Clinical Outcomes and Biomarker Findings From a Trial of Bezisterim in Subjects With Probable Alzheimer's Disease

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BACKGROUND

- **Bezisterim (formerly NE3107) is an investigative oral anti-inflammatory and insulin sensitizer being evaluated in both AD and PD[1]**
  - Bezisterim binds ERK and inhibits inflammation-specific ERK, NF-κB, and TNF signaling, but does not impact their homeostatic functions[1]
  - In obese animal models and subjects with IGT or T2D, bezisterim decreased pro-inflammatory mediators and insulin resistance[1]
  - Bezisterim improved motor activity and decreased neurodegeneration in a PD marmoset model[2] and improved MDS-UPDRS Part III scores in a Phase 2a PD study[3]
  - In an open-label 14-week Phase 2 study in MCI and mild AD, bezisterim improved neurological signs and symptoms, neuroimaging outcomes, and CSF/peripheral biomarkers; these findings were also correlated with changes in several biomarkers and neuroimaging analyses of change[4,5]
  - Bezisterim has a well-tolerated safety profile to date[3,4]
- **We evaluated the efficacy, safety, and tolerability of bezisterim in a larger sample with probable AD and over a longer duration (30 weeks)**

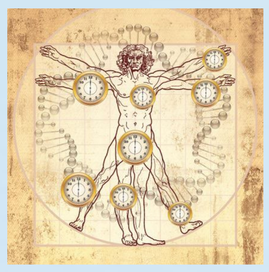
OBJECTIVES

Are processes of aging pharmacologically mutable?

- **Individuals appear to age at different rates**
  - Analysis of DNA methylation "clocks" can provide evidence of changes in gene expression related to aging (biological age)
  - The difference between biological age and chronological age (dAge) is a measure of age acceleration
  - A positive age acceleration (biological age greater than chronological age) is an independent predictor of impaired cognitive performance and earlier mortality[6,7]
- The primary goal of our study was to investigate the associations between metabolic inflammation, biological aging, and dementia in a human clinical investigation
- **The specific aim tested whether bezisterim could impact physiologic processes consistent with neurocognitive decline and diseases of aging**

METHODS

- NM101 (NCT04669028) was a Phase 3, randomized, placebo-controlled trial to assess the safety and efficacy of 20-mg oral bezisterim twice daily in subjects aged 60 to 85 years with probable AD
- Co-primary endpoints:
  - CDR-SB
  - ADAS-Cog12
- Secondary endpoints:
  - MMSE
  - ADCS-ADL
  - ADCS-CGIC
  - ADCOMS
- Exploratory endpoints:
  - The Epigenetic Clock Development Foundation analyzed available DNA samples using the Horvath SkinBlood epigenetic aging clock[8]
  - Hurdle.bio / Chronomics analyzed available DNA samples using PhenoAge,[9] Grim Age,[10] AgeHannum,[11] and InflammAge epigenetic aging clocks[12]



RESULTS

Patient disposition and data source

- The trial started during the COVID-19 pandemic and enrolled a total of 439 subjects through 39 sites
- We reported that upon trial completion, the Company found significant deviation from protocol and Good Clinical Practice (GCP) violations at 15 sites, causing the Company to exclude all subjects from these sites
- After exclusions for GCP violations, 57 subjects remained in the per-protocol population; those assigned to bezisterim were verified to have taken study drug from pharmacokinetics data, and 7 subjects randomized to placebo discontinued before day 150
- Baseline and completion data were available for 50 subjects (bezisterim, n=24 and placebo, n=26); and DNA methylation data were available for 33 of this cohort
- The cohorts were well-balanced overall (**Table 1**); of 31 metabolic, inflammatory, and dementia biomarkers, only RANTES was significantly different (placebo, 16.2 pg/mL; bezisterim, 23.1 pg/mL)

Table 1. Demographics and baseline characteristics

Characteristic	Placebo (n=26)	Bezisterim (n=24)
Mean age, years	75.3	75.7
Female, n (%)	12 (46)	15 (63)
Caucasian, n (%)	22 (92)	22 (92)
Mean CDR-SB	6.92	6.58
Mean ADAS-Cog12	33.3	31.0
Mean waist-hip ratio	0.939	0.902
Mean weight, pounds	174	159
Mean SBP/DBP, mm Hg	133/76	134/79
Mean total cholesterol, mg/dL	182	189
Mean glucose, mg/dL	94.7	98.8
HOMA2 IR	1.10	1.34

Safety findings (Per-Protocol population, n=57)

- TEAEs occurred in 62.5% (n=15) of patients in the bezisterim group and 72.7% (n=24) of patients in the Placebo group
- Bezisterim TEAEs ≥5% and > Placebo: Headache (12.5%; n=3) vs (0%; n=0)
- Treatment-related TEAEs occurred in 12.5% (n=3) of patients in the bezisterim group and 18.2% (n=6) of patients in the placebo group
- SAEs occurred in 4 patients (bezisterim, n=1; placebo, n=3); none were treatment-related
- There was 1 non-treatment-related death in the bezisterim group; the patient was a 70-year-old male who died of a respiratory arrest
- 3 patients in the placebo group and none in the bezisterim group discontinued due to an AE

Efficacy findings (primary and secondary endpoints)

- Week 30 data suggest bezisterim improved neurological assessments vs placebo (**Figure 1**) and was comparable to results reported from clinical trials of approved medications (**Table 2**)

Figure 1. Bezisterim showed improvements over time in primary and secondary endpoints

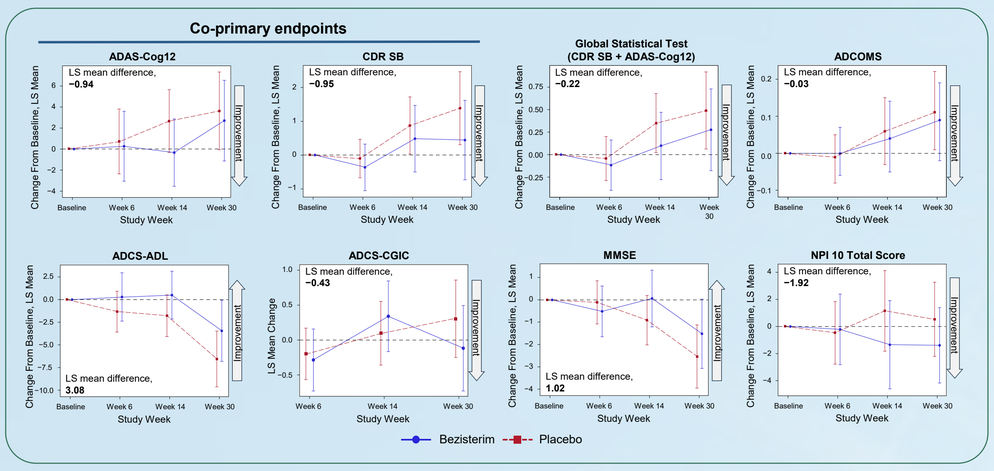


Table 2. Bezisterim vs placebo change from baseline

	Bezisterim vs placebo	Published data at 1.5 years[a]
Co-Primary		
CDR-SB (Lower=improvement)	−0.95 (68%)	−0.45 (27%)[7] −0.39 (22%)[8]
ADAS-Cog12 (Lower=improvement)	−0.94 (26%)	−1.44 (25%)[7] −1.40 (27%)[8]
Secondary		
MMSE (Higher=improvement)	+1.02 (40%)	+0.6 (18%)[8]
ADCS-ADL (Higher=improvement)	+3.08 (47%)	+2.0 (36%)[7]
ADCS-CGIC (Lower=improvement)	−0.43 (139%)	
ADCOMS (Lower=improvement)	−0.03 (27%)	−0.05 (23%)[8]

[a]Other published data at 18 months data for lecanemab[13] and aducanumab.[14]

Exploratory endpoints

- Bezisterim significantly modified clinical measure correlations with biomarkers (**Figure 2**)
- Bezisterim modified age acceleration (**Figure 3**), which was associated with change in monocyte phenotype (**Figure 4**)
- Decreased age acceleration correlated with improvements in CDR-SB, ADAS-Cog12, Global Statistical Test, MMSE, ADCOMS, and CGIC; there were no correlations with any neurological assessments in the placebo group
- Principal Component Analysis (PCA) was then carried out to reduce data for the age-acceleration correlations into lower dimensions or Principal Components (PCs) (**Figure 5**)

Figure 2. Bezisterim modified interdependence of neurological assessments[b]

Placebo was correlated with worsening biomarkers/measures

Dementia biomarkers
↑GFAP with ↑GST, ↓MMSE, ↑CDR-SB, ↑ADCOMS, ↑Cog12
↑pTau217 with ↑GST, ↑ADCOMS, ↑Cog12, ↑CGIC

Metabolic measures
↑Blood Pressure with ↑GST, ↑CDR-SB, ↑ADCOMS
↑Glucose with ↑CDR-SB
↑Beta cell insulin production with ↓ADL

Inflammatory measures
↑CRP with ↑CDR-SB, ↓ADL
↓C1q with ↑CGIC

Bezisterim yielded inverse correlations with improvements vs placebo

- ↓Fructosamine with ↑ADL (placebo: ↓Fructosamine with ↓ADL) p=0.003[a]
- ↑RANTES with ↓CDR-SB (placebo: ↑RANTES with ↑CDR-SB) p=0.003[a]

Bezisterim correlated with additional improvements

- ↓NfL with ↓GST, ↑MMSE, ↓CDR-SB, ↓Cog12, ↓ADCOMS, ↑ADL
- ↓Cholesterol with ↓GST, ↑MMSE, ↓CDR-SB, ↓Cog12
- ↑RANTES with ↓GST, ↑MMSE, ↓ADCOMS, ↓Cog12

For neurological assessments: red = decline; green = improvement

[b]Z test statistic of Fisher's to Z transformation.

Figure 3. Bezisterim modified dAge for 5 aging clocks

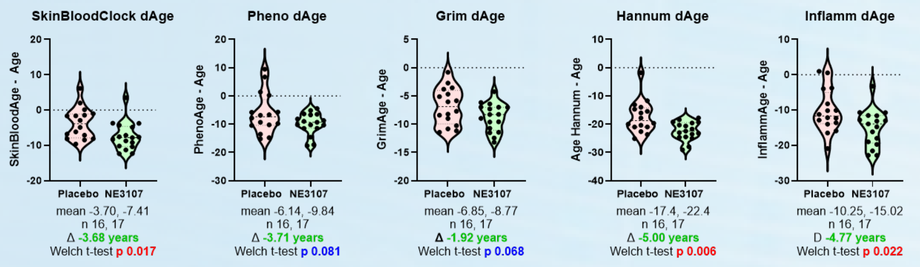
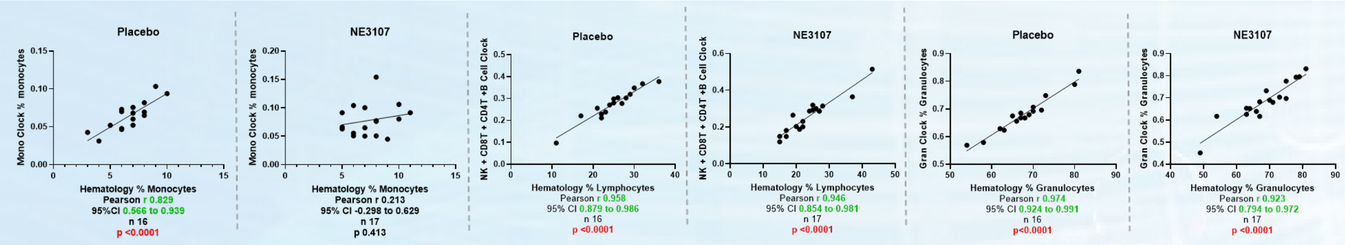


Figure 4. Bezisterim modification of monocyte phenotype

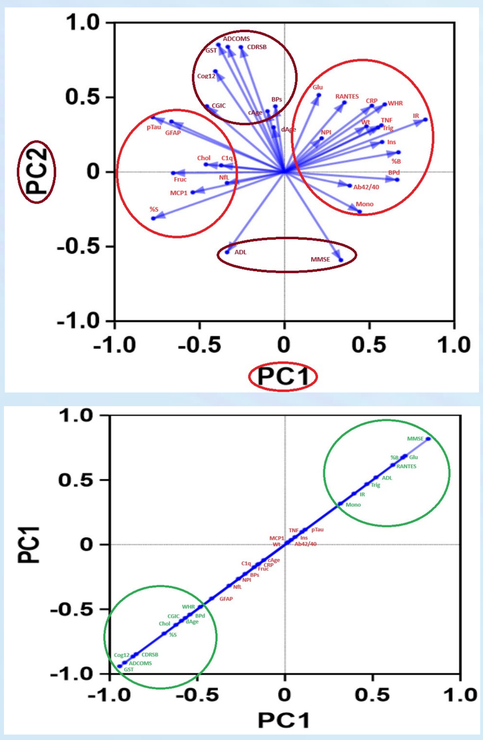


Cell type	Placebo		Bezisterim			
	Pearson r	P	Pearson r	P	Z'	P
Monocytes	0.829	**<0.0001**	0.213	0.413	2.51	**0.012**
Lymphocytes	0.958	**<0.0001**	0.946	**<0.0001**	0.334	0.738
Granulocytes	0.974	**<0.0001**	0.923	**<0.0001**	1.44	0.149

Correlations of cell type clocks and hematology results (as assessed by Z test statistic of Fisher's to Z transformation):

- Placebo: hematology % monocytes is correlated with monocyte internal molecular clock
- Bezisterim: hematology % monocytes is **not correlated** with monocyte internal molecular clock
- **Bezisterim's impact on dAge may be explained by modification of monocyte DNA methylome, changing from a pro-inflammatory to an anti-inflammatory state (M1→M2 transition hypothesis)**

Figure 5. Principal components associated with placebo and bezisterim differed



Placebo dAge PC analysis

- PC1 included metabolic, inflammation, and dementia biomarkers
- PC2 included neurological assessments, and chronological age; dAge was only weakly associated with outcomes
- **Divergent PC1 vs PC2 suggests systems dysregulation in placebo-treated patients**

Bezisterim dAge PC analysis

- Only PC1 was identified, which included metabolism, innate immunity, and dAge; inflammation and dementia biomarkers were excluded
- All 31 measures in PC1 significantly correlated with dAge in PC regression
- **Single PC suggests systems re-regulation in bezisterim-treated patients**

CONCLUSIONS

- In this small per-protocol sample compared to placebo, bezisterim appeared to:
  - Improve neurological assessments
  - Decrease biological age
  - Realign biological aging with neurological assessments
- Principal Component Analysis and Principal Component Regression correlations were consistent with the hypothesis that bezisterim, by decreasing TNF- and MCP1-stimulated NF-κB and neuroinflammation, might promote a transition of microglia from inflammatory and destructive to anti-inflammatory, phagocytic, and restorative cells
- DNA methylation aging clocks may prove to be valuable biomarkers for neurodegeneration
- These data suggest bezisterim may improve probable AD via pathways related to inflammation and warrant confirmation

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DISCLOSURES

CA, **CLR**, **JD**, **JP**, **JY**, and **LW** are employees of BioVie Inc.

JZ is a consultant for BioVie Inc.

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DCS and **HY** have nothing to disclose.

AD, Alzheimer's disease; ADAS-Cog12, 12-subunit Cognitive Subscale of the Alzheimer's Disease Assessment Scale; ADCOMS, Alzheimer's Disease Composite Score; ADCS-ADL, Alzheimer's Disease Cooperative Study-Activities of Daily Living; ADCS-CGIC, Alzheimer's Disease Cooperative Study-Clinical Global Impression of Change; AE, adverse events; CDR-SB, Clinical Dementia Rating-Sum of Boxes; COVID-19, coronavirus 2019; CSF, cerebrospinal fluid; ERK, extracellular signal-regulated kinases; GST, glutathione S-transferase; IGT, impaired glucose tolerance; ITT, intent-to-treat; MCI, mild cognitive impairment; LS, least squares; MCP1, monocyte chemoattractant protein-1; MDS-UPDRS, Movement Disorder Society-Unified Parkinson's Disease Rating Scale; MMSE, Mini-Mental State Examination; NF-κB, nuclear factor kappa-light-chain-enhancer of activated B cells; PD, Parkinson's disease; RANTES, Regulated upon Activation, Normal T Cell Expressed and Presumably Secreted; SAE, serious adverse event; T2D, type 2 diabetes; TEAE, treatment-emergent adverse event; TNF-α, tumor necrosis factor alpha.

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