



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.

Ligustilide

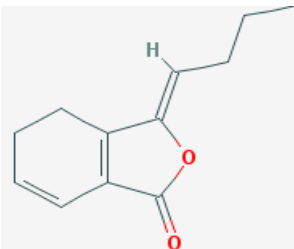
Evidence Summary

Protects against inflammatory and oxidative stress damage through induction of Klotho and Nrf2, but has poor stability and bioavailability. Novel formulations will be needed for it to be clinically viable.

Neuroprotective Benefit: Protects against inflammatory and oxidative stress induced cognitive impairment in preclinical studies, but suffers from extremely poor stability and bioavailability in its current form.

Aging and related health concerns: Protects against inflammatory damage and vascular dysfunction in preclinical studies, but novel bioavailable formulations will be necessary to offer clinically meaningful benefits.

Safety: No major adverse events reported in animals, but pure forms of ligustilide have not been tested in humans.

Availability: In traditional Chinese medicine (Dong quai)	Dose: Wide range used, no clinically beneficial dose established	Chemical formula: C₁₂H₁₄O₂ MW: 190.242 g/mol  Source: Pubchem
Half-life: 3-5 hours (in rats)	BBB: Penetrant	
Clinical trials: Dong quai has been tested in 2 RCTS for treatment of menopausal symptoms, but no trials have been conducted with pure ligustilide.	Observational studies: No human studies for pure ligustilide.	

What is it? Ligustilide (Z-ligustilide) is a phthalide that is contained in *Angelica* subspecies. It is the primary active ingredient in the root of *Angelica sinensis*, also known as Dong quai, which is an herb used in traditional Chinese medicine. It is commonly used for cardiovascular health, relief from menstrual pain and menopausal symptoms, such as hot flashes. Ligustilide has anti-inflammatory and antioxidant activity through the induction of Klotho and the Nrf2 antioxidant system.

Neuroprotective Benefit: Protects against inflammatory and oxidative stress induced cognitive impairment in preclinical studies, but suffers from extremely poor stability and bioavailability in its current form.

Types of evidence:

- Several laboratory studies

Human research to suggest prevention of dementia, prevention of decline, or improved cognitive function? None

Human research to suggest benefits to patients with dementia: None



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

While there have been no human studies examining the efficacy of ligustilide for dementia prevention, rodent studies have provided evidence of its neuroprotective capabilities. Furthermore, its projected mechanism of action involves induction of two of the body's most potent defenses against oxidative stress and inflammatory damage: Klotho and the Nrf2 antioxidant system. Due to its **poor stability and bioavailability profile** [1; 2], novel formulations of ligustilide would need to be developed in order for it to be a viable option for testing in clinical trials.

Alzheimer's disease: Potential benefit (rodents)

In the APP/PS1 transgenic Alzheimer's disease (AD) mouse model, ligustilide was able to reduce the progression of cognitive impairment and AD related pathology through induction of Klotho and the reduction of oxidative stress.

Ligustilide treatment (40 mg/kg daily for 15 days) attenuated neuronal loss and A β -associated pathology (accumulations of A β , APP, p-Tau) in rats following intracerebral injection of A β_{25-35} [3]. The treated rats also had better performance on a spatial memory task (Morris water maze) than untreated animals. Nine month-old AD mice treated with ligustilide (40 mg/kg daily intragastric gavage) for 8 weeks showed improvements in spatial memory on the Morris water maze test, including shortened escape latency, and increased time in platform quadrant [4]. This was accompanied by decreased levels of senile plaques, more structurally intact mitochondria, enhanced mitochondrial respiratory capacity, decreased oxidative stress (ROS, MDA), and less synaptic loss in the hippocampus. Similar cognitive and neuronal protecting benefits were seen with ligustilide treatment (30 mg/kg daily for 14 weeks) in 12-month-old AD mice. In these animals, the neuroprotection was attributed to an increase in Klotho [5]. Ligustilide may act as an enhancer of the protease ADAM10, which promotes the alpha-cleavage and processing of APP, and cleavage of transmembrane Klotho to increase the level of soluble-Klotho. Ligustilide also promotes the expression and secretion of Klotho in the choroid plexus.

Cognitive aging: Potential benefit (rodents)

The Klotho induction and antioxidant activities of ligustilide may help protect against aging-associated cognitive decline.

In 10-month-old senescence-accelerated (SAMP8) mice, ligustilide treatment (40 mg/kg daily for 8 weeks) reduced A β_{1-42} accumulation, p-Tau levels, and neuronal loss, and improved performance on short term (Y maze) and long-term (passive avoidance) memory tasks compared to vehicle [3]. The reduction of AD pathology was correlated with the upregulation of Klotho expression in the choroid



plexus. Klotho led to the induction of the transcriptional FOXO oxidative stress resistance system by regulating the phosphorylation of FoxO1 and Akt. The antioxidant induction capacity of ligustilide was also able to attenuate accelerated-aging/cognitive impairment induced by reactive oxygen species (ROS) generating d-galactose [6].

Stroke: Potential benefit (rodents)

Ligustilide provides neuroprotection in ischemic rodent stroke models (cerebral artery occlusion) by **reduction of inflammation and oxidative stress**. However, ligustilide was more effective at reducing ischemia-related pathology than preventing neurological impairments. The beneficial effects appear to be mediated by the **induction of CNS Klotho and the Nrf2 antioxidant system**.

Ligustilide treatment has been shown to prevent neuronal loss (similar to sham controls)[7; 8; 9; 10; 11; 12], attenuate astroglial and microglial activation [12; 13; 14], greatly reduce induction of pro-inflammatory mediators (NF- κ B, TNF- α , IL-6, IL-1 β , TLR4) [8; 12; 14], increase anti-inflammatory mediators (IL-10) [12], and reduce levels of oxidative stress (decreased MDA, MPO, and increased SOD) [12; 13]. Treatment also reduced the permeability of the blood brain barrier (BBB) by regulating expression of tight-junction adhesion proteins (occludin, MMP-2, MMP-9, ICAM-1), leading to less brain edema [9; 10; 12; 15]. Ligustilide (32 or 40 mg/kg) administered 2 hours after ischemic injury, reduced infarct volumes to approximately 40% of the infarct size in untreated animals [11; 12], and had similar efficacy as the antioxidant edavarone [11].

Ligustilide promotes activation (nuclear translocation) of Nrf2 and induction of its antioxidant pathway, including expression of the target gene HO-1 [11]. It also reduces oxidative stress damage through activation of Klotho, via modulation of the FOXO transcription factors (by inhibiting phosphorylation of FoxO1 and Akt)[8] and the peroxiredoxin redox system (by reducing secretion of Prx6) [12]. Prx6 promotes the TLR4 signaling pathway and associated microglial activation. In a stroke model (MCAO), ligustilide (40 mg/kg) reduced Prx6 levels to 2.3 \pm 1.2 times the level in sham animals, while Prx6 levels increased to 11.7 \pm 3.1 times higher than sham in untreated rats following stroke [12].

Despite, the highly neuroprotective effects which reduced neuronal pathology toward levels seen in sham controls, ligustilide treatment was only partially protective in preventing neurological deficits. Neurological deficit scores were reduced up to half relative to untreated animals with MCAO, and ligustilide treated animals had better performance on the Morris water maze, however, they still had significant neurological impairments relative to sham controls [7; 9; 11; 12; 13; 14].



Ligustilide has also been tested for its ability to improve the therapeutic efficacy of adipose-derived stem cells in the MCAO stroke model. Pre-treatment of the stem cells with ligustilide did not enhance their ability to promote recovery [16]. Additionally, high doses of ligustilide (>10 µg/mL) interfered with the proliferation and viability of the stem cells in culture.

APOE4 interactions: Unknown

Aging and related health concerns: Protects against inflammatory damage and vascular dysfunction in preclinical studies, but novel bioavailable formulations will be necessary to offer clinically meaningful benefits.

Types of evidence:

- 2 clinical trials (for ligustilide containing traditional Chinese medicine for menopause)
- Numerous laboratory studies

Inflammation: Potential benefit (rodents)

Ligustilide treatment has been shown to exhibit anti-inflammatory activity in rodent models.

Pretreatment of ligustilide (i.v.) prior to lipopolysaccharide (LPS), attenuated intrathecal LPS-mediated inflammatory pain by **reducing the induction of pro-inflammatory mediators** (TNF-α, IL-1β, and IL-6) and microglial activation in the spinal cord [17]. Spinal pain induced by injection of Freund's adjuvant was also alleviated by treatment with ligustilide (60 mg/kg i.v. for 4 days) starting 1 day after injection [18]. The pain relief was mediated by inhibition of TLR4 activation and signaling, likely due to Klotho induction. Pretreatment with *Angelica sinensis* extract (active ingredients are ligustilide and ferulic acid) was also able to inhibit TNF-α and protect mice against LPS-induced endotoxic shock [19]. Treatment increased survival from 18% to 44%. Ligustilide (40 mg/kg daily for 8 weeks) was also beneficial in reducing cartilage damage and inflammation (TNF-α, IL-6) in a mouse model of osteoarthritis [20].

A nanoemulsion formulation of ligustilide given orally at 20 mg/kg was found to significantly improve the absorption rate (C_{max} and AUC), and was superior to untreated ligustilide in reducing inflammation (TNF-α, IL-1β, VEGF, IL-17) in a mouse model of endotoxin-induced eye inflammation (uveitis) [21]. Most notably, this formulation was found to be stable at room temperature for at least 90 days, whereas untreated ligustilide decomposes rapidly (oxidizes) at high temperature [2].



Vascular dysfunction: Potential benefit (rodents)

Ligustilide **inhibits inflammation and promotes vasodilation** through Nrf2 and regulation of voltage-gated calcium channels. In human umbilical vein endothelial cells, ligustilide suppressed monocyte activation, NF- κ B activation, and ROS production in response to inflammatory stimuli (TNF- α) [22]. It also promoted the activation (nuclear translocation) of Nrf2 and nitric oxide (NO) production. Ligustilide was able to dose-dependently relax pre-constricted rat arteries by inhibiting receptor-mediated calcium influx and release [23].

Cancer: Potential benefit (rodents)

Ligustilide was shown to have anti-tumorigenic properties in cell culture, and possibly utility in helping overcome chemoresistance. In an ovarian cancer cell line, ligustilide promoted oxidative-stress mediated apoptosis [24], and helped to re-sensitize tamoxifen-resistant breast cancer cells by inhibiting autophagy [25]. In a mouse xenograft model, mice treated with *Angelica sinensis* extract (500 mg/kg s.c.) had reduced glioblastoma tumor size (at day 38) ($155.1 \pm 56.4 \text{ mm}^3$) compared to controls ($849.9 \pm 150.1 \text{ mm}^3$, $p < 0.05$) [26].

Menopausal symptoms: No benefit

Angelica sinensis (Dong quai) has long been used to provide relief from post-menopausal symptoms because it was believed to have estrogenic effects, however, clinical trials demonstrated that it is no more effective than placebo and does not have estrogen-like effects.

In a Phase 2 non-placebo-controlled RCT of 60 menopausal women treatment with Dang Gui Buxue Tang (composed of *Astragalus membranaceus* and *Angelica sinensis*) for 12 weeks improved psychosocial scores on the Menopause Quality of Life scale ($P = 0.013$) relative to baseline, but had no effects on estrogen or luteinizing hormones, or blood lipid content [27]. A separate 24-week placebo-controlled trial of 71 post-menopausal women found that Dong quai (*Angelica sinensis*) treatment was not better than placebo in reducing hot flashes and did not produce any estrogen-like effects on endometrial thickness or vaginal maturation [28].

Safety: No major adverse events reported in animals, but pure forms of ligustilide have not been tested in humans.

Types of evidence:

- 2 clinical trials (for ligustilide containing traditional Chinese medicine for menopause)
- Numerous laboratory studies

Since pure ligustilide has not yet been tested in humans, its safety profile is not known. Dong quai is rated as possibly safe for occasional use by [WebMD](#), but may increase sensitivity to the sun, and there is not enough information to determine whether long-term chronic use is safe. In clinical trials, women taking Dong quai with warfarin were at an increased risk for bleeding [27], and it should not be taken in conjunction with other blood thinners. According to [Drugs.com](#), Dong quai has moderate interaction with 77 drugs, which are primarily anticoagulants.

Sources and dosing:

Ligustilide can be found in a variety of traditional Chinese medicines, Dong quai (*Angelica sinensis*), Dang gui buxue tang, Danzhi xiaoyao san, and Rhizoma chuanxiong. [Doses](#) range widely (up to 15g/day), but **no clinically beneficial dose has been established**. None of these preparations have been rated by reputable sources. Additionally, these herbs contain a variety of other active compounds and due to its poor oral bioavailability, it is unlikely that the consumption of these herbs would provide levels of ligustilide sufficient to provide clinically meaningful benefits. Due to extensive first pass metabolism in the liver, the oral bioavailability in rats was estimated to be 2.6% [1].

Novel formulations are needed for ligustilide to be clinically viable. There have been some efforts to improve the stability and oral bioavailability of ligustilide. A nanoemulsion improved the pharmacokinetic profile, and *in vivo* anti-inflammatory activity of ligustilide in rats [21]. A complex of ligustilide with hydroxypropyl- β -cyclodextrin, which increases the solubility of lipophilic drugs, improved oral bioavailability to 35.9% [29].

Research underway:

According to [Clinicaltrials.gov](#), there are currently 6 active or recruiting trials using traditional Chinese preparations that include ligustilide containing *Angelica sinensis*. Notably, one of these is a retrospective study examining the effectiveness of Chinese herbal medicine in Alzheimer's disease ([NCT03221894](#)).

Search terms:

Pubmed, Google: Ligustilide or *Angelica sinensis* or Dong quai +

Dementia, Alzheimer's disease, neuroprotection, neurodegeneration, stroke, cognitive, klotho, Nrf2, aging, lifespan, cardiovascular, inflammation, cancer, menopause, clinical trials, safety, meta-analysis, pharmacokinetics, bioavailability

Websites visited for Ligustilide:

- Clinicaltrials.gov (Dong quai)
- Drugs.com (Dong quai)
- WebMD.com (Dong quai)
- PubChem (z-ligustilide)
- DrugBank.ca (Angelica sinensis)
- Patientslikeme.com (Dong quai)

References:

1. Yan R, Ko NL, Li S-L *et al.* (2008) Pharmacokinetics and Metabolism of Ligustilide, a Major Bioactive Component in Rhizoma Chuanxiong, in the Rat. *Drug Metabolism and Disposition* 36, 400-408. <http://dmd.aspetjournals.org/content/dmd/36/2/400.full.pdf>
2. Guo J, Shang E-x, Liu J-L *et al.* (2011) *Determination of ligustilide in the brains of freely moving rats using microdialysis coupled with ultra performance liquid chromatography/mass spectrometry.* vol. 82.
3. Kuang X, Du J-R, Chen Y-S *et al.* (2009) Protective effect of Z-ligustilide against amyloid β -induced neurotoxicity is associated with decreased pro-inflammatory markers in rat brains. *Pharmacology Biochemistry and Behavior* 92, 635-641. <http://www.sciencedirect.com/science/article/pii/S009130570900094X>
4. Xu Y-J, Mei Y, Qu Z-L *et al.* (2018) Ligustilide Ameliorates Memory Deficiency in APP/PS1 Transgenic Mice via Restoring Mitochondrial Dysfunction. *BioMed Research International* 2018, 15. <https://doi.org/10.1155/2018/4606752>
5. Kuang X, Zhou H-J, Thorne AH *et al.* (2017) Neuroprotective Effect of Ligustilide through Induction of α -Secretase Processing of Both APP and Klotho in a Mouse Model of Alzheimer's Disease. *Frontiers in aging neuroscience* 9, 353-353. <https://www.ncbi.nlm.nih.gov/pubmed/29163135> <https://www.ncbi.nlm.nih.gov/pmc/PMC5673635/>
6. Li J-J, Zhu Q, Lu Y-P *et al.* (2015) Ligustilide prevents cognitive impairment and attenuates neurotoxicity in d-galactose induced aging mice brain. *Brain Research* 1595, 19-28. <http://www.sciencedirect.com/science/article/pii/S0006899314013870>
7. Feng Z, Lu Y, Wu X *et al.* (2012) Ligustilide alleviates brain damage and improves cognitive function in rats of chronic cerebral hypoperfusion. *Journal of Ethnopharmacology* 144, 313-321. <http://www.sciencedirect.com/science/article/pii/S0378874112006095>



8. Long F-Y, Shi M-Q, Zhou H-J *et al.* (2018) Klotho upregulation contributes to the neuroprotection of ligustilide against cerebral ischemic injury in mice. *European Journal of Pharmacology* 820, 198-205. <http://www.sciencedirect.com/science/article/pii/S0014299917308026>
9. Chen D, Tang J, Khatibi NH *et al.* (2011) Treatment with Z-Ligustilide, a Component of *Angelica sinensis*, Reduces Brain Injury after a Subarachnoid Hemorrhage in Rats. *Journal of Pharmacology and Experimental Therapeutics* 337, 663-672. <http://jpet.aspetjournals.org/content/jpet/337/3/663.full.pdf>
10. Li L, Wang N, Jin Q *et al.* (2017) Protection of Tong-Qiao-Huo-Xue Decoction against Cerebral Ischemic Injury through Reduction Blood-Brain Barrier Permeability. *Chemical and Pharmaceutical Bulletin* 65, 1004-1010
11. Peng B, Zhao P, Lu Y-P *et al.* (2013) Z-ligustilide activates the Nrf2/HO-1 pathway and protects against cerebral ischemia-reperfusion injury in vivo and in vitro. *Brain Research* 1520, 168-177. <http://www.sciencedirect.com/science/article/pii/S0006899313006549>
12. Kuang X, Wang L-F, Yu L *et al.* (2014) Ligustilide ameliorates neuroinflammation and brain injury in focal cerebral ischemia/reperfusion rats: involvement of inhibition of TLR4/peroxiredoxin 6 signaling. *Free Radical Biology and Medicine* 71, 165-175. <http://www.sciencedirect.com/science/article/pii/S0891584914001452>
13. Kuang X, Du J-R, Liu Y-X *et al.* (2008) Posts ischemic administration of Z-Ligustilide ameliorates cognitive dysfunction and brain damage induced by permanent forebrain ischemia in rats. *Pharmacology Biochemistry and Behavior* 88, 213-221. <http://www.sciencedirect.com/science/article/pii/S0091305707002560>
14. Han L, Liu D-L, Zeng Q-K *et al.* (2018) The neuroprotective effects and probable mechanisms of Ligustilide and its degradative products on intracerebral hemorrhage in mice. *International Immunopharmacology* 63, 43-57. <http://www.sciencedirect.com/science/article/pii/S1567576918302984>
15. Li J, Yu J, Ma H *et al.* (2017) Intranasal Pretreatment with Z-Ligustilide, the Main Volatile Component of *Rhizoma Chuanxiong*, Confers Prophylaxis against Cerebral Ischemia via Nrf2 and HSP70 Signaling Pathways. *Journal of Agricultural and Food Chemistry* 65, 1533-1542. <https://doi.org/10.1021/acs.jafc.6b04979>
16. Chi K, Fu R-H, Huang Y-C *et al.* (2016) Therapeutic Effect of Ligustilide-Stimulated Adipose-Derived Stem Cells in a Mouse Thromboembolic Stroke Model. *Cell Transplantation* 25, 899-912. <https://journals.sagepub.com/doi/abs/10.3727/096368916X690539>
17. Zhu M-D, Zhao L-X, Wang X-T *et al.* (2014) Ligustilide inhibits microglia-mediated proinflammatory cytokines production and inflammatory pain. *Brain Research Bulletin* 109, 54-60. <http://www.sciencedirect.com/science/article/pii/S0361923014001506>
18. Qian B, Li F, Zhao L-X *et al.* (2016) Ligustilide Ameliorates Inflammatory Pain and Inhibits TLR4 Upregulation in Spinal Astrocytes Following Complete Freund's Adjuvant Peripheral Injection. *Cellular and Molecular Neurobiology* 36, 143-149. <https://doi.org/10.1007/s10571-015-0228-0>
19. Chao W-W, Hong Y-H, Chen M-L *et al.* (2010) Inhibitory effects of *Angelica sinensis* ethyl acetate extract and major compounds on NF- κ B trans-activation activity and LPS-induced inflammation. *Journal of Ethnopharmacology* 129, 244-249. <http://www.sciencedirect.com/science/article/pii/S0378874110001844>
20. Li X, Wu D, Hu Z *et al.* (2018) The Protective Effect of Ligustilide in Osteoarthritis: An in Vitro and in Vivo Study. *Cellular Physiology and Biochemistry* 48, 2583-2595. <https://www.karger.com/DOI/10.1159/000492701>



21. Ma Z, Bai L (2013) Anti-inflammatory Effects of Z-Ligustilide Nanoemulsion. *Inflammation* 36, 294-299. <https://doi.org/10.1007/s10753-012-9546-2>
22. Choi ES, Yoon JJ, Han BH *et al.* (2018) Ligustilide attenuates vascular inflammation and activates Nrf2/HO-1 induction and, NO synthesis in HUVECs. *Phytomedicine* 38, 12-23. <http://www.sciencedirect.com/science/article/pii/S0944711317301393>
23. Cao Y-X, Zhang W, He J-Y *et al.* (2006) Ligustilide induces vasodilatation via inhibiting voltage dependent calcium channel and receptor-mediated Ca²⁺ influx and release. *Vascular Pharmacology* 45, 171-176. <http://www.sciencedirect.com/science/article/pii/S1537189106001182>
24. Lang F, Qu J, Yin H *et al.* (2018) Apoptotic cell death induced by Z-Ligustilide in human ovarian cancer cells and role of NRF2. *Food and Chemical Toxicology* 121, 631-638. <http://www.sciencedirect.com/science/article/pii/S0278691518306847>
25. Qi H, Jiang Z, Wang C *et al.* (2017) Sensitization of tamoxifen-resistant breast cancer cells by Z-ligustilide through inhibiting autophagy and accumulating DNA damages. *Oncotarget* 8, 29300-29317. <https://www.ncbi.nlm.nih.gov/pubmed/28431397> <https://www.ncbi.nlm.nih.gov/pmc/PMC5438731/>
26. Tsai N-M, Lin S-Z, Lee C-C *et al.* (2005) The Antitumor Effects of Angelica sinensis on Malignant Brain Tumors In vitro and In vivo. *Clinical Cancer Research* 11, 3475-3484. <http://clincancerres.aacrjournals.org/content/clincanres/11/9/3475.full.pdf>
27. Wang CC, Cheng KF, Lo WM *et al.* (2013) A randomized, double-blind, multiple-dose escalation study of a Chinese herbal medicine preparation (Dang Gui Buxue Tang) for moderate to severe menopausal symptoms and quality of life in postmenopausal women. *Menopause* 20, 223-231. https://journals.lww.com/menopausejournal/Fulltext/2013/02000/A_randomized_double_blind_multiple_dose.18.aspx
28. Hirata JD, Swiersz LM, Zell B *et al.* (1997) Does dong quai have estrogenic effects in postmenopausal women? A double-blind, placebo-controlled trial. *Fertility and Sterility* 68, 981-986. <http://www.sciencedirect.com/science/article/pii/S001502829700397X>
29. Lu Y, Liu S, Zhao Y *et al.* (2014) Complexation of Z-ligustilide with hydroxypropyl- β -cyclodextrin to improve stability and oral bioavailability. 64, 211. <https://content.sciendo.com/view/journals/acph/64/2/article-p211.xml>



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