KKL RESEARCH

SUPPLEMENT RESEARCH

MARCH 2016

KKL RESEARCH

Contents

Magnesium Malate

Abraham, G. E., & Flechas, J. D. (1992). Management of fibromyalgia: Rationale for the use of magnesium and malic acid. *Journal of Nutritional Medicine*, *3*(1), 49.

Proposes that primary fibromyalgia (FM) symptoms are caused by enhanced gluconeogenesis with breakdown of muscle proteins, resulting from a deficiency of oxygen and other substances need for ATP synthesis. Role of magnesium and malate in ATP production; Worsening of muscle pain upon placebo administration; Integrity of mitochondrial membrane and capacity of respiratory chain.

Kim, Y., Kim, K., Lee, D., Kim, B., Park, S., Cho, D., & ... Joo, N. (2011). Women with fibromyalgia have lower levels of calcium, magnesium, iron and manganese in hair mineral analysis. *Journal of Korean Medical Science*, *26*(10), 1253-1257. doi:10.3346/jkms.2011.26.10.1253

Little is known about hair mineral status in fibromyalgia patients. This study evaluated the characteristics of hair minerals in female patients with fibromyalgia compared with a healthy reference group. Forty-four female patients diagnosed with fibromyalgia according to the American College of Rheumatology criteria were enrolled as the case group. Age and body mass index-matched data were obtained from 122 control subjects enrolled during visit for a regular health check-up. Hair minerals were analyzed and compared between the two groups. The mean age was 43.7 yr. General characteristics were not different between the two groups. Fibromyalgia patients showed a significantly lower level of calcium (775 μg/g vs 1,093 μg/g), magnesium (52 μg/g vs 72 μg/g), iron (5.9 μg/g vs 7.1 μg/g), copper (28.3 μg/g vs 40.2 μg/g) and manganese (140 ng/g vs 190 ng/g). Calcium, magnesium, iron, and manganese were loaded in the same factor using factor analysis; the mean of this factor was significantly lower in fibromyalgia group in multivariate analysis with adjustment for potential confounders. In conclusion, the concentrations of calcium, magnesium, iron, and manganese in the hair of female patients with fibromyalgia are lower than of controls, even after adjustment of potential confounders.

Malic Acid

Abraham, G. E., & Flechas, J. D. (1992). Management of fibromyalgia: Rationale for the use of magnesium and malic acid. *Journal of Nutritional Medicine*, *3*(1), 49.

Proposes that primary fibromyalgia (FM) symptoms are caused by enhanced gluconeogenesis with breakdown of muscle proteins, resulting from a deficiency of oxygen and other substances need for ATP synthesis. Role of magnesium and malate in ATP production; Worsening of muscle pain upon placebo administration; Integrity of mitochondrial membrane and capacity of respiratory chain.

Russell, I. J., Michalek, J. E., Flechas, J. D., & Abraham, G. E. (1995). Treatment of fibromyalgia syndrome with Super Malic: a randomized, double blind, placebo controlled, crossover pilot study. *The Journal of Rheumatology*, *22*(5), 953–8.

Objective: To study the efficacy and safety of Super Malic, a proprietary tablet containing malic acid (200 mg) and magnesium (50 mg), in treatment of primary fibromyalgia syndrome (FM). Methods: Twenty-four sequential patients with primary FM were randomized to a fixed dose (3 tablets bid), placebo controlled, 4-week/course, pilot trial followed by a 6-month, open label, dose escalation (up to 6 tablets bid) trial. A 2-week, medication free, washout period was required before receiving treatment, between blinded courses, and again before starting open label treatment. The 3 primary outcome variables were measures of pain and tenderness but functional and psychological measures were also assessed. Results: No clear treatment effect attributable to Super Malic was seen in the blinded, fixed low dose trial. With dose escalation and a longer duration of treatment in the open label trial, significant reductions in the severity of all 3 primary pain/tenderness measures were obtained without limiting risks. Conclusions: These data suggest that Super Malic is safe and may be beneficial in the treatment of patients with FM. Future placebo-controlled studies should utilize up to 6 tablets of Super Malic bid and continue therapy for at least 2 months. (Full-text unavailable)

Quercetin

Aguirre-Hernández, E., González-Trujano, M. E., Terrazas, T., Herrera Santoyo, J., & Guevara-Fefer, P. (2016). Anxiolytic and sedative-like effects of flavonoids from Tilia americana var. mexicana: GABAergic and serotonergic participation. *Salud Mental*, *39*(1), 37-46. doi:10.17711/SM.0185-3325.2015.066

Introduction The inflorescences of Tilia americana var. mexicana are used as an infusion in Mexican traditional medicine due to their tranquilizing effects; however, pharmacological and phytochemical studies of the leaves are lacking. In this research, the anxiolytic and sedative-like efficacy of the Tilia americana var. mexicana leaves was compared to that obtained with its inflorescences and flavonoids therein identified, as well as the possible mechanism of action. Methods The sorted and dried inflorescences and leaves were macerated subsequently in hexane, ethyl acetate and methanol. The methanol extracts were qualitative- and quantitative-analyzed by HPLC, using commercial flavonoids standards selected on the basis of their previously reported presence in Tilia species. The pharmacological activity was evaluated in CD-1 mice in the tests: open-field, elevated plus-maze, hole-board, and the sodium pentobarbital-induced sleep potentiation test. In regard to the mechanism of action, participation of benzodiazepine and 5-HT1A serotonin receptors was tested with the respective antagonists: flumazenil and WAY100635. Results The presence of quercetin, rutin and isoquercitrin was confirmed in the extracts of the inflorescences and leaves. The anxiolytic-like effects were the same between the two organs, which were inhibited in the presence of flumazenil and WAY100635. Discussion and conclusion Our results provide evidence that the extracts of the leaves of T. americana var. mexicana are as efficacious as the inflorescences to produce anxiolytic and sedative-like effects, where flavonoids like quercetin, rutin and isoquercitrin are partially responsible for these activities by the involvement of GABA/BDZ and 5HT1A serotonergic receptors.

Ashrafpour, M., Parsaei, S., & Sepehri, H. (2015). Quercetin improved spatial memory dysfunctions in rat model of intracerebroventricular streptozotocin-induced sporadic Alzheimer's disease. *National Journal of Physiology, Pharmacy & Pharmacology*, *5*(5), 411-415. doi:10.5455/njppp.2015.5.2308201563

The article presents research on the use of quercetin in spatial memory dysfunction improvement in the intracerebroventricular streptozotocin-induced sporadic Alzheimer's disease (AD) of rats. Results of the study show the impairment in spatial memory acquisition and retrieval and the decrease of escape latency in training trials.

Calixto-Campos, C., Corrêa, M. P., Carvalho, T. T., Zarpelon, A. C., Hohmann, M. N., Rossaneis, A. C., & ... Verri, W. J. (2015). Quercetin reduces Ehrlich tumor-induced cancer pain in mice. *Analytical Cellular Pathology (Amsterdam)*, *2015*285708. doi:10.1155/2015/285708

Cancer pain directly affects the patient's quality of life. We have previously demonstrated that the subcutaneous administration of the mammary adenocarcinoma known as Ehrlich tumor induces pain in mice. Several studies have shown that the flavonoid quercetin presents important biological effects, including anti-inflammatory, antioxidant, analgesic, and antitumor activity. Therefore, the analgesic effect

and mechanisms of quercetin were evaluated in Ehrlich tumor-induced cancer pain in mice. Intraperitoneal (i.p.) treatments with quercetin reduced Ehrlich tumor-induced mechanical and thermal hyperalgesia, but not paw thickness or histological alterations, indicating an analgesic effect without affecting tumor growth. Regarding the analgesic mechanisms of quercetin, it inhibited the production of hyperalgesic cytokines IL-1β and TNFα and decreased neutrophil recruitment (myeloperoxidase activity) and oxidative stress. Naloxone (opioid receptor antagonist) inhibited quercetin analgesia without interfering with neutrophil recruitment, cytokine production, and oxidative stress. Importantly, cotreatment with morphine and quercetin at doses that were ineffective as single treatment reduced the nociceptive responses. Concluding, quercetin reduces the Ehrlich tumor-induced cancer pain by reducing the production of hyperalgesic cytokines, neutrophil recruitment, and oxidative stress as well as by activating an opioid-dependent analgesic pathway and potentiation of morphine analgesia. Thus, quercetin treatment seems a suitable therapeutic approach for cancer pain that merits further investigation.

Casuso, R. A., Martínez-Amat, A. a., Martínez-López, E. J., Camiletti-Moirón, D., Porres, J. M., & Aranda, P. (2013). Ergogenic effects of quercetin supplementation in trained rats. *Journal of the International Society of Sports Nutrition*, *10*(1), 1-7.

Background: Quercetin is a natural polyphenolic compound currently under study for its ergogenic capacity to improve mitochondrial biogenesis. Sedentary mice have exhibited increased endurance performance, but results are contradictory in human models. Methods: We examined the effects of six weeks of endurance training and quercetin supplementation on markers of endurance performance and training in a rodent model. Rats were randomly assigned to one of the following groups: placebo+sedentary (PS), quercetin+sedentary (QS), placebo+endurance training (PT) and quercetin +endurance training (QT). Quercetin was administered at a dose of 25 mg/kg on alternate days. During six weeks of treatment volume parameters of training were recorded, and after six weeks all groups performed a maximal graded VO_2 max test and a low-intensity endurance run-to-fatigue test. Results: No effects were found in VO_2 peak ($p>0.999$), nor in distance run during low-intensity test, although it was 14% greater in QT when compared with PT ($P = 0.097$). Post-exercise blood lactate was increased in QT when compared with PT ($p=0.023$) and also in QS compared with PS ($p=0.024$). Conclusions: This study showed no effects in VO_2 peak, speed at VO_2 peak or endurance time to exhaustion after six weeks of quercetin supplementation compared with placebo in trained rats. Quercetin was show to increase blood lactate production after high-intensity exercise.

Casuso, R. A., Martínez-Amat, A., Martínez-Romero, R., Camiletti-Moiron, D., Hita-Contreras, F., & Martínez-López, E. (2013). Plasmatic nitric oxide correlates with weight and red cell distribution width in exercised rats supplemented with quercetin. *International Journal of Food Sciences & Nutrition*, *64*(7), 830-835. doi:10.3109/09637486.2013.803521

Quercetin is suggested as a nitric oxide regulator which may in turn influence blood parameters and weight gain. Wistar rats were classified as: quercetin-exercise training, QT; placebo-exercise training, PT; quercetin-sedentary, QS; and placebo sedentary, PS. After 6 weeks of treatment with quercetin and/or exercise, an incremental test was run to measure oxygen consumption. QT had lower levels of NO compared with PS ($p =$

0.029) and QS (p = 0.002). Red cell distribution width increased in both exercised groups, especially in the QT group (p < 0.001). Pearson correlation analysis showed that nitric oxide levels were associated with weight (r = 0.675) and red distribution width (r = −0814) in the QT group. Quercetin effect on NO production seems to be more powerful when it is supplemented during exercise training. Moreover, RDW relationship with NO production need to be further investigated in regards to health.

Chakraborty, J., Singh, R., Dutta, D., Naskar, A., Rajamma, U., & Mohanakumar, K. P. (2014). Quercetin Improves Behavioral Deficiencies, Restores Astrocytes and Microglia, and Reduces Serotonin Metabolism in 3-Nitropropionic Acid-Induced Rat Model of Huntington's Disease.*CNS Neuroscience & Therapeutics*, *20*(1), 10-19. doi:10.1111/cns.12189

Aim Huntington's disease (HD) is an autosomal dominant disorder, for which clinically available drugs offer only symptomatic relief. These prescription drugs are not free of side effects, and the patients usually suffer from anxiety and depression. We investigated quercetin, a dietary flavonoid with free radical scavenging properties, for its beneficial potential if any, in 3-nitropropionic acid (3- NP)-induced HD in rats where both drugs were administered simultaneously. Methods Performance of rats on beam balancing, elevated plus maze and gait traits were investigated following 3- NP and/or quercetin treatments for 4 days. Striatal biogenic amine levels and monoamine oxidase activity were assayed. Striatal sections were examined for Cd11B and glial fibrillary acidic protein immunoreactivity, and for evidences of neuronal lesion. Results Quercetin significantly attenuated 3- NP-induced anxiety, motor coordination deficits, and gait despair. While the dopaminergic hyper-metabolism was unaffected, quercetin provided a significant reduction of 3-NP mediated increase in serotonin metabolism. Quercetin failed to affect 3- NP-induced striatal neuronal lesion, but decreased microglial proliferation, and increased astrocyte numbers in the lesion core. Conclusion These results taken together suggest that quercetin could be of potential use not only for correcting movement disturbances and anxiety in HD, but also for addressing inflammatory damages.

Fenglian, X., Proft, J., Gibbs, S., Winkfein, B., Johnson, J. N., Syed, N., & Braun, J. A. (2010). Quercetin Targets Cysteine String Protein (CSPa) and Impairs Synaptic Transmission. *Plos ONE*, *5*(6), 1-13. doi:10.1371/journal.pone.0011045

Background: Cysteine string protein (CSPα) is a synaptic vesicle protein that displays unique anti-neurodegenerative properties. CSPα is a member of the conserved J protein family, also called the Hsp40 (heat shock protein of 40 kDa) protein family, whose importance in protein folding has been recognized for many years. Deletion of the CSPα in mice results in knockout mice that are normal for the first 2-3 weeks of life followed by an unexplained presynaptic neurodegeneration and premature death. How CSPα prevents neurodegeneration is currently not known. As a neuroprotective synaptic vesicle protein, CSPα represents a promising therapeutic target for the prevention of neurodegenerative disorders. Methodology/Principal Findings: Here, we demonstrate that the flavonoid quercetin promotes formation of stable CSPα- CSPα dimers and that quercetin-induced dimerization is dependent on the unique cysteine string region. Furthermore, in primary cultures of Lymnaea neurons, quercetin induction of CSPα dimers correlates with an inhibition of synapse formation and synaptic transmission suggesting that quercetin interferes with CSPα

function. Quercetin's action on CSPα is concentration dependent and does not promote dimerization of other synaptic proteins or other J protein family members and reduces the assembly of CSPα:Hsc70 units (70kDa heat shock cognate protein). Conclusions/Significance: Quercetin is a plant derived flavonoid and popular nutritional supplement proposed to prevent memory loss and altitude sickness among other ailments, although its precise mechanism(s) of action has been unclear. In view of the therapeutic promise of upregulation of CSPα and the undesired consequences of CSPα dysfunction, our data establish an essential proof of principle that pharmaceutical agents can selectively target the neuroprotective J protein CSPα.

Hassanzadeh, P. & Ahmadiani A. (2007). Inflammatory pain induces neuronal alterations in NO and JNK dependent manners. *Daru*, *15*(4), 183-187.

Background: Dark neurons are generated in vivo as an acute or delayed consequence of several pathological situations and lesions. The present study was designed to evaluate whether inflammatory pain induces formation of dark neurons in the dorsal horn of rat spinal cord. Since NO and JNK pathway are involved in the mechanisms of pain generation and degenerative neuronal alteration, their roles were also considered. Methods: Histological procedures were employed for detection of dark neurons following induction of inflammatory pain. Results: On the fifth day; following daily injections of 5% formalin, numbers of dark neurons increased significantly. Acute and chronic administration of 1% or 2.5% formalin did not induce any remarkable neuronal alteration in the dorsal horn of lumbar spinal cord. Daily intrathecal administration of quercetin (inhibitor of JNK pathway) 100µg/rat, or PTIO (NO scavenger) 30µg/rat before injection of 5% formalin, led to a reliable reduction of formation of dark neurons. Conclusion: Results indicate that induction of inflammatory pain for longer periods may result in a serious central disorder, and administration of neutralizers or inhibitors of NO and JNK may exert neuroprotective effects.

Heeba, G. H., Mahmoud, M. E., & Hanafy, A. E. (2014). Anti-inflammatory potential of curcumin and quercetin in rats: Role of oxidative stress, heme oxygenase-1 and TNF-α. *Toxicology & Industrial Health*, *30*(6), 551-560. doi:10.1177/0748233712462444

Flavonoids are group of compounds that have been shown to possess potent anti-inflammatory effects in both cellular and animal models of inflammation. In the current study, the single and combined effects of the two flavonoids, curcumin and quercetin, against carrageenan-induced acute inflammation in rats were evaluated with emphasis on the role of oxidative stress, anti-inflammatory enzyme, heme oxygenase-1 (HO-1) and proinflammatory cytokine, tumor necrosis factor-alpha (TNF-α). Curcumin (50 mg/kg), quercetin (50 mg/kg) and a combination of both were orally administered for 14 days before carrageenan injection in rats and compared with the reference nonsteroidal anti-inflammatory drug, indomethacin (10 mg/kg). The percentage increase in paw thickness was calculated. Frozen hind paws were used for the estimation of lipid peroxides (malondialdehyde, MDA), nitric oxide (NO), reduced glutathione (GSH), TNF-α level and HO-1 messenger RNA (mRNA) expression. Formalin-fixed hind paws were used for histopathological examination. Results showed that both curcumin and quercetin caused reduction in carrageenin-induced edema and lymphocytes infiltration along with the decrease is being even higher in case of their combination.

Additionally, both flavonoids reduced MDA and NO formation, and restored GSH contents in the paw. Furthermore, both flavonoids increased HO-1 mRNA expression and decreased the elevated TNF-α level. Results showed that both flavonoids moderately lowered inflammation, while their combination was more effective. Accordingly, this study suggests that the reduction in oxidative stress and modulation of HO-1 mRNA expression and TNF-α release by curcumin and quercetin may contribute to the synergistic anti-inflammatory effects of these two flavonoids upon combination.

Islam, M. R., Zaman, A., Jahan, I., Chakravorty, R., & Chakraborty, S. (2013). In silico QSAR analysis of quercetin reveals its potential as therapeutic drug for Alzheimer's disease. *Journal of Young Pharmacists*, *5*(4), 173-179. doi:10.1016/j.jyp.2013.11.005

Acetylcholine-esterase (AchE) inhibitors are one of the most potent drug molecules against Alzheimer's disease (AD). But, patients treated with current AchE inhibitors often experience severe side effects. Quercetin is a plant flavonoid compound which can act as AchE inhibitor and it may be a better alternative to current AchE inhibitors in terms of effectiveness with no or fewer side effects. Aims: The aim of the study was to compare quercetin with conventional AchE inhibitors to search for a better drug candidate. Methods and materials: Physico-chemical properties of conventional drugs and quercetin were predicted using bioinformatics tools. Molecular docking of these compounds on the active site of AchE was performed using AutoDock and comparative analysis was performed. Later, modification on the basic structure of quercetin with different functional groups was done to perform QSAR analysis. Result and discussion: Quercetin showed a similar drug likeness score to the conventional drugs. The binding strength for quercetin in the active site of the enzyme was –8.8 kcal/mol, which was considerably higher than binding scores for some of the drugs such as donepezil (binding score –7.9 kcal/mol). Fifteen hydrogen bonds were predicted between quercetin and the enzyme whereas conventional drugs had fewer or even no hydrogen bonds. It implies that quercetin can act as a better inhibitor than conventional drugs. To find out even better inhibitor, similar structures of quercetin were searched through SIMCOMP database and a methylation in the 4-OH position of the molecule showed better binding affinity than parent quercetin. Quantitative structure activity relationship study indicated that O-4 methylation was specifically responsible for better affinity. Conclusion: This in silico study has conclusively predicted the superiority of the natural compound quercetin over the conventional drugs as AchE inhibitor and it sets the need for further in-vitro study of this compound in future.

Kumar, A., & Goyal, R. (2008). Quercetin protects against acute immobilization stress-induced behaviors and biochemical alterations in mice. *Journal of Medicinal Food*, *11*(3), 469-473. doi:10.1089/jmf.2006.0207

Oxidative stress is a major contributor to the alterations of various pathological conditions, including neurodegenerative and neuropsychiatric problems. Antioxidative flavonoids, ubiquitously included in vegetables, fruits, and teas, are expected to prevent degenerative diseases. Recently, flavonoids have been characterized as neuroprotectants in the treatment of various neurological disorders. The present study was designed to investigate protective effects of quercetin, a bioflavonoid, against acute immobilization-induced

behavioral and biochemical alterations in mice. Mice were immobilized for a period of 6 hours. Quercetin (20 and 40 mg/kg, i.p.) was administered 30 minutes before subjecting the animals to acute stress. Behavioral tests (mirror chamber, actophotometer, and tail flick test) and biochemical analysis (malondialdehyde, reduced glutathione, catalase, nitrite, and protein levels) were subsequently performed. Acute immobilization stress for a period of 6 hours caused severe anxiety, analgesia, and impaired motor activity in mice. Biochemical analyses revealed an increase in malondialdehyde and nitrite levels as well as partial depletion of reduced glutathione and catalase activity in immobilization-stressed brain. Behavioral and biochemical parameters were significantly altered as compared to naive mice. Pretreatment with quercetin (20 and 40 mg/kg, i.p.) significantly reversed immobilized stress-induced anxiety and analgesia and reduced locomotor activity. Biochemically, quercetin treatment attenuated malondialdehyde accumulation and nitrite activity and restored the depleted reduced glutathione and catalase activity. Neuroprotective effects of quercetin were significantly improved as compared to control (immobilized stressed) animals. Results suggest that neuroprotective properties of quercetin can be used in the treatment and management of stress and related disorders. (Full-text unavailable)

McAnulty, L. S., Miller, L. E., Hosick, P. A., Utter, A. C., Quindry, J. C., & McAnulty, S. R. (2013). Effect of resveratrol and quercetin supplementation on redox status and inflammation after exercise. *Applied Physiology, Nutrition & Metabolism*, *38*(7), 760-765. doi:10.1139/apnm-2012-0455

Resveratrol and quercetin function as antioxidants and anti-inflammatories in vitro, but these mechanisms have been minimally examined in combination in exercising humans. The purpose of this investigation was to examine supplementation as a countermeasure against oxidative stress and inflammation in response to exercise. Fourteen athletes were randomly assigned, in a double-blind crossover design, to a resveratrol and quercetin combination (RQ) (120 mg resveratrol and 225 mg quercetin for 6 days and 240 mg resveratrol and 450 mg quercetin on day 7 just prior to exercise) or to placebo (P). There was a 1-week washout between trials. Blood was taken at baseline, pre-exercise, immediately after exercise, and 1 h after exercise. Plasma was analyzed for oxidative stress (F_2-isoprostanes and protein carbonyls), antioxidant capacity (ferric-reducing ability of plasma (FRAP), Trolox equivalent antioxidant capacity (TEAC), oxygen radical absorptive capacity (ORAC)), and inflammation (cytokine interleukin (IL)-8 and C-reactive protein (CRP)). Statistical design utilized a 2 × 3 ANOVA and Student's t test. Pre-exercise values were not different from baseline for any measure. The postexercise increase in F_2-isoprostanes was significantly less ($p = 0.039$ interaction) with RQ (68%) than with P (137%). Protein carbonyls, FRAP, ORAC, and TEAC significantly increased after exercise but were not affected by treatment. IL-8 and CRP increased significantly immediately after exercise but were not affected by treatment. These data indicate that RQ significantly reduces exercise-induced lipid peroxidation without associated changes in inflammation or plasma antioxidant status.

Liu, P., Zou, D., Yi, L., Chen, M., Gao, Y., Zhou, R., & ... Mi, M. (2015). Quercetin ameliorates hypobaric hypoxia-induced memory impairment through mitochondrial and neuron function adaptation via the PGC-1α pathway. *Restorative Neurology & Neuroscience*, *33*(2), 143-157. doi:10.3233/RNN-140446.

Purpose: Acute hypobaric hypoxia (HH) causes persistent cognitive impairment, affecting memory function specifically. Mitochondrial dysfunction and synaptic morphological change were the prominent pathological features of HH exposure on brain. Quercetin, a flavonoid found in fruits, vegetables, leaves and grains, is reported to prevent ischemia induced by neuronal injury. This study investigated the efficacy of quercetin to ameliorate HH-induced memory deficit. Methods: Rats were exposed to HH equivalent to 5000 m for 7 days in a decompression chamber and received quercetin daily (50, 75 or 100 mg/kg·bw) via gavage during the period of exposure. Cognitive performance was assessed by the Morris water maze test. In vitro, the effect of quercetin was tested in hippocampus tissue. Results: Quercetin, especially at 100 mg/kg·bw, significantly reduced HH-induced memory decline. Meanwhile, HH-induced hippocampus mitochondrial and synaptic lesions were ameliorated by quercetin. Furthermore, quercetin regulated the expression of sirtuin 1(Sirt1), PGC-1α, and the proteins related with mitochondrial biogenesis and dynamics. Moreover, quercetin increased expression of fibronectin type III domain-containing protein 5 (FNDC5) and brain-derived neurotrophic factor (BDNF), showing the PGC-1α/FNDC5/BNDF pathways might be involved in neuronal adaptation. Conclusions: The results suggest quercetin has prophylactic potential for amelioration of HH-induced memory impairment, which is associated with the mitochondrial and neuronal adaptation in hippocampus.

Moh San, A. M., Thongpraditchote, S., Sithisarn, P., & Gritsanapan, W. (2013). Total Phenolics and Total Flavonoids Contents and Hypnotic Effect in Mice of Ziziphus mauritiana Lam. Seed Extract. *Evidence-Based Complementary & Alternative Medicine (Ecam)*, *2013*1-4.

The seeds of Ziziphus mauritiana Lam. have been traditionally used for treatment of various complications including insomnia and anxiety. They are popularly used as sedative and hypnotic drugs in China, Korea, Myanmar, Vietnam, and other Asian countries. However, no scientific proof on hypnotic activity of Z. mauritiana seeds (ZMS) was reported. In this study, the hypnotic activity of 50% ethanolic extract from ZMS was observed on the loss of righting reflex in mice using pentobarbital-induced sleep mice method. The contents of total phenolics and total flavonoids in the extract were also determined. The results showed that the 50% ethanolic extract from ZMS contained total phenolics 27.62 ± 1.43 mg gallic acid equivalent (GAE)/g extract and total flavonoids 0.74 ± 0.03 mg quercetin equivalent (QE)/g extract. Oral administration of the extract at the dose of 200 mg/kg significantly increased the sleeping time in mice intraperitoneally administered with sodium pentobarbital (50 mg/kg body weight). These results supported the traditional use of ZMS for the treatment of insomnia. The seeds of Z. mauritiana should be further developed as an alternative sedative and/or hypnotic product.

Napatr, S., Jintanaporn W., Supaporn, M., Somsak, T., Kamoltip, B., & Kowit, C. (2012). Cognitive-Enhancing Effect of Quercetin in a Rat Model of Parkinson's Disease Induced by 6-Hydroxydopamine. *Evidence-Based Complementary & Alternative Medicine (Ecam)*,*2012*1-9.

Oxidative stress has been reported to induce cognitive impairment in Parkinson's disease. This paper aimed to determine the effect of quercetin, a substance possessing antioxidant activity, on the cognitive function in a rat model of Parkinson's disease. Male Wistar rats, weighing 200-250 g, were orally given quercetin at doses of 100, 200, 300 mg/kg BWonce daily for a period of 14 days before and 14 days after the unilateral lesion of right substantia nigra induced by 6-hydroxydopamine (6-OHDA). Their spatial memory was assessed at 7 and 14 days of treatment and neuron density was determined, malondialdehyde (MDA) level, the activity of superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) were evaluated at the end of the experiment. In addition, the activity of acetylcholinesterase (AChE) was also measured. It was found that all doses of quercetin enhanced spatial memory. Therefore, it is suggested that the cognitive-enhancing effect of quercetin occurs partly because of decreased oxidative damage resulting in increased neuron density.

Pu, F., Mishima, K., Irie, K., Motohashi, K., Tanaka, Y., Orito, K., & ... Fujiwara, M. (2007). Neuroprotective effects of quercetin and rutin on spatial memory impairment in an 8-arm radial maze task and neuronal death induced by repeated cerebral ischemia in rats. *Journal of Pharmacological Sciences*, *104*(4), 329-334.

In order to determine the differential effects of flavonoids on cerebral ischemia, we investigated the effects of (-)-epigallocatechin gallate (EGCG), catechin, rutin, and quercetin on spatial memory impairment and neuronal death induced by repeated cerebral ischemia in rats. Both rutin and quercetin (50 mg/kg x 2) improved spatial memory impairment in the 8-arm radial maze task and neuronal death in the hippocampal CA1 area; however, catechin (200 mg/kg x 2) and EGCG (50 mg/kg x 1) did not. Administration of EGCG (50 mg/kg x 2) resulted in a high mortality rate. These results suggest that in this repeated cerebral ischemia model, the 4-oxo group and the 2,3-double bond in the C ring of rutin and quercetin are related to their neuroprotective action. (Full-text unavailable)

Sah, S. P., Tirkey, N., Kuhad, A., & Chopra, K. (2011). Effect of quercetin on lipopolysaccharide induced-sickness behavior and oxidative stress in rats. *Indian Journal of Pharmacology*, *43*(2), 192-196. doi:10.4103/0253-7613.77365

Objectives: Gram-negative infections and control infusion of recombinant cytokines in human have been shown to induce sickness behavior characterized by fever, prolong sleep, decreased food and water intake, reduced mobility, depression, and anxiety. Therefore, the present study was undertaken to investigate the effect of bioflavonoid quercetin in lipopolysaccharide (LPS)-induced sickness behavior. Materials and Methods: Wistar albino rats were divided into six groups (n=6). Three groups received vehicle and two doses of quercetin (2 and 25 mg/kg, i.p.) respectively for 2 weeks before being challenged with LPS (1 mg/kg, i.p). One group received vehicle for 2 weeks and was challenged with saline on day 15. The per se effect of

quercetin (2 and 25 mg/kg, i.p.) was also seen after 2 weeks of dosing. LPS-induced sickness behavior in rats was quantified by measuring time in social exploration, anxiety, food and water consumption, and weight loss. Levels of cytokines (TNF-α, IL-1β, and IL-6) and oxidative stress in rat brain were also analyzed. Results: Quercetin (2 and 25 mg/kg) administration significantly ($P<0.05$) attenuated LPS-induced sickness behavior by modulating cytokines production as well inhibiting LPS-induced oxidative stress. Conclusions: Adequate intake of dietary flavonoids (like quercetin) may help promote recovery from sickness behavior.

Selvakumar, K., Prabha, R. L., Saranya, K., Bavithra, S., Krishnamoorthy, G., & Arunakaran, J. (2013). Polychlorinated biphenyls impair blood–brain barrier integrity via disruption of tight junction proteins in cerebrum, cerebellum and hippocampus of female Wistar rats: Neuropotential role of quercetin. *Human & Experimental Toxicology*, *32*(7), 706-720. doi:10.1177/0960327112464798

Polychlorinated biphenyls (PCBs) comprise a ubiquitous class of toxic substances associated with carcinogenic and tumor-promoting effects as well as neurotoxic properties. Reactive oxygen species, which is produced from PCBs, alters blood–brain barrier (BBB) integrity, which is paralleled by cytoskeletal rearrangements and redistribution and disappearance of tight junction proteins (TJPs) like claudin-5 and occludin. Quercetin, a potent antioxidant present in onion and other vegetables, appears to protect brain cells against oxidative stress, a tissue-damaging process associated with Alzheimer's and other neurodegenerative disorders. The aim of this study is to analyze the role of quercetin on oxidative stress markers and transcription of transmembrane and cytoplasmic accessory TJPs on cerebrum, cerebellum and hippocampus of female rats exposed to PCBs. Rats were divided into the following four groups. Group I: received only vehicle (corn oil) intraperitoneally (i.p.); group II: received Aroclor 1254 at a dose of 2 mg/kg body weight (bwt)/day (i.p); group III: received Aroclor 1254 (i.p.) and simultaneously quercetin 50 mg/kg bwt/day through gavage and group IV: received quercetin alone gavage. From the experiment, the levels of hydrogen peroxide, lipid peroxidation and thiobarbituric acid reactive substances were observed to increase significantly in cerebrum, cerebellum and hippocampus as 50%, 25% and 20%, respectively, after exposure to PCB, and the messenger RNA expression of TJP in rats exposed to PCBs is decreased and is retrieved to the normal level simultaneously in quercetin-treated rats. Hence, quercetin can be used as a preventive medicine to PCBs exposure and prevents neurodegenerative disorders.

Shao-Xiao, Y., Jun-Lian, L., Yan-Ying, S., Yu-Ze, W., Meng-Han, L., Xia, Z., & ... Chun-Yan, X. (2015). Studies on Anti-Depressant Activity of Four Flavonoids Isolated from Apocynum venetum Linn (Apocynaceae) Leaf in Mice. *Tropical Journal of Pharmaceutical Research*, *14*(12), 2269-2277. doi:10.4314/tjpr.v14i12.17

Purpose: To investigate the anti-depressant activity of kaempferol, quercetin, kaempferol-3-O-β-Dglucose and quercetin-3-O-β-D-glucose isolated from Apocynum venetum Linn. (Apocynaceae) leaf and their mechanisms of action. Methods: The four flavonoids were isolated from Apocynum venetum leaf by chromatography. Mice were divided into vehicle, fluoxetine, kaempferol, quercetin, kaempferol-3-O-β-D-glucose and quercetin- 3-O-β-D-glucose groups (n = 10). Forced swimming (FST), tail suspension (TST) and locomotor activity (LAT) tests were used to evaluate the effects of the four flavonoids (0.35 mM/kg) on

immobility time, monoamine neurotransmitters, viz, norepinephrine (NE), dopamine (DA) and 5-hydroxytryptamine (5- HT), as well as on the metabolite (5-HIAA) in mice brain and central nervous system (CNS) with the aid of video camera, HPLC-ECD and activity-monitoring system. Results: The four flavonoids significantly ($p < 0.05$) reduced mice immobility time (72.58 - 90.24; 52.58 - 70.24 s), 5-HIAA levels (940.8 - 1244.7; 880.8 - 1164.1 ng/g) and 5-HIAA/5-HT ratio (1.77 - 4.76; 1.83 - 4.16), but increased NE, DA and 5-HT levels (238.7 - 405.7, 308.4 - 528.1, 261.4 - 531.9; 243.9 - 423.6, 296.7 - 534.9, 279.8 - 481.4 ng/g) in FST and TST, compared with control group (146.18, 126.18 s; 1363.4, 1240.9 ng/g; 7.43, 6.16; 138.4, 235.4, 183.4 and 143.7, 218.6, 201.4 ng/g). The effects of the four flavonoids on the above indices were significant ($p < 0.05$) and positively related to their polarity. They had no CNS-stimulating effects in LAT. Conclusion: The anti-depressant activities of the four flavonoids are positively related to their polarity, and the mechanisms may be due to increased NE, DA and 5-HT and reduced 5-HT metabolism.

Sharma, D., Wani, W., Sunkaria, A., Kandimalla, R., Verma, D., Cameotra, S., & Gill, K. (2013). Quercetin Protects Against Chronic Aluminum-Induced Oxidative Stress and Ensuing Biochemical, Cholinergic, and Neurobehavioral Impairments in Rats. *Neurotoxicity Research*, *23*(4), 336-357. doi:10.1007/s12640-012-9351-6

In this study, we investigated the protective effect of chronic quercetin (a natural flavanoid) administration against Al-induced cognitive impairments, oxidative damage, and cholinergic dysfunction in male Wistar rats. Al lactate (10 mg/kg b.wt./day) was administered intragastrically to rats which were pre-treated with quercetin (10 mg/kg b.wt./day, intragastrically) for 12 weeks. At the end of 6 or 12 weeks of the study, several behavioral parameters were carried out to evaluate cognitive functions. Further after 12 weeks of exposure, various biochemical tests and H&E staining were performed to assess the extent of oxidative damage and neurodegeneration, respectively. Al levels were also estimated in HC and CS regions of rat brain. Chronic administration of quercetin caused significant improvement in the muscle coordination, cognition, anxiety, locomotion, and initial exploratory patterns in Al-treated rats. Quercetin supplementation to Al-treated animals also reduced oxidative stress, decreased ROS production, increased MnSOD activity and glutathione levels with decreased lipid peroxidation and protein oxidation. It increased AChE activity and ATP levels in HC and CS regions of rat brain compared to Al-treated rats. Quercetin administration ameliorates Al-induced neurodegenerative changes in Al-treated rats as seen by H&E staining. Further with the help of atomic absorption spectrophotometer, we found that quercetin supplementation to Al-treated rats also decreases the accumulation of Al in the HC and CS regions of rat brain. Taken together the results of this study show that quercetin offers neuroprotection against Al-induced cognitive impairments, cholinergic dysfunction, and associated oxidative damage in rats.

Theoharides, T. C. (2007). Treatment Approaches for Painful Bladder Syndrome/Interstitial Cystitis. *Drugs*, *67*(2), 215-235.

Painful bladder syndrome/interstitial cystitis (PBS/IC) is a disease of unknown aetiology, characterized by severe pressure and pain in the bladder area or lower pelvis that is frequently or typically relieved by voiding, along with urgency or frequency of urination in the absence of urinary tract infections. PBS/IC

occurs primarily in women, is increasingly recognized in young adults, and may affect as many as 0.1% of adult women. PBS/IC is often comorbid with allergies, endometriosis, fibromyalgia, irritable bowel syndrome and panic syndrome, all of which are worsened by stress. As a result, patients may visit as many as five physicians, including family practitioners, internists, gynecologists, urologists and pain specialists, leading to confusion and frustration. There is no curative treatment; intravesical dimethyl sulfoxide, as well as oral amitriptyline, pentosan polysulfate and hydroxyzine have variable results, with success more likely when these drugs are given together. Pilot clinical trials suggest that the flavonoid quercetin may be helpful. Lack of early diagnosis and treatment can affect outcomes and leads to the development of hyperalgesia/allodynia.

Velázquez, K. T., Enos, R. T., Narsale, A. A., Puppa, M. J., Davis, J. M., Murphy, E. A., & Carson, J. A. (2014). Quercetin supplementation attenuates the progression of cancer cachexia in ApcMin/+ mice. *The Journal of Nutrition*, *144*(6), 868-875. doi:10.3945/jn.113.188367

Although there are currently no approved treatments for cancer cachexia, there is an intensified interest in developing therapies because of the high mortality index associated with muscle wasting diseases. Successful treatment of the cachectic patient focuses on improving or maintaining body weight and musculoskeletal function. Nutraceutical compounds, including the natural phytochemical quercetin, are being examined as potential treatments because of their anti-inflammatory, antioxidant, and anticarcinogenic properties. The purpose of this study was to determine the effect of quercetin supplementation on the progression of cachexia in the adenomatous polyposis coli (Apc)(Min/+) mouse model of colorectal cancer. At 15 wk. of age, C57BL/6 and male Apc(Min/+) mice were supplemented with 25 mg/kg of quercetin or vehicle solution mix of Tang juice and water (V) daily for 3 wk. Body weight, strength, neuromuscular performance, and fatigue were assessed before and after quercetin or V interventions. Indicators of metabolic dysfunction and inflammatory signaling were also assessed. During the treatment period, the relative decrease in body weight in the Apc(Min/+) mice gavaged with V (Apc(Min/+)V; -14% ± 2.3) was higher than in control mice gavaged with V (+0.6% ± 1.0), control mice gavaged with quercetin (-2% ± 1.0), and Apc(Min/+) mice gavaged with quercetin (Apc(Min/+)Q; -9% ± 1.3). At 18 wk. of age, the loss of grip strength and muscle mass shown in Apc(Min/+)V mice was significantly attenuated ($P < 0.05$) in Apc(Min/+)Q mice. Furthermore, Apc(Min/+)V mice had an induction of plasma interleukin-6 and muscle signal transducer and activator of transcription 3 phosphorylation, which were significantly ($P < 0.05$) mitigated in Apc(Min/+)Q mice, despite having a similar tumor burden. Quercetin treatment did not improve treadmill run-time-to-fatigue, hyperglycemia, or hyperlipidemia in cachectic Apc(Min/+) mice. Overall, quercetin supplementation positively affected several aspects of cachexia progression in mice and warrants further exploration as a potential anticachectic therapeutic.

Wu, J., Gao, W., Wei, J., Yang, J., Pu, L., & Guo, C. (2012). Quercetin alters energy metabolism in swimming mice. *Applied Physiology, Nutrition, And Metabolism = Physiologie Appliquée, Nutrition Et Métabolisme*, *37*(5), 912-922. doi:10.1139/h2012-064

Quercetin has been demonstrated to be effective in increasing physical endurance in mice and humans. However, the mechanisms involved are not fully understood. In this study, male Kunming mice were fed a diet containing 0.1% quercetin for 14 days before swimming for 60 min. The overall serum metabolic profile was investigated by a ^{1}H nuclear magnetic resonance-based metabolomic approach. Serum glucose, lactate, nonesterified fatty acids (NEFA), and nonprotein nitrogen (NPN), as well as hepatic and muscular glycogen were measured biochemically. The results of metabolomic analysis showed that swimming induced a significant change in serum metabolic profile. Relative increases in the levels of lactate, alanine, low-density lipoprotein-very low-density lipoprotein, and unsaturated fatty acids, and decreases in choline, phosphocholine, and glucose were observed after swimming. With quercetin supplementation, these changes were attenuated. The results of biochemical assays were consistent with the data obtained from metabolomic analysis, in that serum NEFA was increased while lactate and NPN decreased after exposed to quercetin in swimming mice. Similar change in NEFA was also found in liver and gastrocnemius muscle tissues. Our current findings suggest that quercetin alters energy metabolism in swimming mice and increased lipolysis may contribute to the actions of quercetin on physical endurance.

Xia, F., Zhong, Y., Li, M., Chang, Q., Liao, Y., Liu, X., & Pan, R. (2015). Antioxidant and Anti-Fatigue Constituents of Okra. *Nutrients*, *7*(10), 8846-8858. doi:10.3390/nu7105435

Okra (Abelmoschus esculentus (L.) Moench), a healthy vegetable, is widely spread in tropical and subtropical areas. Previous studies have proven that okra pods possess anti-fatigue activity, and the aim of this research is to clarify the anti-fatigue constituents. To achieve this, we divided okra pods (OPD) into seeds (OSD) and skins (OSK), and compared the contents of total polysaccharides, total polyphenols, total flavonoids, isoquercitrin, and quercetin-3-O-gentiobiose and the antioxidant activity in vitro and anti-fatigue activity in vivo between OSD and OSK. The contents of total polyphenols and total polysaccharides were 29.5% and 14.8% in OSD and 1.25% and 43.1% in OSK, respectively. Total flavonoids, isoquercitrin and quercetin-3-O-gentiobiose (5.35%, 2.067% and 2.741%, respectively) were only detected in OSD. Antioxidant assays, including 1-diphenyl-2-picrylhydrazyl (DPPH) scavenging, ferric reducing antioxidant power (FRAP) and reducing power test, and weight-loaded swimming test showed OSD possessed significant antioxidant and anti-fatigue effects. Moreover, biochemical determination revealed that that anti-fatigue activity of OSD is caused by reducing the levels of blood lactic acid (BLA) and urea nitrogen (BUN), enhancing hepatic glycogen storage and promoting antioxidant ability by lowering malondialdehyde (MDA) level and increasing superoxide dismutase (SOD) and glutathione peroxidase (GSH-PX) levels. These results proved okra seeds were the anti-fatigue part of okra pods and polyphenols and flavonoids were active constituents.

Ye, M., Liu, Z., Lou, S., Chen, Z., Yu, A., Liu, C., & ... Zhang, J. (2015). Flos Albiziae aqueous extract and its active constituent quercetin potentiate the hypnotic effect of pentobarbital via the serotonergic system. *Biomedical Reports*, *3*(6), 835-838.

Flos albiziae (FA) is reportedly used for treatment of insomnia and anxiety in traditional medicine. The hypnotic effect of an extract of FA (FAE) and its constituent quercetin [2-(3,4-dihydroxyphenyl)-3,5,7-trihydroxy-4H-chromen-4-one, QR] was examined in mice. QR is a widely distributed natural flavonoid abundant in FA flowers and other tissues. The possible mechanisms underlying the hypnotic effects of FAE and QR were investigated using behavioral pharmacology. FAE and QR significantly potentiated pentobarbital-induced [50 mg/kg, intraperitoneal (ip)] sleep (prolonged sleeping time; shortened sleep latency) in a dose-dependent manner, and these effects were augmented by administration of 5-hydroxytryptophan (5-HTP), a precursor of 5-hydroxytryptamine. With a sub-hypnotic dose of pentobarbital (28 mg/kg, ip), FAE and QR significantly increased the rate of sleep onset and were synergistic with 5-HTP (2.5 mg/kg, ip). Pretreatment with p-chlorophenylalanine, an inhibitor of tryptophan hydroxylase, significantly decreased sleeping time and prolonged sleep latency in pentobarbital-treated mice, whereas FAE and QR significantly reversed this effect. Data show that FAE and QR have hypnotic activity, possibly mediated by the serotonergic system. The present study offers a rationale for the use of FA in treating sleep disorders associated with serotonin system dysfunction.

Choline Bitartrate

Borges, A. A., EI-Batah, P. N., Yamashita, L. F., Santana, A. S., Lopes, A. C., Freymuller-Haapalainen, E., & ... Sinigaglia-Coimbra, R. (2015). Neuroprotective effect of oral choline administration after global brain ischemia in rats. *Nutritional Neuroscience*, *18*(6), 265-274. doi:10.1179/1476830514Y.0000000125

Choline - now recognized as an essential nutrient - is the most common polar group found in the outer leaflet of the plasma membrane bilayer. Brain ischemia-reperfusion causes lipid peroxidation triggering multiple cell death pathways involving necrosis and apoptosis. Membrane breakdown is, therefore, a major pathophysiologic event in brain ischemia. The ability to achieve membrane repair is a critical step for survival of ischemic neurons following reperfusion injury. The availability of choline is a rate-limiting factor in phospholipid synthesis and, therefore, may be important for timely membrane repair and cell survival. This work aimed at verifying the effects of 7-day oral administration with different doses of choline on survival of CA1 hippocampal neurons following transient global forebrain ischemia in rats. The administration of 400 mg/kg/day divided into two daily doses for 7 consecutive days significantly improved CA1 pyramidal cell survival, indicating that the local availability of this essential nutrient may limit postischemic neuronal survival.

Glenn, M. L. (2007). Prenatal choline availability modulates hippocampal neurogenesis and neurogenic responses to enriching experiences in adult female rats. *European Journal of Neuroscience*, *25*(8), 2473-2482.

Increased dietary intake of choline early in life improves performance of adult rats on memory tasks and prevents their age-related memory decline. Because neurogenesis in the adult hippocampus also declines with age, we investigated whether prenatal choline availability affects hippocampal neurogenesis in adult Sprague–Dawley rats and modifies their neurogenic response to environmental stimulation. On embryonic days (ED) 12–17, pregnant rats ate a choline-supplemented (SUP-5 g/kg), choline sufficient (SFF-1.1 g/kg), or choline-free (DEF) semisynthetic diet. Adult offspring either remained in standard housing or were given 21 daily visits to explore a maze. On the last ten exploration days, all rats received daily injections of 5-bromo-2-deoxyuridine (BrdU, 100 mg/kg). The number of BrdU+ cells was significantly greater in the dentate gyrus in SUP rats compared to SFF or DEF rats. While maze experience increased the number of BrdU+ cells in SFF rats to the level seen in the SUP rats, this enriching experience did not alter cell proliferation in DEF rats. Similar patterns of cell proliferation were obtained with immunohistochemical staining for neuronal marker doublecortin, confirming that diet and exploration affected hippocampal neurogenesis. Moreover, hippocampal levels of the brain-derived neurotrophic factor (BDNF) were increased in SUP rats as compared to SFF and DEF animals. We conclude that prenatal choline intake has enduring effects on adult hippocampal neurogenesis, possibly via up-regulation of BDNF levels, and suggest that these alterations of neurogenesis may contribute to the mechanism of life-long changes in cognitive function governed by the availability of choline during gestation.

Hoffman, J. R., Ratamess, N. A., & Gonzalez, A. (2010). The effects of acute and prolonged CRAM supplementation on reaction time and subjective measures of focus and alertness in healthy college students. *Journal of the International Society of Sports Nutrition*, B1-B8.

Background: The purpose of this study was to examine the effect of acute and prolonged (4-weeks) ingestion of a supplement designed to improve reaction time and subjective measures of alertness, energy, fatigue, and focus compared to placebo. Methods: Nineteen physically-active subjects (17 men and 2 women) were randomly assigned to a group that either consumed a supplement (21.1 ± 0.6 years; body mass: 80.6 ± 9.4 kg) or placebo (21.3 ± 0.8 years; body mass: 83.4 ± 18.5 kg). During the initial testing session (T1), subjects were provided 1.5 g of the supplement (CRAM; α-glycerophosphocholine, choline bitartrate, phosphatidylserine, vitamins B3, B6, and B12, folic acid, L-tyrosine, anhydrous caffeine, acetyl-L-carnitine, and naringin) or a placebo (PL), and rested quietly for 10-minutes before completing a questionnaire on subjective feelings of energy, fatigue, alertness and focus (PRE). Subjects then performed a 4-minute quickness and reaction test followed by a 10-min bout of exhaustive exercise. The questionnaire and reaction testing sequence was then repeated (POST). Subjects reported back to the lab (T2) following 4-weeks of supplementation and repeated the testing sequence. Results: Reaction time significantly declined ($p = 0.050$) between PRE and POST at T1 in subjects consuming PL, while subjects under CRAM supplementation were able to maintain ($p = 0.114$) their performance. Significant performance declines were seen in both groups from PRE to POST at T2. Elevations in fatigue were seen for CRAM at both T1 and T2 ($p = 0.001$ and $p = 0.000$, respectively), but only at T2 for PL ($p = 0.029$). Subjects in CRAM maintained focus between PRE and POST during both T1 and T2 trials ($p = 0.152$ and $p = 0.082$, respectively), whereas significant declines in focus were observed between PRE and POST in PL at both trials ($p = 0.037$ and $p = 0.014$, respectively). No difference in alertness was seen at T1 between PRE and POST for CRAM ($p = 0.083$), but a significant decline was recorded at T2 ($p = 0.005$). Alertness was significantly lower at POST at both T1 and T2 for PL ($p = 0.040$ and $p = 0.33$, respectively). No differences in any of these subjective measures were seen between the groups at any time point. Conclusion: Results indicate that acute ingestion of CRAM can maintain reaction time, and subjective feelings of focus and alertness to both visual and auditory stimuli in healthy college students following exhaustive exercise. However, some habituation may occur following 4-weeks of supplementation.

Turmeric Root Extract

Bengmark, S., Mesa, M. D., & Gil, A. (2009). Plant-derived health - the effects of turmeric and curcuminoids. *Nutricion Hospitalaria*, *24*(3), 273-281.

Plants contain numerous polyphenols, which have been shown to reduce inflammation and hereby to increase resistance to disease. Examples of such polyphenols are isothiocyanates in cabbage and broccoli, epigallocatechin in green tee, capsaicin in chili peppers, chalones, rutin and naringenin in apples, resveratrol in red wine and fresh peanuts and curcumin/curcuminoids in turmeric. Most diseases are maintained by a sustained discreet but obvious increased systemic inflammation. Many studies suggest that the effect of treatment can be improved by a combination of restriction in intake of proinflammatory molecules such as advanced glycation end products (AGE), advanced lipoperoxidation end products (ALE), and rich supply of antiinflammatory molecules such as plant polyphenols. To the polyphenols with a bulk of experimental documentation belong the curcuminoid family and especially its main ingredient, curcumin. This review summarizes the present knowledge about these turmeric-derived ingredients, which have proven to be strong antioxidants and inhibitors of cyclooxigenase-2 (COX-2), lipoxygenase (LOX) and nuclear factor κ B (NF-κB) but also AGE. A plethora of clinical effects are reported in various experimental diseases, but clinical studies in humans are few. It is suggested that supply of polyphenols and particularly curcuminoids might be value as complement to pharmaceutical treatment, but also prebiotic treatment, in conditions proven to be rather therapy-resistant such as Crohn's, long-stayed patients in intensive care units, but also in conditions such as cancer, liver cirrhosis, chronic renal disease, chronic obstructive lung disease, diabetes and Alzheimer's disease.

Bundy, R., Walker, A. F., Middleton, R. W., & Booth, J. (2004). Turmeric Extract May Improve Irritable Bowel Syndrome Symptomology in Otherwise Healthy Adults: A Pilot Study. *Journal of Alternative & Complementary Medicine*, *10*(6), 1015-1018.

To assess the effects of turmeric () extract on irritable bowel syndrome (IBS) symptomology in otherwise healthy adults. Partially blinded, randomized, two-dose, pilot study. Five hundred (500) volunteers were screened for IBS using the Rome II criteria. Two hundred and seven (207) suitable volunteers were randomized. One or two tablets of a standardized turmeric extract taken daily for 8 weeks. IBS prevalence, symptom-related quality of life (IBSQOL) and self-reported effectiveness. IBS prevalence decreased significantly in both groups between screening and baseline (41% and 57%), with a further significant drop of 53% and 60% between baseline and after treatment, in the one- and two-tablet groups respectively (<0.001). A post-study analysis revealed abdominal pain/discomfort score reduced significantly by 22% and 25% in the one- and two-tablet group respectively, the difference tending toward significance (= 0.071). There were significant improvements in all bar one of the IBSQOL scales of between 5% and 36% in both groups, approximately two thirds of all subjects reported an improvement in symptoms after treatment, and there was a favorable shift in self-reported bowel pattern. There were no significant differences between groups. Turmeric may help reduce IBS symptomology. Placebo controlled trials are now warranted to confirm these findings.

Conrozier, T., Mathieu, P., Bonjean, M., Marc, J., Renevier, J., & Balblanc, J. (2014). A Complex of Three Natural Anti-inflammatory Agents Provides Relief of Osteoarthritis Pain. *Alternative Therapies in Health & Medicine*, *20*(Supp 1), 32-37.

Background * Devil's claw (Harpagophytum procumbens), turmeric (Curcuma longa), and bromelain are nutraceuticals that have demonstrated anti-inflammatory and analgesic properties and may be potential solutions in the treatment of acute or chronic joint pain. Their analgesic effect, however, is generally considered mild to moderate, and the relevance of their clinical use remains subject to discussion. Objectives * The aim of the study was to evaluate the clinical relevance of the efficacy of a marketed complex of 3 plant extracts--H procumbens, C longa, and bromelain (AINAT, 650 mg)--in the treatment of degenerative joint pain. Methods * A multicenter, observational, prospective, open-label survey was conducted in 8 rheumatology centers. The study included 2 groups, 1 group with participants suffering from chronic osteoarthritis (OA) pain and 1 group suffering from acute OA pain. Setting * The research team carried out the study under daily practice conditions. Participants * A total of 42 patients (36 women; mean age = 67 y) suffering from acute or chronic, degenerative spine or joint pain participated. Intervention * Two 650-mg capsules of AINAT were administered 3 x/d to patients with acute pain and 2 x/d to patients with chronic pain. Outcome Measures * At baseline, and during a follow-up visit at 15 d for the acute pain group and 60 d for the chronic pain group, the research team obtained each participant' s global assessment (PGA) and each rheumatologist's global assessment (RGA), as well as each participant's pain score, using for each of them a 100-mm visual analogue scale (VAS). The clinical relevance of the efficacy was evaluated by comparing the outcome measures at endpoint to the values defining the patient acceptable symptom state (PASS) and by comparing the variations (in mm and %) between baseline and endpoint to those defining the minimal clinically important improvement (MCII). Tolerance was also assessed by collecting adverse events at each visit and by using a 4-point scale (very good to bad) at the endpoint. Results * At baseline, the VAS pain score (standard deviation) was 69.1 mm (15.4) and 68.0 mm (18.2) for patients with acute and chronic pain, respectively. At the endpoint, the scores decreased to 42.1 mm (21.1) and 37.8 mm (25.9), respectively. his reduction of pain, as a percentage as well as an absolute value, corresponds to the required definition of MCII, particularly in patients with chronic joint pain. At the endpoint, most of the patients in both groups reached the level of pain defined as the PASS. No withdrawals occurred due to treatment side effects. Conclusion * The improvement of joint pain was clinically relevant in patients treated with AINAT for both acute and chronic OA pain. Considering its excellent tolerance profile, the tested complex of 3 plant extracts with anti-inflammatory properties may be a valuable and safe alternative to NSAIDs in patients suffering from degenerative joint diseases.

Gupta, S. C., Kismali, G., & Aggarwal, B. B. (2013). Curcumin, a component of turmeric: From farm to pharmacy. *Biofactors*, *39*(1), 2-13. doi:10.1002/biof.1079

Curcumin, an active polyphenol of the golden spice turmeric, is a highly pleiotropic molecule with the potential to modulate the biological activity of a number of signaling molecules. Traditionally, this polyphenol has been used in Asian countries to treat such human ailments as acne, psoriasis, dermatitis, and rash. Recent studies have indicated that curcumin can target newly identified signaling pathways including those associated with microRNA, cancer stem cells, and autophagy. Extensive research from preclinical and

clinical studies has delineated the molecular basis for the pharmaceutical uses of this polyphenol against cancer, pulmonary diseases, neurological diseases, liver diseases, metabolic diseases, autoimmune diseases, cardiovascular diseases, and numerous other chronic diseases. Multiple studies have indicated the safety and efficacy of curcumin in numerous animals including rodents, monkeys, horses, rabbits, and cats and have provided a solid basis for evaluating its safety and efficacy in humans. To date, more than 65 human clinical trials of curcumin, which included more than 1000 patients, have been completed, and as many as 35 clinical trials are underway. Curcumin is now used as a supplement in several countries including the United States, India, Japan, Korea, Thailand, China, Turkey, South Africa, Nepal, and Pakistan. In this review, we provide evidence for the pharmaceutical uses of curcumin for various diseases.

Huang, W., Chiu, W., Chuang, H., Tang, D., Lee, Z., Wei, L., & ... Huang, C. (2015). Effect of curcumin supplementation on physiological fatigue and physical performance in mice. *Nutrients*, *7*(2), 905-921. doi:10.3390/nu7020905

Curcumin (CCM) is a well-known phytocompound and food component found in the spice turmeric and has multifunctional bioactivities. However, few studies have examined its effects on exercise performance and physical fatigue. We aimed to evaluate the potential beneficial effects of CCM supplementation on fatigue and ergogenic function following physical challenge in mice. Male ICR mice were divided into four groups to receive vehicle or CCM (180 μg/mL) by oral gavage at 0, 12.3, 24.6, or 61.5 mL/kg/day for four weeks. Exercise performance and anti-fatigue function were evaluated after physical challenge by forelimb grip strength, exhaustive swimming time, and levels of physical fatigue-associated biomarkers serum lactate, ammonia, blood urea nitrogen (BUN), and glucose and tissue damage markers such as aspartate transaminase (AST), alanine transaminase (ALT), and creatine kinase (CK). CCM supplementation dose-dependently increased grip strength and endurance performance and significantly decreased lactate, ammonia, BUN, AST, ALT, and CK levels after physical challenge. Muscular glycogen content, an important energy source for exercise, was significantly increased. CCM supplementation had few subchronic toxic effects. CCM supplementation may have a wide spectrum of bioactivities for promoting health, improving exercise performance and preventing fatigue.

Jin, X., Zhang, Y., Li, Q., & Zhao, J. (2013). Mechanisms underlying the beneficial effects of Kaiyu Granule for depression. *Neural Regeneration Research*, *8*(34), 3241-3248.

The proprietary Chinese medicine preparation Kaiyu Granule is made of bupleurum, nutgrass galingale rhizome, szechwan lovage rhizome, turmeric root tuber, white peony alba, cape jasmine fruit, fried semen ziziphi jujubae, and prepared liquorice root. It is a common recipe for the clinical treatment of depression in China. In this study, after 21 days of unpredictable stress exposure, Wistar rats exhibited similar behavioral changes to patients with depression. Moreover, G-protein-coupled inwardly rectifying K(+) channel 1 mRNA and protein expression were significantly reduced in rat hippocampal CA1 and CA3 regions. However, G-protein-coupled inwardly rectifying K(+) channel 1 mRNA, protein expression, and rat behavior were clearly better after administration of 12, 8, or 4 g/kg of Kaiyu Granule when depression model rats underwent stress. 12 g/kg of Kaiyu Granule had the most obvious effects on the increased expression of G-protein-

coupled inwardly rectifying K(+) channel 1 mRNA and protein in rat hippocampal CA1 and CA3 regions. These results suggested that Kaiyu Granule improved depression by affecting G-protein-coupled inwardly rectifying K(+) channel 1 expression in the rat hippocampus.

Katsidoni, V. G. (2014). Curcumin, demethoxycurcumin and bisdemethoxycurcumin differentially inhibit morphine's rewarding effect in rats. *Psychopharmacology*, *231*(23), 4467-4478.

Rationale: Recent animal studies reported that curcumin, the active constituent of Curcuma longa, has several central actions and may attenuate morphine tolerance. Objectives: In the present study, we utilized the intracranial self-stimulation (ICSS) paradigm to examine the effects of the commercially available curcuminoid mixture and each one of its components, individually, on brain stimulation reward and on the reward-facilitating effect of morphine. Methods: Male Sprague-Dawley rats were implanted with an electrode into the medial forebrain bundle and trained to respond for electrical stimulation using a rate-frequency paradigm. In the first study, rats were injected with graded doses either of the curcuminoid mixture, or curcumin I, or II, or III. In the second study, we examined whether a low dose of the curcuminoid mixture or each individual curcumin analogue composing it could counteract the reward-facilitating effect of morphine. Results: At low doses, both the curcuminoid mixture and curcumin I did not affect brain stimulation reward, whereas, higher doses increased ICSS thresholds. Curcumin II and curcumin III did not affect brain stimulation reward at any doses. Subthreshold doses of the curcuminoid mixture and curcumin I inhibited the reward-facilitating effect of morphine. Conclusion: Both the curcuminoid mixture and curcumin I lack hedonic properties and moderate the reward-facilitating effect of morphine. Our data suggest that curcumin interferes with brain reward mechanisms responsible for the expression of the acute reinforcing properties of opioids and provide evidence that curcumin may be a promising adjuvant for attenuating morphine's rewarding effects in patients who are under long-term opioid therapy.

Kulkarni, S. K., & Dhir, A. (2010). An Overview of Curcumin in Neurological Disorders. *Indian Journal of Pharmaceutical Sciences*, *72*(2), 149-154.

Curcumin, the principal curcuminoid found in spice turmeric, has recently been studied for its active role in the treatment of various central nervous system disorders. Curcumin demonstrates neuroprotective action in Alzheimer's disease, tardive dyskinesia, major depression, epilepsy, and other related neurodegenerative and neuropsychiatric disorders. The mechanism of its neuroprotective action is not completely understood. However, it has been hypothesized to act majorly through its anti-inflammatory and antioxidant properties. Also, it is a potent inhibitor of reactive astrocyte expression and thus prevents cell death. Curcumin also modulates various neurotransmitter levels in the brain. The present review attempts to discuss some of the potential protective role of curcumin in animal models of major depression, tardive dyskinesia and diabetic neuropathy. These studies call for well-planned clinical studies on curcumin for its potential use in neurological disorders.

Lakhan, S. E., Ford, C. T., & Tepper, D. (2015). Zingiberaceae extracts for pain: a systematic review and meta-analysis. *Nutrition Journal*, *14*50. doi:10.1186/s12937-015-0038-8

Background: Members of the family Zingiberaceae including turmeric, ginger, Javanese ginger, and galangal have been used for centuries in traditional medicine. Preclinical studies of Zingiberaceae extracts have shown analgesic properties. This study aims to systematically review and meta-analyze whether extracts from Zingiberaceae are clinically effective hypoalgesic agents. Methods: Literature was screened from electronic databases using the key words Zingiberaceae AND pain OR visual analogue score (VAS) to identify randomized trials. From this search, 18 studies were identified, and of these, 8 randomized, double-blinded, placebo-controlled trials were found that measured pain by VAS for inclusion in the meta-analysis. Results: Findings indicated significant efficacy of Zingiberaceae extracts in reducing subjective chronic pain (SMD - 0.67; 95 % CI - 1.13 to - 0.21; P = 0.004). A linear dose-effect relationship was apparent between studies (R(2) = 0.71). All studies included in the systematic review reported a good safety profile for extracts, without the renal risks associated with non-steroidal anti-inflammatory drugs, and with similar effectiveness. Conclusion: Our findings indicated that Zingiberaceae extracts are clinically effective hypoalgesic agents and the available data show a better safety profile than non-steroidal anti-inflammatory drugs. However, both non-steroidal anti-inflammatory drugs and Zingiberaceae have been associated with a heightened bleeding risk, and there have been no comparator trials of this risk. Further clinical studies are recommended to identify the most effective type of Zingiberaceae extract and rigorously compare safety, including bleeding risk.

Mishra, S., & Palanivelu, K. (2008). The effect of curcumin (turmeric) on Alzheimer's disease: An overview. *Annals of Indian Academy of Neurology*, *11*(1), 13-19.

This paper discusses the effects of curcumin on patients with Alzheimer's disease (AD). Curcumin (Turmeric), an ancient Indian herb used in curry powder, has been extensively studied in modern medicine and Indian systems of medicine for the treatment of various medical conditions, including cystic fibrosis, hemorrhoids, gastric ulcer, colon cancer, breast cancer, atherosclerosis, liver diseases and arthritis. It has been used in various types of treatments for dementia and traumatic brain injury. Curcumin also has a potential role in the prevention and treatment of AD. Curcumin as an antioxidant, anti-inflammatory and lipophilic action improves the cognitive functions in patients with AD. A growing body of evidence indicates that oxidative stress, free radicals, beta amyloid, cerebral deregulation caused by bio-metal toxicity and abnormal inflammatory reactions contribute to the key event in Alzheimer's disease pathology. Due to various effects of curcumin, such as decreased Beta-amyloid plaques, delayed degradation of neurons, metal-chelation, anti-inflammatory, antioxidant and decreased microglia formation, the overall memory in patients with AD has improved. This paper reviews the various mechanisms of actions of curcumin in AD and pathology.

Ng, T., Chiam, P., Lee, T., Chua, H., Lim, L., & Kua, E. (2006). Curry consumption and cognitive function in the elderly. *American Journal of Epidemiology*, *164*(9), 898-906 9p. doi:aje/kwj267

Curcumin, from the curry spice turmeric, has been shown to possess potent antioxidant and antiinflammatory properties and to reduce beta-amyloid and plaque burden in experimental studies, but epidemiologic evidence is lacking. The authors investigated the association between usual curry consumption level and cognitive function in elderly Asians. In a population-based cohort (n = 1,010) of non-demented elderly Asian subjects aged 60-93 years in 2003, the authors compared Mini-Mental State Examination (MMSE) scores for three categories of regular curry consumption, taking into account known sociodemographic, health, and behavioral correlates of MMSE performance. Those who consumed curry 'occasionally' and 'often or very often' had significantly better MMSE scores than did subjects who 'never or rarely' consumed curry. The authors reported tentative evidence of better cognitive performance from curry consumption in non-demented elderly Asians, which should be confirmed in future studies.

Nieman, D., Shanely, R., Luo, B., Dew, D., Meaney, M., & Sha, W. (2013). A commercialized dietary supplement alleviates joint pain in community adults: a double-blind, placebo-controlled community trial. *Nutrition Journal*, *12*(1), 1-19.

Background: The purpose of this study was to assess the effect of 8-weeks ingestion of a commercialized joint pain dietary supplement (InstaflexTM Joint Support, Direct Digital, Charlotte, NC) compared to placebo on joint pain, stiffness, and function in adults with self-reported joint pain. InstaflexTM is a joint pain supplement containing glucosamine sulfate, methylsufonlylmethane (MSM), white willow bark extract (15% salicin), ginger root concentrate, boswella serrata extract (65% boswellic acid), turmeric root extract, cayenne, and hyaluronic acid. Methods: Subjects included 100 men and women, ages 50-75 years, with a history (>3 months) of joint pain, and were randomized to InstaflexTM or placebo (3 colored gel capsules per day for 8 weeks, double-blind administration). Subjects agreed to avoid the use of non-steroidal antiinflammatory drugs (NSAID) and all other medications and supplements targeted for joint pain. Primary outcome measures were obtained pre- and post-study and included joint pain severity, stiffness, and function (Western Ontario and McMaster Universities [WOMAC]), and secondary outcome measures included health-related quality of life (Short Form 36 or SF-36), systemic inflammation (serum C-reactive protein and 9 plasma cytokines), and physical function (6-minute walk test). Joint pain symptom severity was assessed bi-weekly using a 12-point Likert visual scale (12-VS). Results: Joint pain severity was significantly reduced in InstaflexTM compared to placebo (8-week WOMAC, ?37% versus ?16%, respectively, interaction effect P = 0.025), with group differences using the 12-VS emerging by week 4 of the study (interaction effect, P = 0.0125). Improvements in ability to perform daily activities and stiffness scores in InstaflexTM compared to placebo were most evident for the 74% of subjects reporting knee pain (8-week WOMAC function score, ?39% versus ?14%, respectively, interaction effect P = 0.027; stiffness score, ?30% versus ?12%, respectively, interaction effect P = 0.081). Patterns of change in SF-36, systemic inflammation biomarkers, and the 6-minute walk test did not differ significantly between groups during the 8-week study Conclusions: Results from this randomized, double blind, placebo-controlled community trial support the use of the InstaflexTM dietary supplement in alleviating joint pain severity in middle-aged and older adults, with mitigation of difficulty performing daily activities most apparent in subjects with knee pain.

Nigam, D., Rani, V., & Singh, K. (2012). Protective Role of Turmeric in Manganese-Induced Oxidative Alterations in Rat Brain. *Journal of Pure & Applied Science & Technology*, *11*(1), 5-11.

Turmeric powder obtained from the rhizomes of Curcuma longa Linn., has been traditionally recognized for treatment of several diseases. Overexposure to manganese (Mn) results in a neurological disorder, termed manganism which shares a similar phenotype to Parkinson's disease. The present study explores the protective effect of turmeric against the toxicity of manganese (Mn) in adult albino male rat brain. Rats were divided into four groups. Group I rats served as control. Group II rats received turmeric (1g/kg body weight/day, orally) for 45 days. Group III rats were received Mn as MnCl2 (8mg/Kg body weight/day), intraperitoneally for 15 days. Group IV rats were orally received turmeric for 45 days. Besides turmeric, group IV rats were also received Mn as MnCl2 (8mg/Kg body weight/day, intraperitoneally) for last 15 days. Levels of reactive oxygen species, lipid peroxidation potential, conjugated dienes, blood-brain barrier permeability, glutathione reductase and glutathione peroxidase were significantly increased, however lower levels of superoxide dismutase, reduced glutathione and membrane fluidity were observed in brain of group III. There was no change found in the activity of catalase in brain of any of the experimental groups. These changes were ameliorated in group IV. The study suggests that turmeric exhibits neuroprotection against free radical-mediated neurotoxicity of Mn.

Nozomi, H., Yoriko, T., Yoshinobu, A., Yuhei, T., Yoshitake, T., Hisayoshi, N., & ... Krishna U., K. (2013). Effects of turmeric on Alzheimer's disease with behavioral and psychological symptoms of dementia. *AYU: An International Quarterly Journal of Research in Ayurveda*, *34*(4), 499-504.

We describe here three patients with the Alzheimer's Disease (AD) whose behavioral symptoms were improved remarkably as a result of the turmeric treatment, which is the traditional Indian medicine. Their cognitive decline and Behavioral and Psychological Symptoms of Dementia (BPSD) were very severe. All three patients exhibited irritability, agitation, anxiety, and apathy, two patients suffer from urinary incontinence and wonderings. They were prescribed turmeric powder capsules and started recovering from these symptoms without any adverse reaction in the clinical symptom and laboratory data. After 12 weeks of the treatment, total score of the Neuro-Psychiatric Inventory-brief questionnaire decreased significantly in both acuities of symptoms and burden of caregivers. In one case, the Mini-Mental State Examination (MMSE) score was up five points, from 12/30 to 17/30. In the other two cases, no significant change was seen in the MMSE; however, they came to recognize their family within 1-year treatment. All cases have been taking turmeric for more than 1 year, re-exacerbation of BPSD was not seen. The present cases suggest a significant improvement of the behavioral symptoms in the AD with the turmeric treatment, leading to probable benefit of the use of turmeric in individuals with the AD with BPSD.

Sarlak, Z., Oryan, S., & Moghaddasi, M. (2015). Interaction between the antioxidant activity of curcumin and cholinergic system on memory retention in adult male Wistar rats. *Iranian Journal of Basic Medical Sciences*, *18*(4), 398-403.

Objective(s): The cholinergic system plays an important role in learning and memory. This study investigated the effects of curcumin (turmeric extract) and the cholinergic system and their interaction on memory retention of passive avoidance learning in adult male Wistar rats. Materials and Methods: At first, an injection cannula was implanted in right ventricles of the animals. One week after the surgery, the animals were trained with a shuttle box set up. Post-training, injections were performed in all experiments. Administration of curcumin increased memory retention. Also administrations of nicotine and pilocarpine, the cholinergic receptor agonists, increased memory retention, while it is decreased by succinylcholine and scopolamine, the cholinergic receptor antagonists. Then co-administration of curcumin and cholinergic drugs were performed. Intraperitoneal and intracerebroventricular injections were applied for the curcumin and cholinergic drugs, respectively. Results: Co-administration of curcumin (45 mg/kg) with a low dose of nicotine (0.1 µg/rat) or pilocarpine (0.5 µg/rat) increased memory retention significantly. Effects of succinylcholine (0.01, 0.1 and 0.5 µg/rat) or scopolamine (0.1, 1 and 5 µg/rat) were attenuated by curcumin markedly (45 mg/kg). Conclusion: The results suggest that curcumin has a close interaction with cholinergic system in memory retention process.

Sikora, E., Scapagnini, G., & Barbagallo, M. (2010). Curcumin, inflammation, ageing and age-related diseases. *Immunity & Ageing*, *7*1-4. doi:10.1186/1742-4933-7-1

A Symposium regarding the Pathophysiology of Successful and Unsuccessful Ageing was held in Palermo, Italy between April 7 and 8th 2009. Here the lecture by Sikora with some input from the chairpersons Scapagnini and Barbagallo is summarized. Ageing is manifested by the decreasing health status and increasing probability to acquire age-related disease such as cancer, Alzheimer's disease, atherosclerosis, metabolic disorders and others. They are likely caused by low grade inflammation driven by oxygen stress and manifested by the increased level of pro-inflammatory cytokines such as IL-1, IL-6 and TNF-α, encoded by genes activated by the transcription factor NF-κB. It is believed that ageing is plastic and can be slowed down by caloric restriction as well as by some nutraceuticals. Accordingly, slowing down ageing and postponing the onset of age-related diseases might be achieved by blocking the NF-κB-dependent inflammation. In this review we consider the possibility of the spice curcumin, a powerful antioxidant and anti-inflammatory agent possibly capable of improving the health status of the elderly.

Stankowska, D. L., Krishnamoorthy, V. R., Ellis, D. Z., & Krishnamoorthy, R. R. (2015). Neuroprotective effects of curcumin on endothelin-1 mediated cell death in hippocampal neurons. *Nutritional Neuroscience*.

Alzheimer's disease is a progressive neurodegenerative disease characterized by loss of hippocampal neurons leading to memory deficits and cognitive decline. Studies suggest that levels of the vasoactive peptide endothelin-1 (ET-1) are increased in the brain tissue of Alzheimer's patients. Curcumin, the main ingredient of the spice turmeric, has been shown to have anti-inflammatory, anti-cancer, and

neuroprotective effects. However, the mechanisms underlying some of these beneficial effects are not completely understood. The objective of this study was to determine if curcumin could protect hippocampal neurons from ET-1 mediated cell death and examine the involvement of c-Jun in this pathway. Methods Primary hippocampal neurons from rat pups were isolated using a previously published protocol. Viability of the cells was measured by the live/dead assay. Immunoblot and immunohistochemical analyses were performed to analyze c-Jun levels in hippocampal neurons treated with either ET-1 or a combination of ET-1 and curcumin. Apoptotic changes were evaluated by immunoblot detection of cleaved caspase-3, cleaved fodrin, and a caspase 3/7 activation assay. Results ET-1 treatment produced a 2-fold increase in the levels of c-Jun as determined by an immunoblot analysis in hippocampal neurons. Co-treatment with curcumin significantly attenuated the ET-1 mediated increase in c-Jun levels. ET-1 caused increased neuronal cell death of hippocampal neurons indicated by elevation of cleaved caspase-3, cleaved fodrin and an increased activity of caspases 3 and 7 which was attenuated by co-treatment with curcumin. Blockade of JNK, an upstream effector of c-Jun by specific inhibitor SP600125 did not fully protect from ET-1 mediated activation of pro-apoptotic enzymes in primary hippocampal cells. Discussion Our data suggests that one mechanism by which curcumin protects against ET-1-mediated cell death is through blocking an increase in c-Jun levels. Other possible mechanisms include decreasing pro-apoptotic signaling activated by ET-1 in primary hippocampal neurons. (Full-text unavailable)

Tizabi, Y., Hurley, L. L., Qualls, Z., & Akinfiresoye, L. (2014). Relevance of the Anti-Inflammatory Properties of Curcumin in Neurodegenerative Diseases and Depression. *Molecules*, *19*(12), 20864-20879. doi:10.3390/molecules191220864

This review is an attempt to summarize our current understanding of curcumin's potential as a neuroprotectant and an antidepressant. This dual property confers a unique advantage to this herbal medication, believed to be devoid of any major side effects, to combat commonly observed co-morbid conditions of a neurodegenerative and a neuropsychiatric disorder. Moreover, in line with the theme of this series, the role of inflammation and stress in these diseases and possible anti-inflammatory effects of curcumin, as well as its interaction with signal transduction proteins as a common denominator in its varied mechanisms of action, are also discussed. Thus, following a brief introduction of curcumin's pharmacology, we present research suggesting how its anti-inflammatory properties have therapeutic potential in treating a devastating neurological disorder (Parkinson's disease = PD) and a debilitating neuropsychiatric disorder (major depressive disorder = MDD). It is concluded that curcumin, or better yet, an analog with better and longer bioavailability could be of important therapeutic potential in PD and/or major depression.

Yan, H., Yuan, Y., Xi, Z., Kun, Z., Shaohua, C., & Zhiyun, D. (2015). Curcumin, Inflammation, and Chronic Diseases: How Are They Linked? *Molecules*, *20*(5), 9183-9213. doi:10.3390/molecules20059183

It is extensively verified that continued oxidative stress and oxidative damage may lead to chronic inflammation, which in turn can mediate most chronic diseases including cancer, diabetes, cardiovascular, neurological, inflammatory bowel disease and pulmonary diseases. Curcumin, a yellow coloring agent extracted from turmeric, shows strong anti-oxidative and anti-inflammatory activities when used as a

remedy for the prevention and treatment of chronic diseases. How oxidative stress activates inflammatory pathways leading to the progression of chronic diseases is the focus of this review. Thus, research to date suggests that chronic inflammation, oxidative stress, and most chronic diseases are closely linked, and the antioxidant properties of curcumin can play a key role in the prevention and treatment of chronic inflammation diseases.

Yimam, M., Young-Chul, L., Moore, B., Ping, J., Mei, H., Jeong-Bum, N., & ... Qi, J. (2016). UP1304, a Botanical Composition Containing Two Standardized Extracts of Curcuma longa and Morus alba, Mitigates Pain and Inflammation in Adjuvant-induced Arthritic Rats. *Pharmacognosy Research*, *8*(2), 112-117. doi:10.4103/0974-8490.172563

Background: Though, the initial etiologies of arthritis are multifactorial, clinically, patients share pain as the prime complaints. Present day pain relief therapeutics heavily relies on the use of prescription and over the counter nonsteroidal anti-inflammatory drugs as the first line of defense where their long-term usage causes gastrointestinal and cardiovascular-related side effects. Hence, the need for evidence-based safer and efficacious alternatives from natural sources to overcome the most prominent and disabling symptoms of arthritis is an overdue. Here, we evaluated the anti-inflammatory and analgesic effect of UP1304, a composition that contains a standardized blend of two extracts from the rhizome of Curcuma longa and the root bark of Morus alba in adjuvant-induced arthritis models in rats. Materials and Methods: The anti-inflammatory and analgesic effects of the botanical composition were demonstrated in adjuvant-induced arthritis models in rats with oral dose ranges of 50-200 mg/kg. Ibuprofen at a dose of 100 mg/kg was used as a reference compound. Ex vivo sulfated glycosaminoglycan inhibition assays were performed. Results: Statistically significant improvements in pain resistance, suppression of paw edema and ankle thickness were observed in animals treated with UP1304 compared to vehicle-treated diseased rats. These results were similar to those achieved by ibuprofen treatment. Inhibitions of proteoglycan degradation were observed in a range of 37.5-61.7% for concentration of UP1304 at 50-200 μg/mL when compared to interleukin-1α-exposed untreated explants. Conclusions: These data suggest that UP1304, for its analgesic and anti-inflammatory effects, could potentially be considered agent of botanical origin for the improvement of arthritis associated symptoms.

Zhang, L., Fang, Y., Xu, Y., Lian, Y., Xie, N., Wu, T., & ... Wang, Z. (2015). Curcumin Improves Amyloid β-Peptide (1-42) Induced Spatial Memory Deficits through BDNF-ERK Signaling Pathway. *Plos ONE*, *10*(6), 1-17. doi:10.1371/journal.pone.0131525

Curcumin, the most active component of turmeric, has various beneficial properties, such as antioxidant, anti-inflammatory, and antitumor effects. Previous studies have suggested that curcumin reduces the levels of amyloid and oxidized proteins and prevents memory deficits and thus is beneficial to patients with Alzheimer's disease (AD). However, the molecular mechanisms underlying curcumin's effect on cognitive functions are not well-understood. In the present study, we examined the working memory and spatial reference memory in rats that received a ventricular injection of amyloid-β1-42 (Aβ1-42), representing a rodent model of Alzheimer's disease (AD). The rats treated with Aβ1-42 exhibited obvious cognitive deficits

in behavioral tasks. Chronic (seven consecutive days, once per day) but not acute (once a day) curcumin treatments (50, 100, and 200 mg/kg) improved the cognitive functions in a dose-dependent manner. In addition, the beneficial effect of curcumin is accompanied by increased BDNF levels and elevated levels of phosphorylated ERK in the hippocampus. Furthermore, the cognition enhancement effect of curcumin could be mimicked by the overexpression of BDNF in the hippocampus and blocked by either bilateral hippocampal injections with lentiviruses that express BDNF shRNA or a microinjection of ERK inhibitor. These findings suggest that chronic curcumin ameliorates AD-related cognitive deficits and that upregulated BDNF-ERK signaling in the hippocampus may underlie the cognitive improvement produced by curcumin.

L-Theanine

Advantageous effect of theanine intake on cognition. (2014). *Nutritional Neuroscience*, *17*(6), 279-283.

Theanine, γ-glutamylethylamide, is one of the major amino acid components in green tea. On the basis of the preventive effect of theanine intake after weaning on stress-induced impairment of recognition memory, the advantageous effect of theanine intake on recognition memory was examined in young rats, which were fed water containing 0.3% theanine for 3 weeks after weaning. The rats were subjected to object recognition test. Object recognition memory was maintained in theanine-administered rats 48 hours after the training, but not in the control rats. When in vivo dentate gyrus long-term potentiation (LTP) was induced, it was more greatly induced in theanine-administered rats than in the control rats. The levels of brain-derived neurotropic factor and nerve growth factor in the hippocampus were significantly higher in theanine-administered rats than in the control rats. The present study indicates the advantageous effect of theanine intake after weaning on recognition memory. It is likely that theanine intake is of advantage to the development of hippocampal function after weaning.

Bryan, J. (2008). Psychological effects of dietary components of tea: caffeine and L-theanine. *Nutrition Reviews*, *66*(2), 82-90. doi:10.1111/j.1753-4887.2007.00011.x

This review summarizes the literature on the association between two dietary components of tea, caffeine and L-theanine, and the psychological outcomes of consumption; it also identifies areas for future research. The studies reviewed suggest that caffeinated tea, when ingested at regular intervals, may maintain alertness, focused attention, and accuracy and may modulate the more acute effects of higher doses of caffeine. These findings concur with the neurochemical effects of L-theanine on the brain. L-theanine may interact with caffeine to enhance performance in terms of attention switching and the ability to ignore distraction; this is likely to be reflective of higher-level cognitive activity and may be sensitive to the detrimental effects of overstimulation. Further research should investigate the interactive effects of caffeine, L-theanine, and task complexity, utilize a range of ecologically valid psychological outcomes, and assess the neuroprotective effects of L-theanine using epidemiological or longer-term intervention studies among individuals at risk of neurodegenerative disease.

Camfield, D. A., Stough, C., Farrimond, J., & Scholey, A. B. (2014). Acute effects of tea constituents L-theanine, caffeine, and epigallocatechin gallate on cognitive function and mood: a systematic review and meta-analysis. *Nutrition Reviews*, *72*(8), 507-522. doi:10.1111/nure.12120

A systematic review and meta-analysis was conducted on 11 randomized placebo-controlled human studies of acute effects of tea constituents L-theanine and epigallocatechin gallate, administered alone or in combination with caffeine, on cognitive function and mood. The outcome measures of mood were alertness, calmness, and contentedness, derived from the Bond- Lader scales, and state anxiety, from the State- Trait Anxiety Inventory. Cognitive measures assessed were attentional switch, intersensory attention, and rapid visual information processing. Standardized mean differences between placebo and treatment

groups are presented for each study and outcome measure. Meta-analysis using a random-effects model was conducted when data were available for three or more studies. Evidence of moderate effect sizes in favor of combined caffeine and L-theanine in the first 2 hours post dose were found for outcome measures Bond- Lader alertness, attentional switching accuracy, and, to a lesser extent, some unisensory and multisensory attentional outcomes. Moderator analysis of caffeine and L-theanine doses revealed trends toward greater change in effect size for caffeine dose than for L-theanine dose, particularly during the first hour post dose.

Cooper, R. (2012). Green tea and theanine: health benefits. *International Journal of Food Sciences & Nutrition*, *63*90-97. doi:10.3109/09637486.2011.629180

Historically, the medicinal use of green tea dates back to China 4700 years ago and drinking tea continues to be regarded traditionally in Asia as a general healthful practice. Numerous scientific publications now attest to the health benefits of both black and green teas, including clinical and epidemiological studies. Although all tea contains beneficial antioxidants, high-quality green and white teas have them in greater concentrations than black tea. Today, scientists believe that the main active ingredients of green tea include the polyphenols, in particular the catechins and the amino acid, theanine. Studies on the health benefits of drinking tea, particularly green tea, are finding exciting results, particularly in cancer research. Modern studies in both Asia and the West have provided encouraging results indicating that drinking green tea contributes to fighting many different kinds of cancers including stomach, oesophageal, ovarian and colon. Recent studies describing the health benefits of these compounds will be reviewed.

Giesbrecht, T., Rycroft, J., Rowson, M., & De Bruin, E. (2010). The combination of L-theanine and caffeine improves cognitive performance and increases subjective alertness. *Nutritional Neuroscience*, *13*(6), 283-290. doi:10.1179/147683010X12611460764840

The non-proteinic amino acid L-theanine and caffeine, a methylxanthine derivative, are naturally occurring ingredients in tea. The present study investigated the effect of a combination of 97 mg L-theanine and 40 mg caffeine as compared to placebo treatment on cognitive performance, alertness, blood pressure, and heart rate in a sample of young adults (n = 44). Cognitive performance, self-reported mood, blood pressure, and heart rate were measured before L-theanine and caffeine administration (i.e. at baseline) and 20 min and 70 min thereafter. The combination of moderate levels of L-theanine and caffeine significantly improved accuracy during task switching and self-reported alertness (both $P < 0.01$) and reduced self-reported tiredness ($P < 0.05$). There were no significant effects on other cognitive tasks, such as visual search, choice reaction times, or mental rotation. The present results suggest that 97 mg of L-theanine in combination with 40 mg of caffeine helps to focus attention during a demanding cognitive task.

Lyon, M. R., Kapoor, M. P., & Juneja, L. R. (2011). The Effects of L-Theanine (Suntheanine) on Objective Sleep Quality in Boys with Attention Deficit Hyperactivity Disorder (ADHD): A Randomized, Double-blind, Placebo-controlled Clinical Trial. *Alternative Medicine Review*, *16*(4), 348-354 7p.

The purpose of this study was to investigate the efficacy and safety of L-theanine as an aid to the improvement of objectively measured sleep quality in a population of 98 male children formally diagnosed with attention-deficit/hyperactivity disorder (ADHD). A randomized, double-blind, placebo-controlled trial was conducted involving boys, ages 8-12 years, who had been previously diagnosed with ADHD. An experienced physician confirmed the diagnosis of ADHD in each subject. Randomization was stratified based upon current use of stimulant medication to ensure an equal distribution of stimulant/non-stimulant treated subjects into active and placebo treated groups. Participants consumed two chewable tablets twice daily (at breakfast and after school), with each tablet containing 100mg of L-theanine (total 400 mg daily Suntheanine, Taiyo Kagaku, Yokkaichi, Japan) or identical tasting chewable placebo for six weeks. Subjects were evaluated for five consecutive nights using wrist actigraphy at baseline, and again at the end of the six-week treatment period. The Pediatric Sleep Questionnaire (PSQ) was completed by parents at baseline and at the end of the treatment period. Actigraph watch data findings indicated that boys who consumed L-theanine obtained significantly higher sleep percentage and sleep efficiency scores, along with a non-significant trend for less activity during sleep (defined as less time awake after sleep onset) compared to those in the placebo group. Sleep latency and other sleep parameters were unchanged. The PSQ data did not correlate significantly to the objective data gathered from actigraphy, suggesting that parents were not particularly aware of their children's sleep quality. L-theanine at relatively high doses was well tolerated with no significant adverse events. This study demonstrates that 400 mg daily of L-theanine is safe and effective in improving some aspects of sleep quality in boys diagnosed with ADHD. Since sleep problems are a common co-morbidity associated with ADHD, and because disturbed sleep may be linked etiologically to this disorder, L-theanine may represeni: a safe and important adjunctive therapy in childhood ADIID. Larger, long-term studies looking at the wider therapeutic role of this agent in this population are warranted.

Neurobiological effects of the green tea constituent theanine and its potential role in the treatment of psychiatric and neurodegenerative disorders. (2014). *Nutritional Neuroscience*, *17*(4), 145-155. doi:10.1179/1476830513Y.0000000079

Theanine (n-ethylglutamic acid), a non-proteinaceous amino acid component of green and black teas, has received growing attention in recent years due to its reported effects on the central nervous system. It readily crosses the blood-brain barrier where it exerts a variety of neurophysiological and pharmacological effects. Its most well-documented effect has been its apparent anxiolytic and calming effect due to its up-regulation of inhibitory neurotransmitters and possible modulation of serotonin and dopamine in selected areas. It has also recently been shown to increase levels of brain-derived neurotrophic factor. An increasing number of studies demonstrate a neuroprotective effects following cerebral infarct and injury, although the exact molecular mechanisms remain to be fully elucidated. Theanine also elicits improvements in cognitive function including learning and memory, in human and animal studies, possibly via a decrease in NMDA-dependent CA1 long-term potentiation (LTP) and increase in NMDA-independent CA1-LTP. Furthermore, theanine administration elicits selective changes in alpha brain wave activity with concomitant increases in

selective attention during the execution of mental tasks. Emerging studies also demonstrate a promising role for theanine in augmentation therapy for schizophrenia, while animal models of depression report positive improvements following theanine administration. A handful of studies are beginning to examine a putative role in attention deficit hyperactivity disorder, and theoretical extrapolations to a therapeutic role for theanine in other psychiatric disorders such as anxiety disorders, panic disorder, obsessive compulsive disorder (OCD), and bipolar disorder are discussed.

Owen, G. N., Parnell, H., De Bruin, E. A., & Rycroft, J. A. (2008). The combined effects of L-theanine and caffeine on cognitive performance and mood. *Nutritional Neuroscience*, *11*(4), 193-198. doi:10.1179/147683008X301513

The aim of this study was to compare 50 mg caffeine, with and without 100 mg L-theanine, on cognition and mood in healthy volunteers. The effects of these treatments on word recognition, rapid visual information processing, critical flicker fusion threshold, attention switching and mood were compared to placebo in 27 participants. Performance was measured at baseline and again 60 min and 90 min after each treatment (separated by a 7-day washout). Caffeine improved subjective alertness at 60 min and accuracy on the attention-switching task at 90 min. The L-theanine and caffeine combination improved both speed and accuracy of performance of the attention-switching task at 60 min, and reduced susceptibility to distracting information in the memory task at both 60 min and 90 min. These results replicate previous evidence which suggests that L-theanine and caffeine in combination are beneficial for improving performance on cognitively demanding tasks.

Park, S., Jung, I., Lee, W. K., Lee, Y. S., Park, H. K., Go, H. J., & ... Rho, S. S. (2011). A combination of green tea extract and L-theanine improves memory and attention in subjects with mild cognitive impairment: a double-blind placebo-controlled study. *Journal of Medicinal Food*,*14*(4), 334-343 10p. doi:10.1089/jmf.2009.1374

A combination of green tea extract and l-theanine (LGNC-07) has been reported to have beneficial effects on cognition in animal studies. In this randomized, double-blind, placebo-controlled study, the effect of LGNC-07 on memory and attention in subjects with mild cognitive impairment (MCI) was investigated. Ninety-one MCI subjects whose Mini Mental State Examination-K (MMSE-K) scores were between 21 and 26 and who were in either stage 2 or 3 on the Global Deterioration Scale were enrolled in this study. The treatment group (13 men, 32 women; 57.58 ± 9.45 years) took 1,680 mg of LGNC-07, and the placebo group (12 men, 34 women; 56.28 ± 9.92 years) received an equivalent amount of maltodextrin and lactose for 16 weeks. Neuropsychological tests (Rey-Kim memory test and Stroop color-word test) and electroencephalography were conducted to evaluate the effect of LGNC-07 on memory and attention. Further analyses were stratified by baseline severity to evaluate treatment response on the degree of impairment (MMSE-K 21-23 and 24-26). LGNC-07 led to improvements in memory by marginally increasing delayed recognition in the Rey-Kim memory test (P = .0572). Stratified analyses showed that LGNC-07 improved memory and selective attention by significantly increasing the Rey-Kim memory quotient and word reading in the subjects with MMSE-K scores of 21-23 (LGNC-07, n = 11; placebo, n = 9). Electroencephalograms were recorded in 24

randomly selected subjects hourly for 3 hours in eye-open, eye-closed, and reading states after a single dose of LGNC-07 (LGNC-07, n = 12; placebo, n = 12). Brain theta waves, an indicator of cognitive alertness, were increased significantly in the temporal, frontal, parietal, and occipital areas after 3 hours in the eye-open and reading states. Therefore, this study suggests that LGNC-07 has potential as an intervention for cognitive improvement. (Full-text unavailable)

Qiangye, Z., Hongchao, Y., Jian, W., Aiwu, L., Wentong, Z., Xinhai, C., & Kelai, W. (2013). Effect of green tea on reward learning in healthy individuals: a randomized, double-blind, placebo-controlled pilot study. *Nutrition Journal*, *12*(1), 1-7. doi:10.1186/1475-2891-12-84.

Background: Both clinical and preclinical studies revealed that regular intake of green tea reduced the prevalence of depressive symptoms, as well as produced antidepressant-like effects in rodents. Evidence proposed that disturbed reward learning has been associated with the development of anhedonia, a core symptom of depression. However, the relationship between green tea and reward learning is poorly investigated. Our goal was to test whether chronic treatment with green tea in healthy subjects affects the process of reward learning and subsequently regulates the depressive symptoms. Methods: Seventy-four healthy subjects participated in a double-blind, randomized placebo-controlled study with oral administration of green tea or placebo for 5weeks. We used the monetary incentive delay task to evaluate the reward learning by measurement of the response to reward trial or no-reward trial. We compared the reaction time of reward responsiveness between green tea and placebo treatment. Furthermore, we selected Montgomery-Asberg depression rating scale (MADRS) and 17-item Hamilton Rating Scale for Depression (HRSD-17) to estimate the depressive symptoms in these two groups. Results: The results showed chronic treatment of green tea increased reward learning compared with placebo by decreasing the reaction time in monetary incentive delay task. Moreover, participants treated with green tea showed reduced scores measured in MADRS and HRSD-17 compared with participants treated with placebo. Conclusions: Our findings reveal that chronic green tea increased the reward learning and prevented the depressive symptoms. These results also raised the possibility that supplementary administration of green tea might reverse the development of depression through normalization of the reward function.

Rogers, P. W. (2008). Time for tea: mood, blood pressure and cognitive performance effects of caffeine and theanine administered alone and together. *Psychopharmacology*, *195*(4), 569-577.

Although both contain behaviorally significant concentrations of caffeine, tea is commonly perceived to be a less stimulating drink than coffee. At least part of the explanation for this may be that theanine, which is present in tea but not coffee, has relaxing effects. There is also some evidence that theanine affects cognitive performance, and it has been found to reduce blood pressure in hypertensive rats. To study the subjective, behavioral and blood pressure effects of theanine and caffeine administered alone and together, in doses relevant to the daily tea consumption of regular tea drinkers. In a randomized, double-blind, placebo-controlled study, healthy adult participants (n = 48) received either 250-mg caffeine, 200-mg theanine, both or neither of these. They completed ratings of mood, including anxiety, and alertness, and had their blood pressure measured before and starting 40 min after drug administration. Anxiety was also

assessed using a visual probe task. Caffeine increased self-rated alertness and jitteriness and blood pressure. Theanine antagonized the effect of caffeine on blood pressure but did not significantly affect jitteriness, alertness or other aspects of mood. Theanine also slowed overall reaction time on the visual probe task. Theanine is a physiologically and behaviorally active compound and, while it is unclear how its effects might explain perceived differences between tea and coffee, evidence suggests that it may be useful for reducing raised blood pressure.

Unno, K., Fujitani, K., Takamori, N., Takabayashi, F., Maeda, K., Miyazaki, H., & ... Hoshino, M. (2011). Theanine intake improves the shortened lifespan, cognitive dysfunction and behavioral depression that are induced by chronic psychosocial stress in mice. *Free Radical Research*, *45*(8), 966-974. doi:10.3109/10715762.2011.566869

To evaluate the psychosocial effect on lifespan and cognitive function, this study investigated the effect of confrontational housing on mice because conflict among male mice is a psychosocial stress. In addition, it investigated the anti-stress effect of theanine (γγ-glutamylethylamide), an amino acid in tea. Mice were housed under confrontation. That is, two male mice were separately housed in the same cage with a partition for establishing the territorial imperative in each mouse. Then, the partition was removed and mice were co-housed confrontationally (confront-housing) using a model mouse of accelerated-senescence (SAMP10) that exhibited cerebral atrophy and cognitive dysfunction with ageing. It was found that mice began to die earlier under confront-housing than group-housed control mice. Additionally, it was found that cerebral atrophy, learning impairment and behavioral depression were higher in mice under the stressed condition of confront-housing than age-matched mice under group-housing. Furthermore, the level of oxidative damage in cerebral DNA was higher in mice housed confrontationally than group-housed control mice. On the other hand, the consumption of purified theanine (20 μμg/ml, 5--6 mg/kg) suppressed the shortened lifespan, cerebral atrophy, learning impairment, behavioral depression and oxidative damage in cerebral DNA. These results suggest that psychosocial stress accelerates age-related alterations such as oxidative damage, lifespan, cognitive dysfunction and behavioral depression. The intake of theanine might be a potential candidate for suppression of disadvantage under psychosocial stress.

Wakabayashi, C. H. (2012). Behavioral and molecular evidence for psychotropic effects in l-theanine. *Psychopharmacology*, *219*(4), 1099-1109.

l-Theanine (N-ethyl- l-glutamine) is an amino acid uniquely found in green tea and historically considered to be a relaxing agent. It is a glutamate derivative and has an affinity for glutamatergic receptors. However, its psychotropic effects remain unclear. Objectives: To elucidate effects of l-theanine on psychiatric disease-related behaviors in mice and its molecular basis focusing on brain-derived neurotrophic factor (BDNF) and N-methyl- d-aspartate (NMDA) receptor. Methods: We examined the effects of l-theanine on behaviors in mice by using the open-field test (OFT), forced swim test (FST), elevated plus-maze test (EPMT), and prepulse inhibition (PPI) of acoustic startle. By western blot analysis, we looked at the effect of l-theanine on the expression of BDNF and related proteins in the hippocampus and cerebral cortex. To determine whether l-theanine has agonistic action on the NMDA receptor, we performed Fluo-3 intracellular Ca imaging in

cultured cortical neurons. Results: Single administration of l-theanine significantly attenuated MK-801-induced deficits in PPI. Subchronic administration (3-week duration) of l-theanine significantly reduced immobility time in the FST and improved baseline PPI. Western blotting analysis showed increased expression of BDNF protein in the hippocampus after subchronic administration of l-theanine. In cultured cortical neurons, l-theanine significantly increased the intracellular Ca concentration, and this increase was suppressed by competitive and non-competitive NMDA receptor antagonists (AP-5 and MK-801, respectively). Conclusions: Our results suggest that l-theanine has antipsychotic-like and possibly antidepressant-like effects. It exerts these effects, at least in part, through induction of BDNF in the hippocampus and the agonistic action of l-theanine on the NMDA receptor.

White, D. J., de Klerk, S., Woods, W., Gondalia, S., Noonan, C., & Scholey, A. B. (2016). Anti-Stress, Behavioural and Magnetoencephalography Effects of an l-Theanine-Based Nutrient Drink: A Randomised, Double-Blind, Placebo-Controlled, Crossover Trial. *Nutrients*, *8*(1), doi:10.3390/nu8010053

l-theanine (γ-glutamylethylamide) is an amino acid found primarily in the green tea plant. This study explored the effects of an l-theanine-based nutrient drink on mood responses to a cognitive stressor. Additional measures included an assessment of cognitive performance and resting state alpha oscillatory activity using magnetoencephalography (MEG). Thirty-four healthy adults aged 18-40 participated in this double-blind, placebo-controlled, balanced crossover study. The primary outcome measure, subjective stress response to a multitasking cognitive stressor, was significantly reduced one hour after administration of the l-theanine drink when compared to placebo. The salivary cortisol response to the stressor was reduced three hours' post-dose following active treatment. No treatment-related cognitive performance changes were observed. Resting state alpha oscillatory activity was significantly greater in posterior MEG sensors after active treatment compared to placebo two hours' post-dose; however, this effect was only apparent for those higher in trait anxiety. This change in resting state alpha oscillatory activity was not correlated with the change in subjective stress response or the cortisol response, suggesting further research is required to assess the functional relevance of these treatment-related changes in resting alpha activity. These findings further support the anti-stress effects of l-theanine.

Rhodiola

Bystritsky, A., Kerwin, L., & Feusner, J. (2008). A pilot study of Rhodiola rosea (RHODAX) for generalized anxiety disorder (GAD). *Journal of Alternative & Complementary Medicine*, *14*(2), 175-180 6p. doi:10.1089/acm.2007.7117

Background: Rhodiola rosea is an herbal supplement that many in the general population in Russia and elsewhere in the world have used for decades to alleviate everyday anxiety, depression, and insomnia. Whether R. rosea is effective in reducing similar symptoms in clinical samples is unknown. The goal of this pilot study was to evaluate whether R. rosea is effective in reducing symptoms of generalized anxiety disorder (GAD). Method: Ten (10) participants with a DSM-IV diagnosis of GAD, recruited from the UCLA Anxiety Disorders Program and between the ages of 34 and 55, were enrolled in this study from November 2005 to May 2006. Participants received a total daily dose of 340 mg of R. rosea extract for 10 weeks. Assessments included the Hamilton Anxiety Rating Scale (HARS), the Four-Dimensional Anxiety and Depression Scale, and the Clinical Global Impressions of Severity/Improvement Scale. Results: Individuals treated with R. rosea showed significant decreases in mean HARS scores at endpoint ($t = 3.27$, $p = 0.01$). Adverse events were generally mild or moderate in severity, the most common being dizziness and dry mouth. Conclusions: Significant improvement in GAD symptoms was found with R. rosea, with a reduction in HARS scores similar to that found in clinical trials. These preliminary findings warrant further exploration of treatment with R. rosea in clinical samples.

Chen, Q., Zeng, Y., Qu, Z., Tang, J., Qin, Y., Chung, P., & ... Hägg, U. (2009). The effects of Rhodiola rosea extract on 5-HT level, cell proliferation and quantity of neurons at cerebral hippocampus of depressive rats. *Phytomedicine*, *16*(9), 830-838 9p. doi:10.1016/j.phymed.2009.03.011

The purpose of this study was to investigate the effects of Rhodiola rosea extract and depression on the serotonin (5-HT) level, cell proliferation and quantity of neurons at cerebral hippocampus of depressive rats induced by Chronic Mild Stress (CMS). Seventy male Sprague-Dawley rats were divided into seven groups (10 per group): normal control group, untreated depressive rat model group, negative control group, positive control group, low dosage Rhodiola rosea extract (1.5g/kg) group, medium dosage Rhodiola rosea extract (3g/kg) group and high dosage Rhodiola rosea extract (6g/kg) group. After the depressive rats induced by CMS had received Rhodiola rosea extract for 3 weeks, the 5-HT levels at cerebral hippocampus were detected by high performance liquid chromatography. Bromodeoxyuridine (BrdU) was injected in vivo to label the proliferating cells at hippocampus, and morphometry was used to count the hippocampal neurons. The results showed that the 5-HT level of the three experimental groups had recovered to normal status. The immunohistochemistry of hippocampus BrdU positive cells had returned to the normal level in the group of depressive rats with low dosage Rhodiola rosea extract. In conclusion the results demonstrated that Rhodiola rosea extract could improve 5-HT level in hippocampus in depressive rats, and low dosage Rhodiola rosea could induce neural stem cell proliferation at hippocampus to return to normal level, repairing the injured neurons at hippocampus.

Darbinyan, V. A. (2007). Clinical trial of Rhodiola rosea L. extract SHR-5 in the treatment of mild to moderate depression. *Nordic Journal of Psychiatry*, *61*(5), 343-348.

The objective of this study was to assess the efficacy and safety of standardized extract SHR-5 of rhizomes of Rhodiola rosea L. in patients suffering from a current episode of mild/moderate depression. The phase III clinical trial was carried out as a randomized double-blind placebo-controlled study with parallel groups over 6 weeks. Participants, males and females aged 18-70 years, were selected according to DSM-IV diagnostic criteria for depression, the severity of which was determined by scores gained in Beck Depression Inventory and Hamilton Rating Scale for Depression (HAMD) questionnaires. Patients with initial HAMD scores between 21 and 31 were randomized into three groups, one of which (group A: 31 patients) received two tablets daily of SHR-5 (340 mg/day), a second (group B: 29 patients) received two tablets twice per day of SHR-5 (680 mg/day), and a third (group C: 29 patients) received two placebo tablets daily. The efficacy of SHR-5 extract with respect to depressive complaints was assessed on days 0 and 42 of the study period from total and specific subgroup HAMD scores. For individuals in groups A and B, overall depression, together with insomnia, emotional instability and somatization, but not self-esteem, improved significantly following medication, whilst the placebo group did not show such improvements. No serious side-effects were reported in any of the groups A-C. It is concluded that the standardized extract SHR-5 shows anti-depressive potency in patients with mild to moderate depression when administered in dosages of either 340 or 680 mg/day over a 6-week period.

Dwyer, A. V., Whitten, D. L., & Hawrelak, J. A. (2011). Herbal Medicines, other than St. John's Wort, in the Treatment of Depression: A Systematic Review. *Alternative Medicine Review*, *16*(1), 40-49 10p.

A computer-based search of Medline, Cinahl, AMED, ALT Health Watch, Psych Articles, Psych Info, Current Contents databases, Cochrane Controlled Trials Register, and Cochrane Database of Systematic Reviews, was performed. Researchers were contacted, and bibliographies of relevant papers and previous meta-analysis were hand searched for additional references. Trials were included in the review if they were prospective human trials assessing herbal medicines, other than St. John's wort, in the treatment of mild-to-moderate depression and utilized validated instruments to assess participant eligibility and clinical endpoints. Nine trials were identified that met all eligibility requirements. Three studies investigated saffron stigma, two investigated saffron petals, and one compared saffron stigma to the petal. Individual trials investigating lavender, Echium, and Rhodiola were also located. Results of the trials are discussed. Saffron stigma was found to be significantly more effective than placebo and equally as efficacious as fluoxetine and imipramine. Saffron petal was significantly more effective than placebo and was found to be equally efficacious compared to fluoxetine and saffron stigma. Lavender was found to be less effective than imipramine, but the combination of lavender and imipramine was significantly more effective than imipramine alone. When compared to placebo, Echium was found to significantly decrease depression scores at week 4, but not week 6. Rhodiola was also found to significantly improve depressive symptoms when compared to placebo. A number of herbal medicines show promise in the management of mild-to-moderate depression.

Hillhouse, B., Dong Sheng, M., French, C., & Towers, N. (2004). Acetylcholine Esterase Inhibitors in Rhodiola rosea. *Pharmaceutical Biology*, *42*(1), 68-72.

The alcohol extract or Rhodiola rosea has been shown to cause 42 ± 3.2% inhibition of acetylcholine esterase (AChE) when tested at 10 g/L. This AChE inhibition provides a physiological explanation for the reported mental and memory enhancing properties of Rhodiola rosea extracts. Active guided fractionation indicated a multitude of components which are responsible for this plants AChE inhibition. Two flavonoid glycosides (gossypetin-7- O -l-rhamnopyranoside and rhodioflavonoside) were isolated and shown to cause 58 ± 15% and 38 ± 4% AChE inhibition respectively when tested at 5 g/L. In view of this new enzymatic activity and previous clinical work indicating memory and mental enhancing properties with no indication of toxicity, this plant needs to be researched for its potential at treating memory impairing disorders such as Alzheimer's disease.

Jurcău, R., Jurcău, I., & Bodescu, C. (2012). Anxiety and salivary cortisol modulation in exercise induced stress, using a phytotherapic product containing Rhodiola Rosea. *Palestrica of the Third Millennium Civilization & Sport*, *13*(3), 213-217.

Background. Phytotherapic products may influence the effect of stress on anxiety and salivary cortisol. Aims. The objective of the study is to highlight the phytotherapic modulation of dynamic peri-stress changes induced by intense short duration physical exercise on two parameters (anxiety and salivary cortisol), in sedentary subjects. Methods. The chosen subjects (n=24) were selected according to the requirements of the study. Stress was represented by intense short duration physical exercise, made with a Monark Ergomedic 839E cycle ergometer. The analyzed indicators were anxiety and salivary cortisol. The phytotherapic product (PP) used contained Rhodiola Rosea. Statistical evaluation was made on the basis of Student test. Results. Anxiety and salivary cortisol were reduced immediately pre- and post-stress in subjects who were administered the PP, compared with subjects who did not follow the phytotherapic treatment. Conclusions. 1) Anxiety and salivary cortisol were significantly reduced immediately pre- and post-exercise, in the case of stress caused by intense short duration physical exercise, in sedentary persons, under PP influence. 2) There were differences in dynamic developments between the PP treated group and the untreated group, for anxiety and salivary cortisol. 3) The influence of the PP used was significantly more intense on anxiety than on salivary cortisol, immediately pre- and post-stress. 4) The PP used may be an effective, safe and accessible modulation path for stress caused by intense short duration physical exercise in sedentary persons.

Kelly, G. (2001). Rhodiola rosea: a possible plant adaptogen. *Alternative Medicine Review*, *6*(3), 293-302 10p.

Rhodiola rosea is a popular plant in traditional medical systems in Eastern Europe and Asia with a reputation for stimulating the nervous system, decreasing depression, enhancing work performance, eliminating fatigue, and preventing high altitude sickness. Rhodiola rosea has been categorized as an adaptogen by Russian researchers due to its observed ability to increase resistance to a variety of chemical, biological, and physical stressors. Its claimed benefits include antidepressant, anticancer, cardioprotective, and central

nervous system enhancement. Research also indicates great utility in asthenic conditions (decline in work performance, sleep difficulties, poor appetite, irritability, hypertension, headaches, and fatigue) developing subsequent to intense physical or intellectual strain. The adaptogenic, cardiopulmonary protective, and central nervous system activities of Rhodiola rosea have been attributed primarily to its ability to influence levels and activity of monoamines and opioid peptides such as beta-endorphins.

Lee, F., Kuo, T., Liou, S., & Chien, C. (2009). Chronic Rhodiola rosea extract supplementation enforces exhaustive swimming tolerance. *American Journal of Chinese Medicine*, *37*(3), 557-572 16p.

We explored the effects and mechanisms of Rhodiola rosea extract supplementation on swimming-induced fatigue in rats. The concentrations of active components in Rhodiola rosea have been determined by high performance liquid chromatography-mass spectrometer. The Rhodiola rosea extract supplementation in water for 2-4 weeks was evaluated in male Wistar rats with 90-min unloaded swimming exercise and 5% body weight loaded swimming up to fatigue. We measured the fatigue biomarkers, including blood urea nitrogen (BUN), glutamic oxaloacetic transaminase (GOT) and glutamic pyruvic transaminase (GPT), lactate dehydrogenase (LDH), hepatic glycogen content, the activity of fat metabolism enzymes, sterol regulatory element-binding protein-1 (SREBP-1) and fatty acid synthase (FAS), the tissue oxygen content and ratio of red and white skeletal muscle fibers in rats. Rhodiola rosea significantly increased liver glycogen, SREBP-1, FAS, heat shock protein 70 expression, Bcl-2/Bax ratio and oxygen content before swimming. Rhodiola rosea supplementation significantly increased the swimming time in a dose-dependent manner and reduced swimming-enhanced serum BUN, GOT and GPT levels. The ratio of red and white muscle fibers was not altered after chronic Rhodiola rosea extract supplementation. Chronic Rhodiola rosea supplementation significantly improved exhaustive swimming-induced fatigue by the increased glycogen content, energy supply of lipogenic enzyme expressions and protective defense mechanisms.

Li, T., Xu, G., Wu, L., & Sun, C. (2007). Pharmacological studies on the sedative and hypnotic effect of salidroside from the Chinese medicinal plant Rhodiola sachalinensis. *Phytomedicine*, *14*(9), 601-604 4p.

This is the preliminary study of sedative and hypnotic activity of salidroside (a major component of Rhodiola sachalinensis) in mice by using synergism with pentobarbital as an index for the hypnotic effect. Loss of the righting reflex was used to determine the start of sleep. Sleep latency and sleeping time were evaluated in this experiment. The results showed that salidroside could obviously shorten the sleep latency and prolong the sleeping time of mice produced by pentobarbital sodium (55 mg/kg, i.p.). Salidroside produces significant sedative-hypnotic effect. The dose-effect relationship is remarkable. (Full-text available)

Mao, J. J., Xie, S. X., Zee, J., Soeller, I., Li, Q. S., Rockwell, K., & Amsterdam, J. D. (2015). Rhodiola rosea versus sertraline for major depressive disorder: A randomized placebo-controlled trial. *Phytomedicine*, *22*(3), 394-399 6p. doi:10.1016/j.phymed.2015.01.010

Background: We performed a proof of concept trial to evaluate relative safety and efficacy of Rhodiola rosea (R. rosea) versus sertraline for mild to moderate major depressive disorder. Hypothesis: We hypothesize that R. rosea would have similar therapeutic effects as sertraline but with less adverse events. Study Design: Phase II randomized placebo controlled clinical trial. Methods: 57 subjects were randomized to 12 weeks of standardized R. rosea extract, sertraline, or placebo. Changes over time in Hamilton Depression Rating (HAM-D), Beck Depression Inventory (BDI), and Clinical Global Impression Change (CGI/C) scores among groups were examined using mixed-effects models. Results: Modest, albeit statistically non-significant, reductions were observed for HAM-D, BDI, and CGI/C scores for all treatment conditions with no significant difference between groups ($p = 0.79$, $p = 0.28$, and $p = 0.17$, respectively). The decline in HAM-D scores was greater for sertraline (-8.2, 95% confidence interval [CI], -12.7 to -3.6) versus R. rosea (-5.1, 95% CI: -8.8 to -1.3) and placebo (-4.6, 95% CI: -8.6 to -0.6). While the odds of improving (versus placebo) were greater for sertraline (1.90 [0.44-8.20]; odds ratio [95% CI]) than R. rosea (1.39 [0.38-5.04]), more subjects on sertraline reported adverse events (63.2%) than R. rosea (30.0%) or placebo (16.7%) ($p = 0.012$). Conclusions: Although R. rosea produced less antidepressant effect versus sertraline, it also resulted in significantly fewer adverse events and was better tolerated. These findings suggest that R. rosea, although less effective than sertraline, may possess a more favorable risk to benefit ratio for individuals with mild to moderate depression. (Full-text unavailable)

Mattioli, L. M. (2012). Effects of a Rhodiola rosea L. extract on the acquisition, expression, extinction, and reinstatement of morphine-induced conditioned place preference in mice. *Psychopharmacology*, *221*(2), 183-193.

Rationale: Opioid addiction is a chronic, recurrent brain disease that is characterized by compulsive drug seeking and a high rate of relapse even after long periods of abstinence. Prevention of relapse is the primary goal of addiction treatment and is still the major limitation in drug therapy. Objectives: The present study investigated the effects of a Rhodiola rosea L. hydroalcoholic extract (RHO), a well-known traditional oriental medicine, on establishment and reinstatement of morphine-induced conditioned place preference (CPP) in mice. Methods: CPP was induced by intraperitoneal injection of morphine (10 mg/kg) as an 8-day conditioning schedule. The effects of RHO on the rewarding properties of morphine were tested in mice receiving oral administration of RHO (10, 15, and 20 mg/kg) 60 min prior to each morphine injection (acquisition) or prior to the CPP test on day 9 (expression). Once established, CPP was extinguished by repeated testing, during which conditioned mice were injected daily with different doses of RHO. Finally, the efficacy of RHO in blocking reinstatement of CPP provoked by priming injections and physical stress was also evaluated. Results: RHO administration showed dose dependency for prevention of establishment of CPP and was effective in facilitating extinction of morphine-induced CPP. RHO suppressed both priming- and stress-induced reinstatement of CPP in a dose-dependent manner. Conclusions: In conclusion, as RHO was effective for reducing craving and vulnerability to relapse, it might be a very effective natural remedy for the treatment of opioid addiction. [ABSTRACT FROM AUTHOR]

Panossian, A., Wikman, G., & Sarris, J. (2010). Rosenroot (Rhodiola rosea): traditional use, chemical composition, pharmacology and clinical efficacy. *Phytomedicine*, *17*(7), 481-493 13p. doi:10.1016/j.phymed.2010.02.002

The aim of this review article was to summarize accumulated information related to chemical composition, pharmacological activity, traditional and official use of Rhodiola rosea L. in medicine. In total approximately 140 compounds were isolated from roots and rhizome - monoterpene alcohols and their glycosides, cyanogenic glycosides, aryl glycosides, phenylethanoids, phenylpropanoids and their glycosides, flavonoids, flavonlignans, proanthocyanidins and gallic acid derivatives. Studies on isolated organs, tissues, cells and enzymes have revealed that Rhodiola preparations exhibit adaptogenic effect including, neuroprotective, cardioprotectiv e, anti-fatigue, antidepressive, anxiolytic, nootropic, life-span increasing effects and CNS stimulating activity. A number of clinical trials demonstrate that repeated administration of R. rosea extract SHR-5 exerts an anti-fatigue effect that increases mental performance (particularly the ability to concentrate in healthy subjects), and reduces burnout in patients with fatigue syndrome. Encouraging results exist for the use of Rhodiola in mild to moderate depression, and generalized anxiety. Several mechanisms of action possibly contributing to the clinical effect have been identified for Rhodiola extracts. They include interactions with HPA-system (cortisol-reducing), protein kinases p-JNK, nitric oxide, and defense mechanism proteins (e.g. heat shock proteins Hsp 70 and FoxO/DAF-16). Lack of interaction with other drugs and adverse effects in the course of clinical trials make it potentially attractive for use as a safe medication. In conclusion, Rhodiola rosea has robust traditional and pharmacological evidence of use in fatigue, and emerging evidence supporting cognition and mood.

Provino, R. (2010). The role of adaptogens in stress management. *Australian Journal of Medical Herbalism*, *22*(2), 41-49.

Stress is a normal part of everyday life but it is important to be able to use tools for its management otherwise chronic stress, if left untreated, can lead to a variety of stress related illnesses including hypertension, heart disease, anxiety, depression, memory impairment and chronic fatigue syndrome. The aim of this literature review is to summarize and critically analyses research conducted on the adaptogenic herbs Withania somnifera, Panax ginseng, Eleutherococcus senticosus, Schisandra chinensis, Glycyrrhiza glabra, Rhodiola rosea, Bacopa monniera and Centella asiatica. The mechanism of adaptogens appears to involve the hypothalamic pituitary adrenal axis with resultant decreases or normalising of nitric oxide and cortisol, which are increased during times of stress. Most adaptogens also have anxiolytic and antioxidant properties and these have been attributed to their adaptogenic effect. Methods: The online databases PUBMED, PROQUEST, EBSCO and the Directory of Open Access Journals were searched to obtain peer-reviewed journal articles on adaptogens. The Library of the National Herbalists Association of Australia was also searched. Most studies used animal experimental models but some limited human clinical trials have been conducted. Fourteen studies using animal experimental models were identified and these have been summarized in Table 1. Twelve human clinical trials were identified and these have been summarized in Table 2. Results and conclusions: Withania somnifera appears to be the most commonly used, and extensively studied, adaptogen followed closely by Panax ginseng and Eleutherococcus senticosus.

Schisandra chinensis is an extensively used adaptogen in Traditional Chinese Medicine (TCM). Other herbs studied that show adaptogenic activity include Rhodiola rosea, Glycyrrhiza glabra, Bacopa monniera and Centella asiatica, with the latter two more specific in improving memory. Most studies on adaptogens have used animal experimental models and whilst positive benefits have been shown, larger scale human clinical trials are needed.

Ulbricht, C., Chao, W., Tanguay-Colucci, S., Costa, D., Culwell, S., Giese, N., & ... Redwanski, J. (2011). Rhodiola (Rhodiola spp.): An Evidence-Based Systematic Review by the Natural Standard Research Collaboration. *Alternative & Complementary Therapies*, *17*(2), 110-119. doi:10.1089/act.2011.17208

The article presents an evidence-based research on the therapeutic use of Rhodiola rosea on various conditions including anxiety, adaptogen, and fatigue. The research utilized systematic review method of several studies on topics such as the use of Rhodiola rosea in stress-related fatigue and the mental performance of pneumonia patients treated with Rhodiola rosea containing product. Preliminary evidences indicate medical efficacy of Rhodiola rosea in reducing depression, anxiety, and fatigue.

Ginger Root Extract

Abascal, K., & Yarnell, E. (2009). Clinical uses of Zingiber officinale (ginger). *Alternative & Complementary Therapies*, *15*(5), 231-237 7p.

This article discusses the clinical uses of Zingiber officinale (ginger) rhizome in Western botanical medicine. Ginger's ability to quell nausea and vomiting in a variety of conditions (pregnancy-induced, drug-induced [e.g., anesthetic, chemotherapy]), and motion-induced as well as digestive ailments) is considered to be the herb's most important effect today. However, ginger has many other clinical uses, some justified by traditional use and now some starting to see preliminary verification in clinical trials. These include ginger's use for treating colds, pulmonary afflictions, joint ailments, headaches, and orchitis. Throughout this article, various doses of ginger products are touched upon. The safety of ginger, focusing on its drug-herb interactions (or lack thereof), is also reviewed. The related plant Zingiber zerumbet (shell ginger) is mentioned. Ginger remains an important herb after thousands of years of recorded use for medicine.

Khayat, S., Kheirkhah, M., Moghadam, Z. B., Fanaei, H., Kasaeian, A., & Javadimehr, M. (2014). Effect of Treatment with Ginger on the Severity of Premenstrual Syndrome Symptoms. *ISRN Obstetrics & Gynecology*, 1-5. doi:10.1155/2014/792708

Premenstrual syndrome (PMS) is a common disorder. Although the etiology of PMS is not clear, to relieve from this syndrome different methods are recommended. One of them is use of medicinal herbs. This study was carried out to evaluate effects of ginger on severity of symptoms of PMS. This study was a clinical trial, double-blinded work, and participants were randomly allocated to intervention (n = 35) and control (n = 35) groups. To determine persons suffering from PMS, participants completed daily record scale questionnaire for two consecutive cycles. After identification, each participant received two ginger capsules daily from seven days before menstruation to three days after menstruation for three cycles and they recorded severity of the symptoms by daily record scale questionnaire. Data before intervention were compared with date 1, 2, and 3 months after intervention. Before intervention, there were no significant differences between the mean scores of PMS symptoms in the two groups, but after 1, 2, and 3 months of treatment, there was a significant difference between the two groups ($P < 0.0001$). Based on the results of this study, maybe ginger is effective in the reduction of severity of mood and physical and behavioral symptoms of PMS and we suggest ginger as treatment for PMS.

Naritsara, S., Jintanaporn, W., Supaporn, M., Terdthai, T., Nawanant, P., Chuleratana, B., & Tanwarat, K. (2012). Zingiber officinale Improves Cognitive Function of the Middle-Aged Healthy Women. *Evidence-Based Complementary & Alternative Medicine (Ecam)*, *2012*1-9.

The development of cognitive enhancers from plants possessing antioxidants has gainedmuch attention due to the role of oxidative stress-induced cognitive impairment. Thus, this study aimed to determine the effect of ginger extract, or Zingiber officinale, on the cognitive function of middle-aged, healthy women. Sixty participants were randomly assigned to receive a placebo or standardized plant extract at doses of 400 and

800 mg once daily for 2months. They were evaluated for working memory and cognitive function using computerized battery tests and the auditory oddball paradigm of event-related potentials at three different time periods: before receiving the intervention, one month, and two months. We found that the ginger-treated groups had significantly decreased P300 latencies, increased N100 and P300 amplitudes, and exhibited enhanced working memory. Therefore, ginger is a potential cognitive enhancer for middle-aged women.

Nozaki, C. L. (2011). Zinc alleviates pain through high-affinity binding to the NMDA receptor NR2A subunit. *Nature Neuroscience*, *14*(8), 1017-1022.

Zinc is abundant in the central nervous system and regulates pain, but the underlying mechanisms are unknown. In vitro studies have shown that extracellular zinc modulates a plethora of signaling membrane proteins, including NMDA receptors containing the NR2A subunit, which display exquisite zinc sensitivity. We created NR2A-H128S knock-in mice to investigate whether Zn^{2+}-NR2A interaction influences pain control. In these mice, high-affinity (nanomolar) zinc inhibition of NMDA currents was lost in the hippocampus and spinal cord. Knock-in mice showed hypersensitivity to radiant heat and capsaicin, and developed enhanced allodynia in inflammatory and neuropathic pain models. Furthermore, zinc-induced analgesia was completely abolished under both acute and chronic pain conditions. Our data establish that zinc is an endogenous modulator of excitatory neurotransmission in vivo and identify a new mechanism in pain processing that relies on NR2A NMDA receptors. The study also potentially provides a molecular basis for the pain-relieving effects of dietary zinc supplementation.

Ozgoli, G., Goli, M., & Moattar, F. (2009). Comparison of effects of ginger, mefenamic acid, and ibuprofen on pain in women with primary dysmenorrhea. *Journal of Alternative & Complementary Medicine*, *15*(2), 129-132 4p. doi:10.1089/acm.2008.0311

Objectives: To compare the effects of ginger, mefenamic acid, and ibuprofen on pain in women with primary dysmenorrhea. Methods: This was a double-blind comparative clinical trial conducted from September 2006 to February 2007. Participants were 150 students (18 years old and over) with primary dysmenorrhea from the dormitories of two medical universities who were alternately divided into three equal groups. Students in the ginger group took 250 mg capsules of ginger rhizome powder four times a day for three days from the start of their menstrual period. Members of the other groups received 250 mg mefenamic acid or 400 mg ibuprofen capsules, respectively, on the same protocol. A verbal multidimensional scoring system was used for assessing the severity of primary dysmenorrhea. Severity of disease, pain relief, and satisfaction with the treatment were compared between the groups after one menstruation. Results: There were not significant differences between groups in baseline characteristics, $p > 0.05$. At the end of treatment, severity of dysmenorrhea decreased in all groups and no differences were found between the groups in severity of dysmenorrhea, pain relief, or satisfaction with the treatment, $p > 0.05$. No severe side effects occurred. Conclusion: Ginger was as effective as mefenamic acid and ibuprofen in relieving pain in women with primary dysmenorrhea. Further studies regarding the effects of ginger on other symptoms associated with dysmenorrhea and efficacy and safety of various doses and treatment durations of ginger are warranted.

Sepahvand, R., Esmaeili-Mahani, S., Arzi, A., Rasoulian, B., & Abbasnejad, M. (2010). Ginger (Zingiber officinale Roscoe) elicits antinociceptive properties and potentiates morphine-induced analgesia in the rat radiant heat tail-flick test. *Journal of Medicinal Food*, *13*(6), 1397-1401 5p. doi:10.1089/jmf.2010.1043

Ginger (Zingiber officinale Roscoe), a well-known spice plant, has been used traditionally in the treatment of a wide variety of ailments. It has been shown that ginger is a calcium channel blocker; however, its influence on morphine analgesic effects has not been elucidated. We examined the effect of ginger root extract on nociceptive threshold and morphine-induced analgesia in male Wistar rats. To determine the effect of ginger on morphine analgesia, ginger extract (200, 400, and 600 mg/kg i.p.) was injected before a sub effective dose of morphine (2.5 mg/kg i.p.). The radiant heat tail-flick test was used to assess the nociceptive threshold before and at different times after drug administration. Our results showed that ginger extract elicited a significant antinociceptive effect. In addition, in groups that received both morphine and ginger, the observed analgesia was higher than that in groups treated with either morphine or ginger extract alone. Thus, the data indicate that ginger extract has a beneficial influence on morphine analgesia and can be an efficacious adjunct for pain management.

Terry, R., Posadzki, P., Watson, L. K., & Ernst, E. (2011). The Use of Ginger (Zingiber officinale) for the Treatment of Pain: A Systematic Review of Clinical Trials. *Pain Medicine*, *12*(12), 1808-1818. doi:10.1111/j.1526-4637.2011.01261.x

Background. Zingiber officinale (Z. officinale), commonly known as ginger, has been widely used traditionally for a variety of medicinal purposes, one of which is for the treatment of pain. The aim of this systematic review was to evaluate the evidence from all human participant clinical trials that have assessed the efficacy of ginger for the treatment of any type of pain. Methods. Following a protocol, multiple databases were sought using comprehensive search strategies for Z. officinale and pain together with a trial filter for randomized or controlled clinical trials. Trials testing the efficacy of Z. officinale, used as a sole oral treatment against a comparison condition in human adults suffering from any pain condition, were included. Results. Seven published articles, reporting a total of eight trials (481 participants), were included in the review. Six trials (two for osteoarthritis, one for dysmenorrhea, and three for experimentally induced acute muscle pain) found that the use of Z. officinale reduced subjective pain reports. The methodological quality of the included articles was variable. When assessed using the Jadad scale, which allows a score of between 0 and 5 to be given, included articles obtained Jadad ratings ranging from 2 to 5. Conclusion. Due to a paucity of well-conducted trials, evidence of the efficacy of Z. officinale to treat pain remains insufficient. However, the available data provide tentative support for the anti-inflammatory role of Z. officinale constituents, which may reduce the subjective experience of pain in some conditions such as osteoarthritis. Further rigorous trials therefore seem to be warranted.

Additional Ingredients

Abokrysha, N. T. (2012). Vitamin D Deficiency in Women with Fibromyalgia in Saudi Arabia. *Pain Medicine*, *13*(3), 452-458. doi:10.1111/j.1526-4637.2011.01304.x

Background. The relation between low levels of 25-hydroxyvitamin D and nonspecific musculoskeletal pain, including fibromyalgia syndrome, is debatable. Many studies have reported 'a positive relation' and others 'found no relation.' Objectives. To determine the prevalence of vitamin D deficiency among patients with fibromyalgia in a neurology clinic in the Kingdom of Saudi Arabia (KSA). Methods. This study was done at a neurology clinic of Bugshan Hospital, Jeddah, KSA, from January to April 2011. Thirty female patients were diagnosed with fibromyalgia according to new clinical fibromyalgia diagnostic criteria; their serum vitamin D levels were screened. Vitamin D deficiency is defined as <20 ng/mL, vitamin D insufficiency is defined as 21-29 ng/mL, and vitamin D sufficiency is equal to or >30 ng/mL. Result. Thirty female patients were included in the study. The mean age was 34.56 ± 8.1 years. Mean vitamin D level was 4.76 ± 1.46 ng/mL. A significant negative correlation between vitamin D level and widespread pain index was found. Thirty percent of the patients were Saudi Arabian of whom 100% were veiled; 70% were non-Saudi Arabian of whom 47.6% were veiled and 52.4% wore long pants and/or full sleeved clothes. Vitamin D deficiency was equally prevalent among veiled (4.77 ± 1.37 ng/mL) and nonveiled (4.75 ± 1.68 ng/mL). Treatment with high-dose vitamin D resulted in clinical improvement in all patients. Conclusion. Vitamin D deficiency is often seen in patients diagnosed with fibromyalgia in our population. This was equally true in veiled and nonveiled, but conservatively dressed populations. Effective treatment with high-dose vitamin D could lead to resolution of almost all symptoms. Further study of these populations and fortification of foods with vitamin D may be essential.

Heidari, B., Shirvani, J. S., Firouzjahi, A., Heidari, P., & Hajian-Tilaki, K. O. (2010). Association between nonspecific skeletal pain and vitamin D deficiency. *International Journal of Rheumatic Diseases*, *13*(4), 340-346. doi:10.1111/j.1756-185X.2010.01561.x

Background: Deficiency of vitamin D has been reported in patients with many types of musculoskeletal pain. The present study was designed to determine the association between serum 25-hydroxyvitamin D [25-(OH)D] deficiency and nonspecific skeletal pain. Methods: A total of 276 patients with nonspecific skeletal pain at different regions of the skeletal system diagnosed as leg pain, widespread pain, arthralgia, rib pain, back pain and fibromyalgia were compared with 202 matched controls with regard to mean serum 25-(OH)D level and 25-(OH)D deficiency. Serum 25-(OH)D was measured by enzyme-linked immunosorbent assay method and levels < 20 ng/mL were considered as deficient. Nonparametric one-way analysis of variance, Kruskal Wallis and Wilcoxon tests were used for group comparisons. Multiple logistic regression analysis with calculation of adjusted odds ratio (OR) and 95% confidence interval (95% CI) were performed to determine associations. Results: In patients with nonspecific skeletal pain the mean 25-(OH)D was significantly lower (P = 0.0001) and the proportion of 25-(OH)D deficiency was significantly higher (63.4%vs. 36.1%, P = 0.0001) compared with controls. There was a significantly positive association between 25-(OH)D deficiency and skeletal pain (OR = 2.94, 95% CI = 1.01-4.3, P = 0.0001). The strength of association varied across the groups with strongest association observed with leg pain (OR = 7.4; 95% CI = 3.9-13.9, P = 0.0001)

followed by arthralgia (OR = 3.9, 95% CI = 2.1-7.1, P = 0.0001) and widespread pain (OR = 2.8, 95% CI = 1.1-6.6, P = 0.020) but no association with back pain and fibromyalgia. There was a greater positive associations in women compared with men (OR = 2.1, 95% CI = 1.1-4.3, P = 0.001). Conclusion: The results of this study indicate a positive association of vitamin D deficiency with a variety of nonspecific bone pain, particularly in women. More studies with larger samples are required to confirm these findings. Increasing serum vitamin D to sufficient levels and longitudinal follow-up of patients may provide further evidence in relation to vitamin D deficiency and skeletal pain.

McCabe, P. S., Pye, S. R., Mc Beth, J., Lee, D. M., Tajar, A., Bartfai, G., & ... Boonen, S. (2016). Low vitamin D and the risk of developing chronic widespread pain: results from the European male ageing study. *BMC Musculoskeletal Disorders*, *17*1-9. doi:10.1186/s12891-016-0881-6

The association between low levels of vitamin D and the occurrence of chronic widespread pain (CWP) remains unclear. The aim of our analysis was to determine the relationship between low vitamin D levels and the risk of developing CWP in a population sample of middle age and elderly men. Methods: Three thousand three hundred sixty-nine men aged 40-79 were recruited from 8 European centers for a longitudinal study of male ageing, the European Male Ageing Study. At baseline participants underwent assessment of lifestyle, health factors, physical characteristics and gave a fasting blood sample. The occurrence of pain was assessed at baseline and follow up (a mean of 4.3 years later) by shading painful sites on a body manikin. The presence of CWP was determined using the ACR criteria for fibromyalgia. Serum 25-hydroxyvitamin D (25-(OH) D) was assessed by radioimmunoassay. Logistic regression was used to determine the relationship between baseline vitamin D levels and the new occurrence of CWP. Results: Two thousand three hundred thirteen men, mean age 58.8 years (SD = 10.6), had complete pain and vitamin data available and contributed to this analysis. 151 (6.5 %) developed new CWP at follow up and 577 (24.9 %) were pain free at both time points, the comparator group. After adjustment for age and center, physical performance and number of comorbidities, compared to those in upper quintile of 25-(OH) D (≥36.3 ng/mL), those in the lowest quintile (<15.6 ng/mL) were more likely to develop CWP (Odds Ratio [OR] = 1.93; 95 % CI = 1.0-3.6). Further adjustment for BMI (OR = 1.67; 95 % CI = 0.93-3.02) or depression (OR = 1.77; 95 % CI = 0.98-3.21), however rendered the association non-significant. Conclusions: Low vitamin D is linked with the new occurrence of CWP, although this may be explained by underlying adverse health factors, particularly obesity and depression.

von Känel, R., Müller-Hartmannsgruber, V., Kokinogenis, G., & Egloff, N. (2014). Vitamin D and Central Hypersensitivity in Patients with Chronic Pain. *Pain Medicine*, *15*(9), 1609-1618. doi:10.1111/pme.12454

Low vitamin D is implicated in various chronic pain conditions with, however, inconclusive findings. Vitamin D might play an important role in mechanisms being involved in central processing of evoked pain stimuli but less so for spontaneous clinical pain. Objective This study aims to examine the relation between low serum levels of 25-hydroxyvitamin D3 (25- OH D) and mechanical pain sensitivity. Design We studied 174 patients (mean age 48 years, 53% women) with chronic pain. A standardized pain provocation test was

applied, and pain intensity was rated on a numerical analogue scale (0-10). The widespread pain index and symptom severity score (including fatigue, waking unrefreshed, and cognitive symptoms) following the 2010 American College of Rheumatology preliminary diagnostic criteria for fibromyalgia were also assessed. Serum 25- OH D levels were measured with a chemiluminescent immunoassay. Results Vitamin deficiency (25- OH D < 50 nmol/L) was present in 71% of chronic pain patients; another 21% had insufficient vitamin D (25- OH D < 75 nmol/L). After adjustment for demographic and clinical variables, there was a mean ± standard error of the mean increase in pain intensity of 0.61 ± 0.25 for each 25 nmol/L decrease in 25- OH D (P = 0.011). Lower 25- OH D levels were also related to greater symptom severity (r = −0.21, P = 0.008) but not to the widespread pain index (P = 0.83) and fibromyalgia (P = 0.51). Conclusions The findings suggest a role of low vitamin D levels for heightened central sensitivity, particularly augmented pain processing upon mechanical stimulation in chronic pain patients. Vitamin D seems comparably less important for self-reports of spontaneous chronic pain.