentration-time curve; IV: intravenous

WVE-N531 restores dystrophin expression in patient-derived myoblasts

Dystrophin protein restoration up to 71% *in vitro*

western blot normalized to primary healthy human myoblast lysate

Standard Curve (% WT lysate in D45-52 lysate)						D45-52 Cells				
						Mock	WVE-N531			
						0	10	3.3	1.1	0.3 Conc [uM]
100%	50%	25%	12%	6%	0%	0%	71%	65%	37%	9.5% % Dystrophin

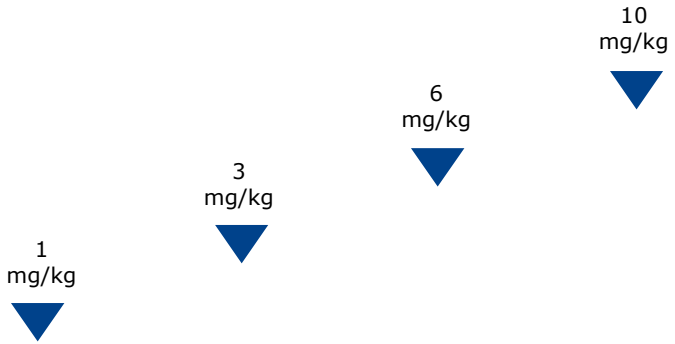


WAVE LIFE SCIENCES

WVE-N531: First-in-human study design

WVE-N531-001: First-in-human study

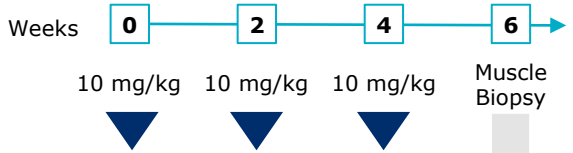
Single-ascending intra-patient doses



Initial cohort
Boys amenable to exon 53 skipping (n=3)

Period before initiating multidosing (~1–2 months)

Multidosing at 10 mg/kg every other week



Endpoints:	
Primary	• Safety and tolerability of WVE-N531
Secondary	• WVE-N531 muscle concentrations • WVE-N531 localization • Exon skipping • Dystrophin protein

WAVE LIFE SCIENCES

▼ Dose WVE-N531

Muscle concentrations and exon skipping indicate WVE-N531 is engaging target

Plasma $t_{1/2}$: 25 days (10 mg/kg single dose)

Patient	Tissue Source	Tissue concentration (µg/g)	% Exon skipping by RT-PCR	Dystrophin by western blot (% of normal)
1	Deltoid	85.5	61.5	0.24
2	Deltoid	33.5	49.8	0.23
3	Bicep	8.3	47.9	0.34

Mean muscle concentration: 42 µg/g

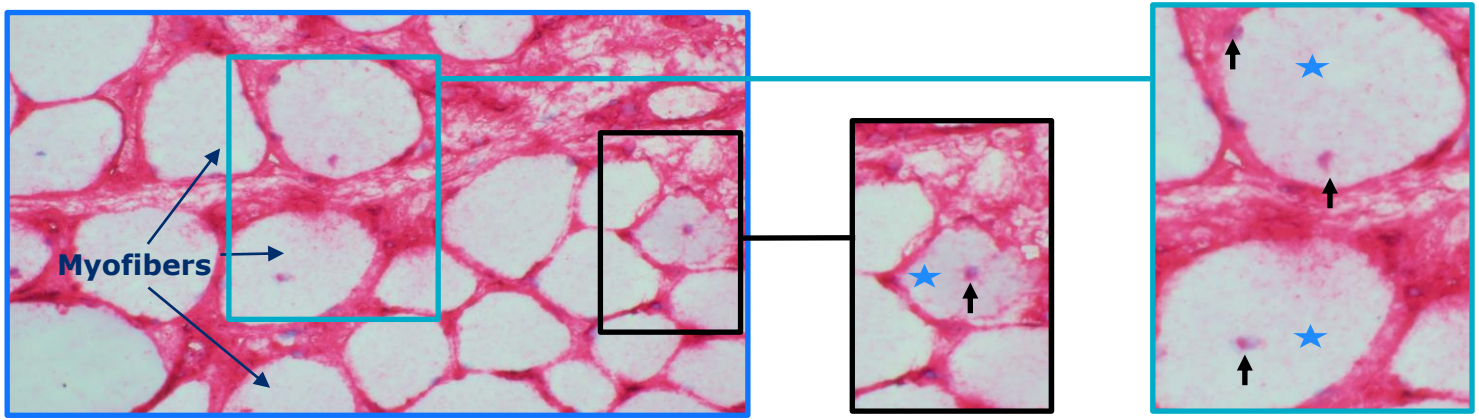
Mean exon skipping: 53%

Mean dystrophin: 0.27% of normal (BLQ)

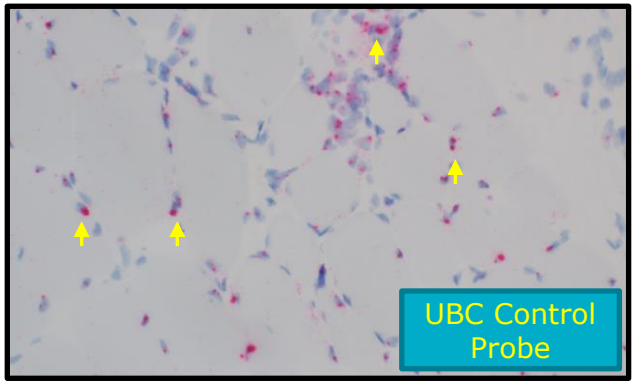
WVE-N531 was generally safe and well-tolerated

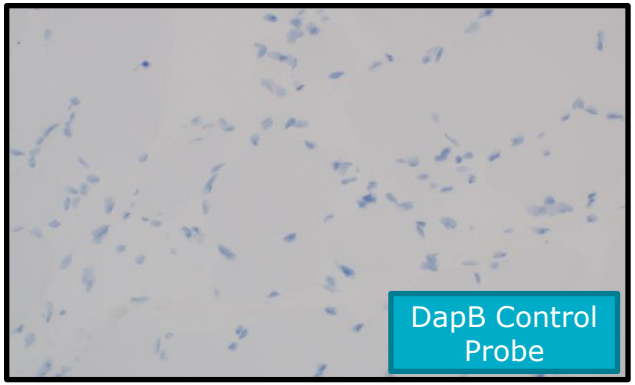
Biopsies collected ~2 weeks post-last dose (3 biweekly doses of 10 mg/kg)
BLQ: Below level of quantification (1%); 42 µg/g = 6.1 µM; $t_{1/2}$: half-life

Intracellular WVE-N531 enabling exon 53 skipping



WVE-N531 (in red) in myofiber cytoplasm (stars) and nuclei (black arrows)





WAVE LIFE SCIENCES

RNAscope (ISH – in situ hybridization). Mag: 40x for WVE-N531; Mag: 20x for control probes
Control Probes: Ubiquitin (UBC) – Positive control probe showing ubiquitin mRNA (yellow arrows); DapB – Negative control

Summary

- WVE-N531 surrogate increased muscle concentration, percentage of exon skipping and dystrophin expression in *mdx23* and dKO mouse models compared with control
- In dKO model, WVE-N531 surrogate substantially extended survival and rescued respiratory function
- In NHPs, WVE-N531 reached high concentrations in heart and diaphragm
- In boys with DMD, WVE-N531 reached high concentrations in skeletal muscle (mean 42 µg/g), and induced 53% mean exon skipping following three biweekly 10 mg/kg doses
- Half-life supports biweekly or less frequent dosing
- WVE-N531's safety profile supports further development
- Wave is initiating Part B: Phase 2 open-label clinical trial for boys amenable to exon 53 skipping, with data expected in 2024

WAVE LIFE SCIENCES

Acknowledgements

Study participants & families

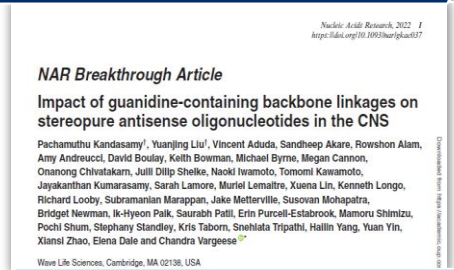
Investigators

- Laurent Servais
- Craig Campbell

Thanks to all colleagues and contributors from **Wave Life Sciences** and our collaborators

Wave publications on PN chemistry

Silencing - antisense



NAR Breakthrough Article

Impact of guanidine-containing backbone linkages on stereopure antisense oligonucleotides in the CNS

Pachamuthu Kandasamy[1], Yuanjing Liu[1], Vincent Aduda, Sandheep Akare, Rowshon Alam, Amy Andreucci, David Boulay, Keith Bowman, Michael Byrne, Megan Cannon, Onanong Chivatakarn, Juill Dilip Shelke, Naoki Iwamoto, Tomomi Kawamoto, Jayakanthan Kumarasamy, Sarah Lamore, Muriel Lemaitre, Xuena Lin, Kenneth Longo, Richard Looby, Subramanian Marappan, Jake Metterville, Susovan Mohapatra, Bridget Newman, Ik-Hyeon Paik, Saurabh Patil, Erin Purcell-Estabrook, Mamoru Shimizu, Pochi Shum, Stephany Standley, Kris Taborn, Snehlata Tripathi, Hailin Yang, Yuan Yin, Xiansi Zhao, Elena Dale and Chandra Vargeese[*]

Wave Life Sciences, Cambridge, MA 02138, USA

Silencing - siRNA

Impact of stereopure chimeric backbone chemistries on the potency and durability of gene silencing by RNA interference

Wei Liu[†], Naoki Iwamoto[†], Subramanian Marappan, Khoa Luu, Snehlata Tripathi, Erin Purcell-Estabrook, Juili Dilip Shelke, Himali Shah, Anthony Lamattina, Qianli Pan, Brett Schrand, Frank Favaloro, Mugdha Bedekar, Arindom Chatterjee, Jigar Desai[*], Tomomi Kawamoto, Genliang Lu, Jake Metterville, Mitinda Samaraweera, Priyanka Shiva Prakasha, Hailin Yang, Yuan Yin, Hui Yu, Paloma H. Giangrande, Michael Byrne, Pachamuthu Kandasamy and Chandra Vargeese[*]

Wave Life Sciences, Cambridge, MA 02138, USA

Splicing

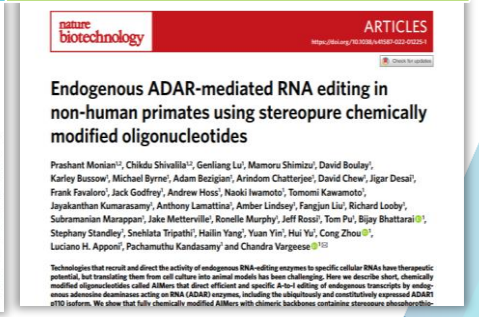
Control of backbone chemistry and chirality boost oligonucleotide splice switching activity

Pachamuthu Kandasamy[1,†], Graham McClorey[2,†], Mamoru Shimizu[1], Nayantara Kothari[1], Rowshon Alam[1], Naoki Iwamoto[1], Jayakanthan Kumarasamy[1], Gopal R. Bommineni[1], Adam Bezigian[1], Onanong Chivatakarn[1], David C. D. Butler[1], Michael Byrne[1], Katarzyna Chwalenia[2], Kay E. Davies[4], Jigar Desai[*,†], Juill Dilip Shelke[1], Ann F. Durbin[1], Ruth Ellerington[2], Ben Edwards[4], Jack Godfrey[1], Andrew Hoss[1], Fangjun Liu[1], Kenneth Longo[1,3], Genliang Lu[1], Subramanian Marappan[1], Jacopo Oieni[2], Ik-Hyeon Paik[1], Erin Purcell Estabrook[1], Chikdu Shivalila[1], Maeve Tischbein[1], Tomomi Kawamoto[1], Carlo Rinaldi[2,3], Joana Rajão-Saraiva[4], Snehlata Tripathi[1], Hailin Yang[1], Yuan Yin[1], Xiansi Zhao[1], Cong Zhou[1], Jason Zhang[1], Luciano Apponi[1], Matthew J. A. Wood[2,3,*] and Chandra Vargeese[*,†]

[1]Wave Life Sciences, Cambridge, MA, USA, [2]Department of Paediatrics, University of Oxford, South Parks Road, Oxford OX1 3QX, UK, [3]MDUK Oxford Neuromuscular Centre, University of Oxford, Oxford OX2 9DU, UK and [4]Department of Physiology, Anatomy and Genetics, University of Oxford, South Parks Road, Oxford OX1 3PT, UK

Received September 01, 2021; Revised December 18, 2021; Editorial Decision January 04, 2022; Accepted January 07, 2022

RNA Editing



nature biotechnology ARTICLES

Endogenous ADAR-mediated RNA editing in non-human primates using stereopure chemically modified oligonucleotides

Prashant Monian[12], Chikdu Shivalila[12], Genliang Lu[1], Mamoru Shimizu[1], David Boulay[1], Karley Bussow[1], Michael Byrne[1], Adam Bezigian[1], Arindom Chatterjee[1], David Chew[1], Jigar Desai[1], Frank Favaloro[1], Jack Godfrey[1], Andrew Hoss[1], Naoki Iwamoto[1], Tomomi Kawamoto[1], Jayakanthan Kumarasamy[1], Anthony Lamattina[1], Amber Lindsey[1], Fangjun Liu[1], Richard Looby[1], Subramanian Marappan[1], Jake Metterville[1], Ronelle Murphy[1], Jeff Rossi[1], Tom Pu[1], Bijay Bhattarai[1], Stephany Standley[1], Snehlata Tripathi[1], Hailin Yang[1], Yuan Yin[1], Hui Yu[1], Cong Zhou[1], Luciano H. Apponi[1], Pachamuthu Kandasamy[1] and Chandra Vargeese[1]

For additional information on this study, please contact clinicaltrials@wavelifesci.com or visit clinicaltrials.gov (NCT04906460)