Cognitive Vitality Reports® are reports written by neuroscientists at the Alzheimer's Drug Discovery Foundation (ADDF). These scientific reports include analysis of drugs, drugs-in-development, drug targets, supplements, nutraceuticals, food/drink, non-pharmacologic interventions, and risk factors. Neuroscientists evaluate the potential benefit (or harm) for brain health, as well as for age-related health concerns that can affect brain health (e.g., cardiovascular diseases, cancers, diabetes/metabolic syndrome). In addition, these reports include evaluation of safety data, from clinical trials if available, and from preclinical models.

Buntanetap

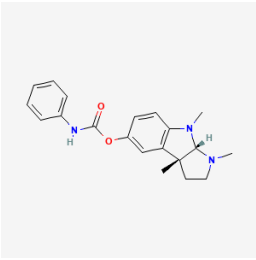
Evidence Summary

Buntanetap suppresses production of proteins like APP and αSyn. In small clinical trials, buntanetap was well tolerated. It may have clinical efficacy on biomarkers and cognition.

Neuroprotective Benefit: Small clinical trials have found improved measures of cognition and trends towards reduced levels of neurotoxic proteins. However, the trials were small and results variable; ongoing larger trials will hopefully be illuminating.

Aging and related health concerns: Buntanetap has not been investigated for non-neuronal aging-related health concerns. There is some preclinical indication for potential benefit in stroke.

Safety: Buntanetap has been tested in four studies and appears to be well tolerated based on the data currently available.

Availability: in clinical development	**Dose**: Doses ranging from 5 to 240 mg total daily have been tested. Buntanetap is taken orally.	**Chemical formula:** $C_{20}H_{23}N_3O_2$ **MW**: 337.4 Source: PubChem 
Half-life: 3.7 – 5.5 hours	**BBB**: Penetrant	
Clinical trials: Buntanetap has been tested in trials with 199 total patients. Ongoing trials plan to enroll a total of 770 patients.	**Observational studies**: Buntanetap has not been evaluated in observational studies.	

What is it?

Proteins are made by first transcribing the relevant gene regions into mRNA, and then ribosomes put together the protein product by translating the mRNA. Cells have a variety of mechanisms to control protein production, including at this translation stage. One mechanism of translational control is having a particular sequence or sequences in the mRNA molecule itself that can be bound by specific protein in a specific state. An example of this mechanism is the iron-responsive element (IRE) which can be bound by iron-regulatory proteins (IRP) 1 or 2, depending on the presence or absence of iron. The binding of IRP1 or IRP2 can then promote or prevent the translation of that mRNA into protein depending on where in the mRNA sequence the IRE is located. Modulating the binding of IRP1 and 2 to mRNA transcripts can therefore modulate the levels of certain proteins present based on the environment in the cell, such as the cellular concentration of iron (Zhang et al., 2014).

As discussed in Chen et al., 2021, there are a variety of mRNA transcripts that code for proteins involved in neurodegenerative diseases, including APP (the precursor to Aβ), alpha-synuclein (αSyn), and huntingtin, that have atypical IRE sequences and can be bound and regulated by IRP1. It is thought that in conditions with low cellular iron concentrations, IRP1 can bind to the IRE in the mRNAs that code for proteins like APP and αSyn and suppress APP and αSyn protein production. In cellular conditions with higher iron levels, IRP1 does not bind to these IREs, and there is increased expression of these proteins.

Higher brain iron levels or other alterations to brain iron homeostasis may be associated with dementia, and treatment with iron chelators may have some clinical benefit; iron driving expression of some of these neurotoxic proteins is a possible mechanism of action (Ayton et al., 2015; Zhang et al., 2022).

Buntanetap, also known as posiphen or ANVS401, is an orally available small molecule. It is closely related to phenserine, which has also been investigated for use in dementia; as buntanetap does not have anticholinesterase activity like phenserine does, buntanetap appears to have more permissive dosing. Phenserine and buntanetap both reduce protein levels of APP and Aβ in cell and animal models (Lahiri et al., 2007, Winblad et al., 2016). It is thought that buntanetap achieves this protein reduction through interacting with the IRE/IRP1 complex of neurotoxic protein transcripts such that protein translation is suppressed. Buntanetap has been shown to reduce levels of Aβ, αSyn, huntingtin, SOD1, and TDP-43 in different systems (Chen et al., 2021).

Neuroprotective Benefit: Small clinical trials have found improved measures of cognition and trends towards reduced levels of neurotoxic proteins. However, the trials were small and results variable; ongoing larger trials will hopefully be illuminating.

Types of evidence:

- 3 clinical trials
- 4 reviews
- 12 laboratory studies

Human research to suggest prevention of dementia, prevention of decline, or improved cognitive function:

Maccicchini et al., 2012 reported on the results from three studies, including an open label proof of mechanism study in patients with mild cognitive impairment. In the proof of mechanism study, they assessed some preliminary biomarker effects after 60 mg doses of buntanetap four times daily for 10 days. The researchers found that the CSF levels of APP and tau decreased to levels of health volunteers after the 10 day treatment, as compared to pre-drug treatment baseline. See "Safety" section for further results from these three studies.

Human research to suggest benefits to patients with dementia:

After the results of the initials trials detailed by Maccicchini et al., 2012, the group proceeded to a larger randomized double-blinded trial in patients with AD and patients with PD, which they described in Fang et al., 2022. According to their baseline characteristics, they randomized 74 total patients: 16 with early AD and 58 with early PD. Of these patients, 50 patients with PD received buntanetap doses of 5, 10, 20, 40, or 80 mg daily for 25±2 days; 10 patients with AD were given 80 mg buntanetap daily for 25±2 days; and 10 patients completed placebo dosing. Their primary endpoint was safety and tolerability; their

secondary endpoint was pharmacokinetics of buntanetap in plasma. Their exploratory endpoints were CSF biomarkers and assessments of cognitive function.

They evaluated cognition in patients with AD using ADAS-Cog1I and WAIS coding test, which measures visual-motor dexterity, associative non-verbal learning, and nonverbal short-term memory. The measures were completed at baseline and after treatment. There was a statistically significant improvement on ADAS-Cog11 compared to baseline, with placebo group improving by 1.1±2.63 points and the treatment group improving by 4.40±2.04 points, for a difference of 3.3±3.32 points. In the buntanetap treatment group, there was also a statistically significant improvement of 6.6±3.04 points on WAIS coding as compared to baseline. There were trends towards improvement on MMSE and CDR Sum of Boxes in the treatment group, but these trends did not reach significance. There appeared to be a lot of variability in patient performance in all groups.

The authors also found statistically significant improvements on the Movement Disorder Society-Unified Parkinson's Disease Rating Scale (MDS-UPDRS) in the treatment group compared to baseline. This scale has different subsections, measuring components such as motor performance and activities of daily living. They found a statistically significant improvement from baseline in motor function in the 10 mg dose group, and a statistically significant improvement from baseline on total score in the 10 mg and 20 mg group. They also found statistically significant improvements on WAIS coding from 5 mg compared to baseline and to placebo, and at higher doses as well.

They additionally examined several CSF biomarkers at the end of the 25±2-day treatment period in patients taking 80 mg daily of buntanetap or placebo. For this analysis they set the biomarker levels as percents of the patient's own baseline level. The study was not powered to detect changes in these biomarkers, but they did see trends towards less increase in total tau, phosphorylated tau, and APP in AD patients as compared to placebo, and in αSyn in PD patients as compared to placebo. They saw trends towards reduction in inflammatory markers in both AD and PD patients, with PD patients showing greater reduction trends as compared to placebo. They also found trends towards less increase in axonal and synaptic markers in both AD and PD patients as compared to placebo.

Mechanisms of action for neuroprotection identified from laboratory and clinical research:

As shown in Chen et al., 2021, buntanetap suppresses translation of proteins with an atypical IRE in their 5' untranslated region (UTR) of their mRNA. Huntingtin, APP, TDP-43, and αSyn all have this atypical IRE present in their coding mRNA, and the levels of these proteins are decreased upon buntanetap administration in cell culture models. It is thought that buntanetap binds to the IRE/IRP1 complex and prevents the ribosome from translating the mRNA transcript. Higher brain iron levels or other alterations to brain iron homeostasis have been associated with dementia, and treatment with iron chelators may have some clinical benefit in dementia patients; buntanetap theoretically could counteract some of the iron homeostasis by suppressing protein translation (Zhou & Tan 2017).

Lowering the expression of these proteins has a variety of potential effects, ranging from restoration of impaired axonal transport and endosomal function (Fang et al., 2022). Buntanetap may also have anti-inflammatory actions, such as suppressing the generation of a pro-inflammatory cytokine IL-1β, based on cell culture models (Yu et al., 2017). Some preclinical work in cell and an animal model indicated that buntanetap may promote neuronal proliferation, neurogenesis, and differentiation (Marutle et al., 2007; Lilja et al., 2013). In a mouse model of AD, buntanetap administration improved synaptic dysfunction as measured by LTP, and mitigated spatial memory and fear learning deficits. Administration of buntanetap to wild-type mice in the same experiments did not have any reported effects (Teich et al., 2018).

One potential caveat of buntanetap is that reducing protein levels of certain neurodegenerative proteins may have unintended consequences. While too much of these proteins is clearly detrimental for neuronal health, there is some evidence that too little of these proteins could also be deleterious, at least in certain diseases.

For instance, some preclinical experiments that reduced expression of αSyn in the developed brain led to neurodegeneration (Benskey et al., 2018). And, while patients with higher levels of αSyn are more likely to develop PD, as evidenced by gene duplications of αSyn causing PD, one study found that PD patients with gene variants associated with higher expression of αSyn progress more slowly towards severe motor and cognitive impairment (Markopoulou et al., 2014). It is therefore possible that there is a 'Goldilocks' level of protein, and tipping too far in either direction can be detrimental to neuronal health. This possibility, particularly in Parkinson's patients, should be kept in mind as these trials progress.

APOE4 interactions:

No preclinical or clinical studies have assessed any differential impact of buntanetap based on APOE4 status.

There are some studies that hint at interactions between APOE allele status and iron homeostasis (Zhang et al., 2022). As buntanetap acts through the iron responsive element and iron regulatory protein, it is theoretically possible there could be a different effect in APOE4 carriers. However, this is speculation based on potential mechanisms in common.

Aging and related health concerns: Buntanetap has not been investigated for non-neuronal aging-related health concerns. There is some preclinical indication for potential benefit in stroke.

Types of evidence:

- 2 laboratory studies

Buntanetap has not been explored for non-neuronal aging or related health concerns in humans.

STROKE: POTENTIAL FOR BENEFIT BASED ON PRECLINICAL WORK

Buntanetap has been assessed for use in stroke in preclinical papers. One group found that buntanetap was neuroprotective against glutamate insult in primary neuron culture models, and post-stroke treatment with buntanetap in a rat model of stroke reduced measures of ER stress, microglia immunoreactivity, cell death, infarction volume, and neurological deficits as compared to vehicle treatment (Yu et al., 2020).

Treatment with buntanetap and another experimental stroke treatment, PFA-α, also enhanced proliferation and neuronal differentiation of progenitor cells in neural stem cell models and improved survival of neural progenitor cells in a mouse model of stroke. Treatment of PFA-α alone improved motor performance in mouse models of stroke as well as combined therapy of PFA-α and buntanetap, but combined treatment mitigated cognitive impairment more than individual treatment of PFA-α (Turcato et al., 2018).

Safety: Buntanetap has been tested in four studies and appears to be well tolerated based on the data currently available.

Types of evidence:

- 3 clinical trials

Macchicchini et al., 2012 reported on the results from three studies: a single ascending dose study in healthy volunteers, a multiple ascending dose in healthy volunteers, and a proof of mechanism study in patients with mild cognitive impairment.

In the single ascending dose study, the researchers enrolled 72 healthy volunteers and planned to administer single ascending doses of 10, 20, 40, 80, 160, or 240 mg of buntanetap or placebo. Due to nausea and vomiting at the 160 mg dose, they did not administer the 240 mg dose. In the multiple ascending dose study, the authors enrolled 48 healthy volunteers and administered 20, 40, or 60 mg of buntanetap or placebo four times daily for 7 or 10 days. For both of these studies, researchers collected

vital signs, ECGs, clinical laboratory tests, adverse event information, and blood and plasma for pharmacokinetics. Both of these studies were randomized and double-blinded.

In the proof of mechanism study, they enrolled 5 patients with MCI. This study was open label. Patients were given 60 mg doses of buntanetap four times daily for 10 days. CSF plasma was analyzed for pharmacokinetics of buntanetap and its metabolites, and also for dementia-related proteins including APP, Aβ, tau, and markers of inflammation.

Side effects included dizziness, nausea and vomiting, headache, and orthostatic hypotension. The side effects were not statistically significantly different than placebo except for dizziness and nausea and vomiting at the single dose of 160 mg. 80 mg single doses and 60 mg four times daily doses were determined as the no observed adverse effects doses.

In a follow-up Phase 2/3 study detailed in Fang et al., 2022, the researchers randomized 74 total patients; 16 with early AD and 58 with early PD. Of these patients, 50 patients with PD received buntanetap doses of 5, 10, 20, 40, or 80 mg daily for 25±2 days; 10 patients with AD were given 80 mg buntanetap daily for 25±2 days; and 10 patients completed placebo dosing. There were no serious adverse events in any group; all but one adverse events were Grade 1 or Grade 2. Most adverse events were related to the lumbar puncture study procedure. There was 1 Grade 1 QT prolongation in a patient receiving buntanetap which resolved and was considered clinically not significant. There was also one patient who received buntanetap and had elevated liver function tests. These resolved and were considered not related to study drug.

Treatment related or possibly related adverse events were headache, erythema, muscle spasms, and movement disorder, and all were mild.

Drug interactions:

Drug interactions of buntanetap are not yet fully clear. Drugbank.ca lists approximately 200 interactions or potential interactions, including with acetylcholine or drugs that affect the cholinergic system and certain drugs that can cause bradycardia as a side effect.

Research underway:

There are two currently ongoing studies investigating the efficacy of buntanetap. Both of these studies are in patients with early or mild to moderate dementia.

NCT05686044 is a Phase 2/3 trial of buntanetap in patients with MCI. The study plans to randomize 320 participants to placebo or one of three doses of buntanetap: 7.5 mg, 15 mg, or 30 mg daily. The participants will be on placebo or study drug for 12 weeks. Assessments will be performed at baseline,

week 6, and week 12. The primary outcome measure is cognitive function as assessed by ADAS-Cog 11 and the Alzheimer's Disease Cooperative Study-Clinical Global Impression of Change (ADCS-CGIC). Secondary outcome measures include other measures of function, including the MMSE and ADL scale. One interim analysis to re-assess sample size is planned for when 90 participants have completed 6 weeks of dosing. This study enrolled its first patient on April 1, 2023. The researchers hope to complete the trial in early 2024.

NCT05357989 is a Phase 3 study of buntanetap in patients with early PD. This study plans to enroll 450 participants and randomize them to placebo or one of two doses of buntanetap: 10 mg or 20 mg daily. The participants will be on study drug for 6 months, with assessments taking place at baseline, 1 month, 2 months, 3 months, and 6 months. The primary outcome measures are safety and tolerability and motor and daily living parts of the MDS-Unified Parkinson's Disease Rating Scale (UPDRS). This scale has subsections to assess different components of PD progression. Secondary outcomes include other measures of PD progression, including scores from other sections of the UPDRS and Clinical Global Impression of Severity (CGIS). This study enrolled its first patient in summer 2022 and the study investigators hope to complete the study at the end of 2023.

Search terms:
Pubmed, Google: buntanetap, posiphen

- Dementia, Alzheimer's, drug interactions

Websites visited for buntanetap / posiphen:

- Clinicaltrials.gov
- PubChem
- DrugBank.ca
- Cafepharma