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I. Introduction

Flavonoids represent one of major polyphenolic groups, which are widely distributed in the plant kingdom and commonly found in vegetables, fruits and teas of the human diet[1]. These compounds have shown low toxicity as well as a variety of physiological effects[1]. In the past few decades, these naturally occurring compounds have attracted much attention by reported beneficial effects on health, such as antioxidant, antitumor, anti-inflammatory and anti-allergic effects[2], as well as presenting low toxicity to human organism[1,3]. In midst the various classes of flavonoids, the flavonols have been the focus of many *in vitro* and *in vivo* studies for their diversified actions on numerous biological pathways[1]. Structurally, flavonoids are composed of a basic flavylium cation, with three phenolic rings[4]. Flavonols present subtle molecular changes to the main ring (Ring C), with the addition of a hydroxyl group (OH) on the third carbon, and a carbonyl group (C=O) on carbon fourth [1,5]. Many reports have shown that structure changes of flavonoid molecules result in variations of bioavailability, by changing absorption efficiency and pathways; thus, also altering biological effects and creating action mechanisms specificity [1,4,6–8].

Flavonoids neuroprotective effects are mostly bound to its related antioxidant, anti-proliferative and anti-inflammatory properties[9,10]. The Central Nervous System (CNS) is highly exposed to oxidative damage, due to high oxygen consumption, high levels of unsaturated lipids and presence of transitional metals[1]. In addition, because of inefficient antioxidant defense mechanisms, alterations of neuronal organic homeostasis can cause grave repercussions[1,9]. Antioxidant properties are correlated since oxidative stress and lipid peroxidation have been linked to a range of neurological pathologies, such as brain trauma, ischemia and neurodegenerative disorders [2,11,12]. Thus, flavonoids ability to scavenge ROS and inhibit lipid peroxidation, protecting neuronal cells from oxidative damage, may be used to prevent and treat neurological pathologies [2].Neurodegenerative disorders, such as Alzheimer, Parkinson's and Huntington's disease cause a progressive functional alteration of neuronal systems[13]. Worldwide, they present variable incidence and constantly relate to high morbidity rates, higher cost in health care and social impairment[13–15]. These pathologies have been well studied; however, new advances in physiopathology and, therefore, in therapeutic modalities are infrequent, maintaining no cure or reversible treatments.

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Other flavonoids, such as resveratrol, fisetin and hyperin have also been considered as potential drugs with neuroprotective effects[9,13,16–22]. Recent studies verified, different flavonoids act on specific organic pathways [1], yet comparable studies between flavonoids are rare. Resveratrol, as an example, has shown neurological and cognitive enhancement properties in a clinical trial[23]. Quercetin and rutin have been widely investigated as therapeutic drugs, especially concerning anti-proliferative and antioxidant effects, for many diseases, including CNS pathologies. However, neuroprotective potential of flavonoids in human trials has been poorly addressed, and Isoquercitrin has rarely been implied in studies investigating neuroprotective effects and comparison studies of the same subject.

II. Methods/Research

a) *Litterature Review*

This paper was based on a literature search of PubMed and Scielo databases using keyword combinations "Flavonoids+Neuroprotecition"; "Quercetin OR Rutin OR Isoquercitrin + Neuroprotection"; "Quercetin OR Rutin OR Isoquercitrin + Alzheimer OR Parkinson OR Huntington", during the period of 2000 to 2016. Clinical trials were assessed in each search. Studies involving flavonoid panel and flavonoid combinations were considered.

b) *Literature Evidence*

Evidence Based Medicine (EBM) is a systematic analysis of present-day research and scientific findings, in which obtained information is classified by authenticity and Evidence Level results [24]. Thus creating a hierarchy system to evaluate information and incorporate in a practical environment of research, as well as to support the conduct of clinical care and therapeutic options[25]. Recommendation levels are the results of extensive research analysis and an important guide to practitioner's clinical decisions[26]. Although animal experiments have always contributed to our understanding of drug action mechanisms and pathophysiological aspects of many diseases[27], some authors debate on whether animal researches are valuable predictors of human conditions and pathogenesis, due to interspecies differences and lack of uniform requirements for reporting animal data and comparable results[28]. Considering the pyramid of medical research, animal experimentation and in vitro studies take their place at the bottom, reflecting basic studies; however, tools of assessment of these studies have emerged in the last decade to improve transparency and accuracy of reports[29]. One of which, ARRIVE guidelines (Animals in Research: Reporting *in Vivo* Experiments)creates a checklist of 20 steps to be met during animal experimentation reports[29]. Although ARRIVE checklist is not mentioned in many animal reports, in the past five years endorsement of this tool amongst journals has risen considerably[30], becoming an important source of evaluation. Thus, it was this paper's intent to grade Animal Experimentation and *In Vitro* Studies´ Evidence Level, through a personal classification (Table 1), derived from the Oxford Centre for Evidence Based Medicine (CEBM). In addition, apply the ARRIVE checklist to*in vivo* animal studies referenced in this paper.

Table 1: Evidence Level adapted from Oxford CEBM to evaluate therapeutic value of scientific studies.

LEVEL	THERAPY
1A	Systematic Reviews of Randomized Controlled Trials (RCT)
1B	Individual RCT with narrow Confidence Interval
1C	All or None Studies
1D*	*In Vivo* Animal Trials
2A	Systematic Reviews of Cohort Studies
2B*	I. Cohort Studies (*In Human*)
	II. *Ex Vivo* Studies
2C*	Outcome Research or Ecological Studies or *In Vitro* Studies
3A	Systematic Review of Case-Control Studies
3B*	Case-Control Studies or Drug Biological Characteristics Assessment Studies
4*	Case-series and poor quality cohort and case-control studies or Literature Review
5	Expert opinion without explicit critical appraisal
*Modified items from Oxford CEBM	

c) *Flavonoids*

Flavonoids represent one of major polyphenolic groups, which are widely distributed in the plant kingdom and commonly found in vegetables, fruits and teas of the human diet[1]. Recent experimental studies and clinical trials conducted with flavonoids, in particular the class of flavonols, have demonstrated a wide variety of physiological effects, including antioxidant, anti-proliferative/antitumor and anti-inflammatory, also correlating some of their action mechanisms to neuroprotective potential. Thus evidencing this class of molecules as an accessible alternative of preventive

therapy or treatment of numerous neurological diseases. Flavonoids can be found in nature as aglycone forms, glycosides or methylated/acylated derivatives[1]. Amongst flavonols representatives, the best-known molecules are (1) Quercetin, an important aglycone form and a pioneer subject mid flavonoid research; (2) Rutin, main hydrophilic glycoside molecule; and (3) Isoquercitrin (IQ), main lipophilic glycoside, also known as quercetin-3-O-glucoside (Q3G) and Isoquercetin, a nearly identical quercetin-3-monoglucoside[1,31].

Flavonoids absorption occur predominantly in the small intestine, however it is limited by molecular weight and hydrophilicity[1,31]. Few studies have been conducted as to elucidate bioavailability and absorption across the blood-brain barrierin human models[32,33] **(Evidence Level 1B)**, whereas most studies have used *in vivo* animal models, biological differences and lack of complete physiological understanding of flavonoid's absorption in the small intestine,have limited new findings. Nevertheless, the type of sugar moiety attached to primary aglycone molecule, has been named the major determinant of small intestine absorption, rather than its position in the same molecule[34].

In food plants, quercetin occurs almost exclusively as glycosides, in which the and the dominant type of glycoside vary amongst foods and is usually located at the 3 or 4 position of the pyrone ring[1,34]. Onions, kale, broccoli and apples are important sources of glucoside molecules, such as Rutin[35]. IQ is a common naturally occurring glycoside also obtained by enzymatic hydrolysis of rutin with hesperedinase, an enzyme produced by specific types of fungisuch as *Penicillium sp.*

Heperedinase has a α-l-ramnosidase selective activity when applied at 58°C for 30 minutes, capable of cleaving the rhamnosidase radical of rutin's basic structure and leaving the glucoside radical intact, transforming it into IQ[36]. This procedure, generates what is called enzymatic modified IQ or Hydrolysed Rutin (HR), both of which consists of a mixture that includes IQ (69,5%), quercetin (7,5%), rutin and other small metabolites[36]. Studies have shown the superiority of anti-oxidant and anti-proliferative properties of HR when compared solely to quercetin and rutin **(Evidence Level 2C)**[36,37].

III. Neurodegenerative Diseases

Neurodegenerative diseases are a consequence of genetic and environmental factors that are strongly associated with age[38]. These disorders arise from multifactorial conditions that interfere directly with cellular oxidative homeostasis and function[38]. Amongst various pathophysiological factors, increased oxidative stress, mitochondrial dysfunction and abnormal protein dynamics represent a common role in the development of different neurodegenerative diseases, such as Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's disease (HD) and amyotrophic lateral sclerosis (ALS)[38]. Also appearing to command numerous neuronal pathways, leading to alterations in neurotransmission and ionic channels, protein aggregation, impaired bioenergetics and even cellular death[38].

Free radicals are indispensable molecules to cellular function by involvement in many biochemical activities[39]. However, oxidative stress arise from disturbances between pro-oxidant/antioxidant equilibrium[39]. Overproduction of free radicals produce cytotoxicity and genotoxicity by damaging biomolecules, such as proteins, lipids and DNA[39] and leading to arise of various chronic diseases, including in the CNS[39]. In the brain glial and neuronal cells are particularly sensitive to free radicals and actively targeted by ROS[39]. Primary neuroprotective potential of flavonoids rely mainly on their antioxidant effect and leading mechanism of action seems to be reduction of cellular oxidative stress by scavenging and decreasing reactive oxygen species (ROS) and reactive nitrogen species (RNS) in brain tissue**(Evidence Level 2C)**[2,9,40–43]. Also exhibiting activation of anti-oxidant enzymes (**Evidence Level 1B)**[44] **(Evidence Level 2C)**[45,46], protein disaggregation and diminished production **(Evidence Level 2C)**[47], decrease of lipid peroxidation **(Evidence Level 1B)**[48] **(Evidence Level 2C)**[42], transitional metal chelation **(Evidence Level 2C e 4)**[2,49–51] and anti-inflammation effects **(Evidence Level 1B)**[52]. Although, Ansari et al. (2009) demonstrated in an *in vitro* study that quercetin showed dual effect on oxidative stress; at lower dosages, antioxidant effects were observed, though in higher dosages, it exhibited a pro-oxidant effect also increasing neuronal dysfunction, it is still clear flavonoids present oxidative-protective effects on various diseases, including those of the CNS.

a) Alzheimer's Disease

Alzheimer's disease (AD) is a complex neurological disorder clinically characterized by progressive loss of memory and cognition; histopathology demonstrates accumulation of extracellular β-amyloid plaques (major constituent of senile plaques), intracellular neurofibrillary tangles, tau protein phosphorylation and neurodegeneration of synaptic neurons, especially in the hippocampus[13,53].Oxidative stress and neuro-inflammation are also considered hallmarks of AD, mainly responsible for increasedneurotoxicity[54] of cognitive-modulating areas [55]. In addition, the decrease in cholinergic neurotransmission has also been implicated in cognitive decline and behavioral changes of AD[56].

Flavonoids represent an interesting class of phytochemicals and, even before scientific studies, have

been widely used as phytotherapy medicines, like Ginkgo biloba, which includes quercetin and kaempferol[57]. In 2009, Shi et al. demonstrated quercetin molecule was responsible for high antioxidant activity of this plant extract, corroborating to the hypothesis of flavonoids, in particular flavonols, as possible therapeutic drugs with neuroprotective potential **(Evidence Level 2C)**[58].

A main pathophysiological mechanism of AD is the transformation of β-amyloid peptides into amyloid-beta (Abeta) plaques and the extracellular aggregation of plaques in areas such as the hippocampus[47], which appear to be an early phenomenon and is currently used as a diagnostic tool of the disease through PET scan markers **(Evidence Level 2A)**[59]**.** In the last few years, some studies have comprised flavonoid's neuroprotective potential and investigated anti-amyloidogenic properties. **Ansari et al. (2009)** demonstrated quercetin's protective effect under low dosages (5 μM and 10 μM), strongly inhibiting Abeta fibril formation and preventing glutathione oxidation **(Evidence Level2C)**[60]. In **2011, Jimenez-Aliaga** evaluated anti-amyloidogenic potential of both quercetin and rutin, demonstrating inhibition of Abeta fibrils formation and disaggregation by both compounds, however, noticing statistical superiority of quercetin's effects. Also reporting the reduction of ROS production and lipid peroxidation index **(Evidence Level2C)**[47]. In addition, **Choi et al. (2014)** studied the*in vitro* activity of a flavonoid panel, including quercetin and rutin, against Abeta-induced toxicity. Results corroborated previous studies showing that flavonoids significantly block Aβ-induced neuronal toxicity**(Evidence Level 2C)**[61].

Liu et al., 2013 investigated *in vivo* the protective effects of quercetin against Abeta-induced toxicity, on both endothelial cells and neurons, after oral administration of quercetin for a period of 8 days; learning and memory were evaluated by Morris Water Maze test, and cerebral flow was closely monitored. Results showed neurovascular coupling protection, with reduction of oxidative stress and maintenance of neurovascular unit**(Evidence Level 1D)**[62].

Recent *in vitro* and *in vivo* studies investigated neuroprotective effects of quercetin on Abeta-induced toxicity models. Results showed quercetin improved cell viability, by diminishing neuronal and endothelial oxidative stress**(Evidence Levels 2C, 1D)**[63,64], and the production of ROS and LDH were decreased, as an increase on superoxide dismutase occurred **(Evidence Level2C)**[63]. **Mohebali et al., 2016** demonstrated both Rutin's and Quercetin's potential to down regulate inflammation-involved genes in AD**(Evidence Level 1D.I)**[65]. Moreover, **Sabogal- Guáqueta et al.**, performed an *in vivo* study with quercetin (25mg/kg) i.p. administration, for 3 months,on triple transgenic AD model mice, observing a decrease of extracellular β-amyloid deposition, as well as a reduction of tau phosphorylation, astrogliosis and microgliosis in the hippocampus and amygdala**(Evidence Level 1D)**[66].

Isolated, rutin has been less studied in AD over the years. **In 2012, Javed et al.** conducted the investigation of rutin's neuroprotective antioxidant effects in an *in vivo* intracerebroventricular-streptozotocin (ICV-STZ) induced toxicity model. Rutin was pre-administered orally (25mg/kg) for 3 weeks, and results indicated attenuation of STZ-induced inflammation by reducing the expression of cyclooxygenase-2 (COX-2), interleukin-8 (IL-8) and nuclear factor-kB, thus preventing neuro-inflammatory morphological changes in the hippocampus**(Evidence Level 1D)**[67].

In addition, recent studies that comprised rutin's antioxidant activity on Abeta-induced neurotoxicity showed the decrease of ROS and RNS as the main action mechanism, consequently reducing lipid peroxidation. Interestingly, rutin was also capable of diminishing glutathione levels and dependent enzymes, also downregulating astrocytosis and microgliosis and, therefore, proving to having similar effects to quercetin **(Evidence Levels 1D)**[68–70].

Another hallmark characteristic of AD is the decrease in cholinergic neurotransmission, which has been implicated in cognitive decline, leading to dementia, and behavioral disorders[56]. The increase on cholinergic neurotransmission is an important focus of recent drug therapy and comprises the inhibition of acetylcholinesterase (AchE), acetylcholine's degrading enzyme [71]. Both quercetin and rutin have been targets of recent studies focusing on AchE inhibition, since it is an important target on Alzheimer's drug therapy. Quercetin is a strong AchE inhibitor**(Evidence Levels 1D, 2B.II)**[72,73], presenting higher binding strength to active site of the enzyme then some of the drugs in the market, like Donezepil[74]; also acting in a dose-dependent manner**(Evidence Level 3B)**[42]. Rutin also seems to show AchE inhibition properties**(Evidence Level 3B)**[75],however no study has solely involved this compound. There are no studies indexed on PubMed or Scielo involving isoquercitrin's specific neuroprotective effects in AD. Nevertheless, IQ exhibits important antioxidant activity[76] and has been implied as a promising molecule for the treatment of several pathologies, especially cancer[77]**.**

b) Parkinson's Disease

Parkinson's disease (PD) is a neurodegenerative movement disorder mainly characterized by progressive loss of dopaminergic neurons within nigrostriatal pathway, affecting *substantia nigra* area, and associated with microglial-mediated neuro-inflammation[22]. Pathogenic aspects of PD involve mitochondrial dysfunction, changes in micro-RNA and α-synuclein levels[13], a major constituent of Lewy bodies and a hallmark of PD[78]. Oxidative stress has been

associated as a risk factor for dopamine cellular degeneration of PD[79], and mitochondrial dysfunction plays an important role on energetic balance as well as regulating oxidative and apoptotic pathways[38].

Quercetin has been widely investigated for its antioxidant effects; however, specific PD model investigations are scarce. On chronic rotenone (ROT)-induced parkinsonian models quercetin (25-75mg/kg i.p.) showed reduced loss of ROT-induced dopaminergic neurons, also decreasing glutathione levels and increasing anti-oxidant enzymes (catalase and superoxide dismutase)**(Evidence Level 2B)**[80]. In 2015, Denny Joseph et al., also investigated on ROT-induced models the beneficial effects of treatment association between fish oil and quercetin. Results corroborated previous studies demonstrating significant behavioral change as well as attenuation of oxidative stress and mitochondrial dysfunction indicators **(Evidence Level 1D)**[81].

Other studies using methyl-4-phenylpyridinium ion (MPP(+)), a parkinsonian toxin that provokes degeneration of dopaminergic neurons[82], and 6-hydroxydopamine (6-OHDA), a selective dopaminergic neurotoxin[83]. Also demonstrated quercetin's neuroprotective potential by reducing apoptotic neuronal death, through modulation of pro-apoptotic (Bax) and anti-apoptotic (Bcl-2) genes, and decreasing oxidative stress and neuro-inflammation (IL-1, TNF-α and COX-2) on microglial cells **(Evidence Levels 2C, 2C, 1D, 1D)**[82–85]. Although these results are consistent with literary review appointing quercetin as an antioxidant molecule, some studies have shown a paradoxical effect of this compound when used in high dosages **(Evidence Level 2C)**[60].

In addition, in 2015, Ahn et al. investigated quercetin's specific effect on α-synuclein expression. During the experiment, PC12 cells were pre-treated with quercetin and results showed quercetin presents neuroprotective effects affecting various mechanisms such as apoptosis and oxidative stress; however, quercetin treatment increased α-synuclein levels, and although cell viability and survival were unaltered with the up-expression, this data suggests quercetin's effects on PD is still not entirely understood**(Evidence Level 2C)**[78].

During the course of 2014 to 2016, Magalingam et al., produced a series of three articles investigating Rutin and Isoquercitrin on 6-OHDA PD-induced models. Both rutin and IQ demonstrated antioxidant effects by reducing lipid peroxidation and increasing anti-oxidant enzymes (catalase, superoxide dismutase and glutathione peroxidase) **(Evidence Level 2C)**[86–88]. Cytoprotective activity of IQ occurred in a dose-dependent manner. Furthermore, Rutin showed upregulation of the TH gene, an important factor in dopamine biosynthesis; as well as modulation of apoptotic pathways by reducing Park2, Park5, Park7, Caspase 3 and Caspase 7 genes**(Evidence Level 2C)**[86].

c) Huntington's Disease

Huntington's disease (HD) is an autosomal dominant, progressive, neurodegenerative disorder clinically characterized by motor, cognitive and behavioral impairment, also presenting high morbidity and mortality rates[89]. Incidence is higher at European countries and mean age of symptoms onset occurs around 40 years old [90]. HD is cause by an expanded CAG trinucleotide repeat in the HTT gene, responsible for encoding the protein huntingtin. The mutation lead to the production of an abnormal protein with long polyglutamine sequences that confers toxic properties and predisposes protein fragmentation, which can result in neuronal death[91].

Quercetin is the only major flavonol whose neuroprotective effect has been associated to Huntington's disease treatment potential. In this review, we found no articles investigating rutin or isoquercitrin effects on HD models. In 2013, **Sandhir et al.** evaluated oral supplementation of quercetin (25mg/kg) on 3-nitropropionic acid-induced (3-NP) Huntington's disease animal model. Posterior analysis was conducted on mitochondrial biogenetics, oxidative stress, neurobiological behaviors and histopathological assays. It was proven quercetin exhibits protective effects by attenuating mitochondrial oxidative stress (reduced lipid peroxidation and mitochondrial swelling), as well as increasing motor skills and antioxidant elements **(Evidence Level 2B)**[92]. Additionally, in 2014, quercetin was once again tested on 3-NP-induced HD models, confirming previous results of antioxidant properties and motor coordination increase, along with display of behavioral changes throughlessening of anxiety and isolation;also reducing neuro-inflammatory responses with an increase of astrocyte numbers in core lesions and decreased microglial proliferation **(Evidence Level 1D)**[93,94].

Table 2: ARRIVE Checklist evaluation of *in vivo* Animal Studies referenced

	Study	*Year*	*Flavonoid Tested*	*ARRIVE Checklist Items Met*	*ARRIVE Checklist Items Not-Met*
Alzheimer's Disease	Liu et al.[62]	2013	Quercetin	1-9, 10a, 11b, 12-20	10b, 11a
	Hayakawa et al.[64]	2015	Quercetin	1,2,3a, 4-9, 11-13, 15-20	3b, 10, 14
	Sabogal-Guaqueta et al.[66]	2015	Quercetin	1,2,3a, 4-8,11b, 12,13, 16-20	3b, 8a, 10, 11a, 14,15
	Javed et al.[67]	2012	Rutin	1, 2, 3a, 4-9, 10a, 12, 13, 15-20	3b, 10b, 10c, 11, 14
	Choi et al.[68]	2015	Rutin	1-9, 10a, 11-20	10b, 10c
	Xu et al.[69]	2014	Rutin	1, 2, 3a, 4-9, 10a, 11-13, 15-20	3b, 10b, 10c, 14
	Moghbelinejad et al.[70]	2014	Rutin	1,2,3a, 4, 5, 6a, 6c, 7, 8b, 9, 12, 13, 15-20	3b, 6b, 8a, 10, 11, 14
	Abdalla et al[72]	2014	Quercetin	1-9, 10a, 11-19	10b, 10c, 20
Parkinson's Disease	Karuppagounder et al.[80]	2013	Quercetin	1-9, 12-14, 16, 17-20	10, 11, 15
	Zhang et al.[83]	2011	Quercetin	1-5, 8, 10, 12-14, 16-20	6, 7, 9, 11, 15
	Haleagrahara et al.[85]	2013	Quercetin	1-20	-
Huntington's Disease	Jain et al.[93]	2014	Quercetin	1, 2, 3a, 4-20	3b
	Chakraborty et al.[94]	2014	Quercetin	1-5, 6a, 6b, 7-10, 12-14, 16-20	6c, 11, 15

IV. Conclusion

Flavonoids have been widely investigated in the past decades and have shown a wide variety of physiological effects, determining a therapeutic potential on innumerous diseases, including neurological pathologies. Their neuroprotective effects are mostly related to anti-oxidant and anti-inflammatory properties; however, specific mechanisms have been reached on both *in vitro* and *in vivo* animal models of neurodegenerative diseases. It is quite difficult to assess on whether animal experimentation is most likely to predict human outcomes and toxicity, nonetheless, it is a vital part of scientific research and discovery. Evidence-based medicine analyses such studies in order to foresee favorable outcomes, and although most studies conducted with flavonols are *in vitro* and animal models of experimentation (Evidence Level 1D/2B and 2C), ARRIVE guidelines offer a tool of assessment, in which the goal is to improve transparency and accuracy of these reports. Our findings on the subject suggest *in vitro* studies are still the majority of literature references, yet *in vivo* animal experimentation references seems to be well-constructed(Table 2) and able to provide key results, leading to the possibility of human clinical trials. Despite the lack of human trials with Quercetin, Rutin and Isoquercitrin, other flavonoids have been tested and results show neurological features, providing a glimpse of the therapeutic potential of these compounds. We also suggest Isoquercitrin as a viable option to future experiments, due to its superiority of anti-oxidant and anti-proliferative properties.

References Références Referencias

1. Appleton J. Evaluating the Bioavailability of Isoquercetin. *Nat Med J*. 2010;2(1):2-6.
2. Dhiman A, Nanda A, Ahmad S. A quest for staunch effects of flavonoids: Utopian protection against hepatic ailments. *Arabian Journal of Chemistry*. http://dx.doi.org/10.1016/j.arabjc.2012.05.001. Published 2012.
3. Amado NG, Predes D, Moreno MM, Carvalho IO, Mendes F a, Abreu JG. Flavonoids and Wnt/β-catenin signaling: potential role in colorectal cancer therapies. *Int J Mol Sci*. 2014; 15(7): 12094-12106. doi:10.3390/ijms150712094.
4. Aherne SA, O'Brien NM. Dietary flavonols: chemistry, food content, and metabolism. *Nutrition*. 2002; 18(1): 75-81. http://www.ncbi.nlm.nih.gov/pubmed/11827770.
5. Hopia A, Heinonen M. Antioxidant activity of flavonol aglycones and their glycosides in methyl linoleate. *J Am Oil Chem Soc*. 1999; 76(1): 139-144. doi:10.1007/s11746-999-0060-0.
6. Heim KE, Tagliaferro AR, Bobilya DJ. Flavonoid antioxidants: Chemistry, metabolism and structure-activity relationships. *J Nutr Biochem*. 2002; 13(10): 572-584. doi:10.1016/S0955-2863(02)00208-5.
7. Liaudanskas M, Viškelis P, Raudonis R, Kviklys D, Uselis N, Janulis V. Phenolic Composition and Antioxidant Activity of Malus domestica Leaves. *Sci World J*. 2014; 2014: 1-10. doi:10.1155/2014/3062 17.
8. Reinboth M, Wolffram S, Abraham G, Ungemach FR, Cermak R. Oral bioavailability of

quercetin from different quercetin glycosides in dogs. *Br J Nutr*. 2010; 104(2): 198-203. doi:10.1017/S000711451000053X.
9. Subash S, Essa MM, Al-Adawi S, Memon MA, Manivasagam T, Akbar M. Neuroprotective effects of berry fruits on neurodegenerative diseases. *Neural Regen Res*. 2014; 9(16): 1557-1566. doi: 10.4103/1673-5374.139483.
10. Costa LG, Garrick JM, Roque PJ, Pellacani C. Mechanisms of Neuroprotection by Quercetin: Counteracting Oxidative Stress and More. *Oxid Med Cell Longev*. 2016; 2016: 2986796. doi:10.1155/2016/2986796.
11. Razavi SM, Zahri S, Zarrini G, Nazemiyeh H, Mohammadi S. Biological activity of quercetin-3-O-glucoside, a known plant flavonoid. *Bioorg Khim*. 2009; 35(3): 414-416. doi:10.1134/S1068162009030133.
12. Boligon AA, Sagrillo MR, Machado LF, et al. Protective effects of extracts and flavonoids isolated from scutia buxifolia reissek against chromosome damage in human lymphocytes exposed to hydrogen peroxide. *Molecules*. 2012; 17(5): 5757-5769. doi:10.3390/molecules17055757.
13. Bhullar KS, Rupasinghe HPV. Polyphenols: multipotent therapeutic agents in neurodegenerative diseases. *Oxid Med Cell Longev*. 2013; 2013: 891748. doi:10.1155/2013/891748.
14. Gazdik Z, Reznicek V, Adam V, et al. Use of liquid chromatography with electrochemical detection for the determination of antioxidants in less common fruits. *Molecules*. 2008; 13(11): 2823-2836. doi:10.3390/molecules131102823.
15. Solanki I, Parihar P, Parihar MS. Neurodegenerative diseases: From available treatments to prospective herbal therapy. *Neurochem Int*. 2015. doi:10.1016/j.neuint.2015.11.001.
16. Zeng K, Wang X, Fu H, Liu G. [Protective effects and mechanism of hyperin on CoCl2-induced PC12 cells]. *Zhongguo Zhong Yao Za Zhi*. 2011;36(17):2409-2412.
17. Moosavi F, Hosseini R, Saso L, Firuzi O. Modulation of neurotrophic signaling pathways by polyphenols. *Drug Des Devel Ther*. 2015; 10: 23-42. doi:10.2147/DDDT.S96936.
18. Ahmad A, Ali T, Park HY, Badshah H, Rehman SU, Kim MO. Neuroprotective Effect of Fisetin Against Amyloid-Beta-Induced Cognitive/Synaptic Dysfunction, Neuroinflammation, and Neurodegeneration in Adult Mice. *Mol Neurobiol*. 2016. doi:10.1007/s12035-016-9795-4.
19. Cho N, Choi JH, Yang H, et al. Neuroprotective and anti-inflammatory effects of flavonoids isolated from Rhus verniciflua in neuronal HT22 and microglial BV2 cell lines. *Food Chem Toxicol*. 2012;50(6):1940-1945. doi:10.1016/j.fct.2012.03.052.
20. Kelsey NA, Wilkins HM, Linseman DA. Nutraceutical antioxidants as novel neuroprotective agents. *Molecules*. 2010; 15(11): 7792-7814. doi:10.3390/molecules15117792.
21. Peritore CS, Ho A, Yamamoto BK, Schaus SE. Resveratrol attenuates L-DOPA-induced hydrogen peroxide toxicity in neuronal cells. *Neuroreport*. 2012; 23(17): 989-994. doi:10.1097/WNR.0b013e32835a4ea4.
22. Bureau G, Longpre F, Martinoli M-G. Resveratrol and quercetin, two natural polyphenols, reduce apoptotic neuronal cell death induced by neuroinflammation. *J Neurosci Res*. 2008;86(2):403-410. doi:10.1002/jnr.21503.
23. Witte AV, Kerti L, Margulies DS, Flöel A. Effects of resveratrol on memory performance, hippocampal functional connectivity, and glucose metabolism in healthy older adults. *J Neurosci*. 2014;34(23):7862-7870. doi:10.1523/JNEUROSCI.0385-14.2014.
24. Masic I, Miokovic M, Muhamedagic B. Evidence Based Medicine - New Approaches and Challenges. *Acta Inform Medica*. 2008;18(1):219. doi:10.5455/aim.2008.16.219-225.
25. Burns PB, Rohrich RJ, Chung KC. The Levels of Evidence and Their Role in Evidence-Based Medicine. *Plast Reconstr Surg*. 2011; 128(1): 305-310. doi:10.1097/PRS.0b013e318219c171.
26. MEDEIROS L., STEIN A. Níveis de evidência e graus de recomendação da medicina baseada em evidências. *Rev AMRIGS*. 2002;46(1,2):42-46.
27. Van der Worp HB, Howells DW, Sena ES, et al. Can Animal Models of Disease Reliably Inform Human Studies? *PLoS Med*. 2010; 7(3): e1000245. doi:10.1371/journal.pmed.1000245.
28. Pound P, Bracken MB. Is animal research sufficiently evidence based to be a cornerstone of biomedical research? *BMJ*. 2014; 348 (may30 1): g3387-g3387. doi:10.1136/bmj.g3387.
29. Sut N. Study Designs in Medicine. *Balkan Med J*. 2015; 31(4): 273-277. doi:10.5152/balkanmedj.2014.1408.
30. Cressey D. Surge in support for animal-research guidelines. *Nature*. 2016. doi:10.1038/nature.2016.19274.
31. Murota K, Shimizu S, Chujo H, Moon JH, Terao J. Efficiency of absorption and metabolic conversion of quercetin and its glucosides in human intestinal cell line Caco-2. *Arch Biochem Biophys*. 2000;c 384(2): 391-397. doi:10.1006/abbi.2000.2123.
32. Kaushik D, O'Fallon K, Clarkson PM, Dunne CP, Conca KR, Michniak-Kohn B. Comparison of quercetin pharmacokinetics following oral supplementation in humans. *J Food Sci*. 2012; 77(11): H231-8. doi:10.1111/j.1750-3841.2012.02934.x.
33. Cialdella-Kam L, Nieman DC, Sha W, Meaney MP, Knab AM, Shanely RA. Dose-response to 3 months

of quercetin-containing supplements on metabolite and quercetin conjugate profile in adults. *Br J Nutr*. 2013; 109(11): 1923-1933. doi:10.1017/S0007114512003972.

34. Arts ICW, Sesink ALA, Faassen-Peters M, Hollman PCH. The type of sugar moiety is a major determinant of the small intestinal uptake and subsequent biliary excretion of dietary quercetin glycosides. *Br J Nutr*. 2004; 91(6): 841-847. doi:10.1079/BJN20041123.
35. Hollman PC, Arts IC. Flavonols, flavones and flavanols - nature, occurrence and dietary burden. *J Sci Food Agric*. 2000; 80(7): 1081-1093. doi:10.1002/(SICI)1097-0010(20000515)80:7<1081::AID-JSFA566>3.0.CO;2-G.
36. De Araújo MEMB, Moreira Franco YE, Alberto TG, et al. Enzymatic de-glycosylation of rutin improves its antioxidant and antiproliferative activities. *Food Chem*. 2013; 141(1): 266-273. doi:10.1016/j.foodchem.2013.02.127.
37. Jung SH, Kim BJ, Lee EH, Osborne NN. Isoquercitrin is the most effective antioxidant in the plant Thuja orientalis and able to counteract oxidative-induced damage to a transformed cell line (RGC-5 cells). *Neurochem Int*. 2010; 57(7): 713-721. doi:10.1016/j.neuint.2010.08.005.
38. Sheikh S, Safia, Haque E, Mir SS. Neurodegenerative Diseases: Multifactorial Conformational Diseases and Their Therapeutic Interventions. *J Neurodegener Dis*. 2013; 2013: 1-8. doi:10.1155/2013/563481.
39. Uttara B, Singh A, Zamboni P, Mahajan R. Oxidative Stress and Neurodegenerative Diseases: A Review of Upstream and Downstream Antioxidant Therapeutic Options. *Curr Neuropharmacol*. 2009; 7(1): 65-74. doi:10.2174/157015909787602823.
40. Halliwell B, Rafter J, Jenner A. Health promotion by flavonoids , tocopherols , tocotrienols , and other phenols : direct or indirect effects ? Antioxidant or not ? 1 – 4. *Am J Clin Nutr*. 2005;81:268-276.
41. Martin S, Gonzalez-Burgos E, Carretero ME, Gomez-Serranillos MP. Neuroprotective properties of Spanish red wine and its isolated polyphenols on astrocytes. *Food Chem*. 2011; 128(1): 40-48. doi:10.1016/j.foodchem.2011.02.074.
42. Moniruzzaman M, Asaduzzaman M, Hossain MS, et al. In vitro antioxidant and cholinesterase inhibitory activities of methanolic fruit extract of Phyllanthus acidus. *BMC Complement Altern Med*. 2015;15:403. doi:10.1186/s12906-015-0930-y.
43. Boligon AA, Pereira RP, Feltrin AC, et al. Antioxidant activities of flavonol derivatives from the leaves and stem bark of Scutia buxifolia Reiss. *Bioresour Technol*. 2009; 100(24): 6592-6598. doi:10.1016/j.biortech.2009.03.091.
44. Boots AW, Drent M, de Boer VCJ, Bast A, Haenen GRMM. Quercetin reduces markers of oxidative stress and inflammation in sarcoidosis. *Clin Nutr*. 2011; 30(4): 506-512. doi:10.1016/j.clnu.2011.01.010.
45. Chung MJ, Lee S, Park Y Il, Lee J, Kwon KH. Neuroprotective effects of phytosterols and flavonoids from Cirsium setidens and Aster scaber in human brain neuroblastoma SK-N-SH cells. *Life Sci*. 2016. doi:10.1016/j.lfs.2016.02.035.
46. Kanter M, Unsal C, Aktas C, Erboga M. Neuroprotective effect of quercetin against oxidative damage and neuronal apoptosis caused by cadmium in hippocampus. *Toxicol Ind Health*. 2016; 32(3): 541-550. doi:10.1177/0748233713504810.
47. Jimenez-Aliaga K, Bermejo-Bescos P, Benedi J, Martin-Aragon S. Quercetin and rutin exhibit antiamyloidogenic and fibril-disaggregating effects in vitro and potent antioxidant activity in APPswe cells. *Life Sci*. 2011; 89(25-26): 939-945. doi:10.1016/j.lfs.2011.09.023.
48. McAnulty LS, Miller LE, Hosick PA, Utter AC, Quindry JC, McAnulty SR. Effect of resveratrol and quercetin supplementation on redox status and inflammation after exercise. *Appl Physiol Nutr Metab*. 2013; 38(7): 760-765. doi:10.1139/apnm-2012-0455.
49. Arora R, Chawla R, Sagar R, et al. Evaluation of radioprotective activities Rhodiola imbricata Edgew--a high altitude plant. *Mol Cell Biochem*. 2005; 273(1-2): 209-223.
50. Senol FS, Acikara OB, Citoglu GS, Orhan IE, Dall' Acqua S, Ozgokce F. Prospective neurobiological effects of the aerial and root extracts and some pure compounds of randomly selected Scorzonera species. *Pharm Biol*. 2014; 52(7): 873-882. doi:10.3109/13880209.2013.872152.
51. Dufour C, Loonis M. Flavonoids and their oxidation products protect efficiently albumin-bound linoleic acid in a model of plasma oxidation. *Biochim Biophys Acta - Gen Subj*. 2007; 1770(6): 958-965. doi:10.1016/j.bbagen.2007.02.005.
52. Dower JI, Geleijnse JM, Gijsbers L, Schalkwijk C, Kromhout D, Hollman PC. Supplementation of the Pure Flavonoids Epicatechin and Quercetin Affects Some Biomarkers of Endothelial Dysfunction and Inflammation in (Pre)Hypertensive Adults: A Randomized Double-Blind, Placebo-Controlled, Crossover Trial. *J Nutr*. 2015; 145(7): 1459-1463. doi:10.3945/jn.115.211888.
53. Swerdlow RH. Pathogenesis of Alzheimer's disease. *Clin Interv Aging*. 2007; 2(3): 347–359.
54. Spagnuolo C, Napolitano M, Tedesco I, Moccia S, Milito A, Russo GL. Neuroprotective role of natural polyphenols. *Curr Top Med Chem*. 2016.
55. Higgins GC, Beart PM, Shin YS, Chen MJ, Cheung NS, Nagley P. Oxidative stress: emerging mitochondrial and cellular themes and variations in neuronal injury. *J Alzheimers Dis*. 2010; 20 Suppl 2:S453-73. doi:10.3233/JAD-2010-100321.

56. Parsons CG, Danysz W, Dekundy A, Pulte I. Memantine and cholinesterase inhibitors: complementary mechanisms in the treatment of Alzheimer's disease. *Neurotox Res*. 2013; 24(3): 358-369. doi:10.1007/s12640-013-9398-z.
57. Chan P-C, Xia Q, Fu PP. Ginkgo biloba leave extract: biological, medicinal, and toxicological effects. *J Environ Sci Health C Environ Carcinog Ecotoxicol Rev*. 2007; 25(3): 211-244. doi:10.1080/10590500701569414.
58. Shi C, Zhao L, Zhu B, et al. Protective effects of Ginkgo biloba extract (EGb761) and its constituents quercetin and ginkgolide B against beta-amyloid peptide-induced toxicity in SH-SY5Y cells. *Chem Biol Interact*. 2009; 181(1): 115-123. doi:10.1016/j.cbi.2009.05.010.
59. Morris E, Chalkidou A, Hammers A, Peacock J, Summers J, Keevil S. Diagnostic accuracy of (18)F amyloid PET tracers for the diagnosis of Alzheimer's disease: a systematic review and meta-analysis. *Eur J Nucl Med Mol Imaging*. 2016; 43(2): 374-385. doi:10.1007/s00259-015-3228-x.
60. Ansari MA, Abdul HM, Joshi G, Opii WO, Butterfield DA. Protective effect of quercetin in primary neurons against Abeta(1-42): relevance to Alzheimer's disease. *J Nutr Biochem*. 2009; 20(4): 269-275. doi:10.1016/j.jnutbio.2008.03.002.
61. Choi S-M, Kim BC, Cho Y-H, et al. Effects of Flavonoid Compounds on beta-amyloid-peptide-induced Neuronal Death in Cultured Mouse Cortical Neurons. *Chonnam Med J*. 2014; 50(2): 45-51. doi:10.4068/cmj.2014.50.2.45.
62. Liu R, Zhang T, Zhou D, et al. Quercetin protects against the Abeta(25-35)-induced amnesic injury: involvement of inactivation of rage-mediated pathway and conservation of the NVU. *Neuropharmacology*. 2013; 67: 419-431. doi:10.1016/j.neuropharm.2012.11.018.
63. Li Y, Zhou S, Li J, et al. Quercetin protects human brain microvascular endothelial cells from fibrillar beta-amyloid1-40-induced toxicity. *Acta Pharm Sin B*. 2015;5(1):47-54. doi:10.1016/j.apsb.2014.12.003.
64. Hayakawa M, Itoh M, Ohta K, et al. Quercetin reduces eIF2α phosphorylation by GADD34 induction. *Neurobiol Aging*. 2015; 36(9): 2509-2518. doi:10.1016/j.neurobiolaging.2015.05.006.
65. Mohebali N, Shahzadeh Fazeli SA, Ghafoori H, et al. Effect of flavonoids rich extract of Capparis spinosa on inflammatory involved genes in amyloid-beta peptide injected rat model of Alzheimer's disease. *Nutr Neurosci*. 2016: 1-8. doi:10.1080/1028415X.2016.1238026.
66. Sabogal-Guáqueta AM, Muñoz-Manco JI, Ramírez-Pineda JR, Lamprea-Rodriguez M, Osorio E, Cardona-Gómez GP. The flavonoid quercetin ameliorates Alzheimer's disease pathology and protects cognitive and emotional function in aged triple transgenic Alzheimer's disease model mice. *Neuropharmacology*. 2015; 93: 134-145. doi:10.1016/j.neuropharm.2015.01.027.
67. Javed H, Khan MM, Ahmad A, et al. Rutin prevents cognitive impairments by ameliorating oxidative stress and neuroinflammation in rat model of sporadic dementia of Alzheimer type. *Neuroscience*. 2012; 210: 340-352. doi:10.1016/j.neuroscience.2012.02.046.
68. Choi JY, Lee JM, Lee DG, et al. The n-Butanol Fraction and Rutin from Tartary Buckwheat Improve Cognition and Memory in an In Vivo Model of Amyloid-beta-Induced Alzheimer's Disease. *J Med Food*. 2015; 18(6): 631-641. doi:10.1089/jmf.2014.3292.
69. Xu P-X, Wang S-W, Yu X-L, et al. Rutin improves spatial memory in Alzheimer's disease transgenic mice by reducing Abeta oligomer level and attenuating oxidative stress and neuroinflammation. *Behav Brain Res*. 2014; 264: 173-180. doi:10.1016/j.bbr.2014.02.002.
70. Moghbelinejad S, Nassiri-Asl M, Farivar TN, et al. Rutin activates the MAPK pathway and BDNF gene expression on beta-amyloid induced neurotoxicity in rats. *Toxicol Lett*. 2014; 224(1): 108-113. doi:10.1016/j.toxlet.2013.10.010.
71. Colovic MB, Krstic DZ, Lazarevic-Pasti TD, Bondzic AM, Vasic VM. Acetylcholinesterase Inhibitors: Pharmacology and Toxicology. *Curr Neuropharmacol*. 2013; 11(3): 315-335. doi:10.2174/1570159X11311030006.
72. Abdalla FH, Schmatz R, Cardoso AM, et al. Quercetin protects the impairment of memory and anxiogenic-like behavior in rats exposed to cadmium: Possible involvement of the acetylcholinesterase and Na(+), K(+)-ATPase activities. *Physiol Behav*. 2014; 135: 152-167. doi:10.1016/j.physbeh.2014.06.008.
73. Baldissarelli J, Santi A, Schmatz R, et al. Hypothyroidism Enhanced Ectonucleotidases and Acetylcholinesterase Activities in Rat Synaptosomes can be Prevented by the Naturally Occurring Polyphenol Quercetin. *Cell Mol Neurobiol*. 2016. doi:10.1007/s10571-016-0342-7.
74. Islam MR, Zaman A, Jahan I, Chakravorty R, Chakraborty S. In silico QSAR analysis of quercetin reveals its potential as therapeutic drug for Alzheimer's disease. *J Young Pharm*. 2013; 5(4): 173-179. doi:10.1016/j.jyp.2013.11.005.
75. Kamal Z, Ullah F, Ayaz M, et al. Anticholinesterase and antioxidant investigations of crude extracts, subsequent fractions, saponins and flavonoids of atriplex laciniata L.: potential effectiveness in Alzheimer's and other neurological disorders. *Biol Res*. 2015; 48: 21. doi:10.1186/s40659-015-0011-1.

76. Vellosa JCR, Regasini LO, Khalil NM, et al. Antioxidant and cytotoxic studies for kaempferol, quercetin and isoquercitrin. *Eclet Quim*. 2011; 36(2): 7-20. doi:10.1590/S0100-46702011000200001.
77. Orfali G di C, Duarte AC, Bonadio V, et al. Review of anticancer mechanisms of isoquercitin. *World J Clin Oncol*. 2016;7(2):189. doi:10.5306/wjco.v7.i2.189.
78. Ahn T-B, Jeon BS. The role of quercetin on the survival of neuron-like PC12 cells and the expression of alpha-synuclein. *Neural Regen Res*. 2015; 10(7): 1113-1119. doi:10.4103/1673-5374.16 0106.
79. Zhu M, Han S, Fink AL. Oxidized quercetin inhibits α-synuclein fibrillization. *Biochim Biophys Acta*. 2013; 1830(4): 2872-2881. doi:10.1016/j.bbagen.2012.12.027.
80. Karuppagounder SS, Madathil SK, Pandey M, Haobam R, Rajamma U, Mohanakumar KP. Quercetin up-regulates mitochondrial complex-I activity to protect against programmed cell death in rotenone model of Parkinson's disease in rats. *Neuroscience*. 2013; 236: 136-148. doi:10.1016/j.neuroscience.2013.01.032.
81. Denny Joseph KM, Muralidhara. Combined oral supplementation of fish oil and quercetin enhances neuroprotection in a chronic rotenone rat model: relevance to Parkinson's disease. *Neurochem Res*. 2015; 40(5): 894-905. doi:10.1007/s11064-015-1542-0.
82. Bournival J, Quessy P, Martinoli M-G. Protective effects of resveratrol and quercetin against MPP+ -induced oxidative stress act by modulating markers of apoptotic death in dopaminergic neurons. *Cell Mol Neurobiol*. 2009; 29(8): 1169-1180. doi:10.1007/s10571-009-9411-5.
83. Zhang ZJ, Cheang LCV, Wang MW, Lee SM-Y. Quercetin exerts a neuroprotective effect through inhibition of the iNOS/NO system and pro-inflammation gene expression in PC12 cells and in zebrafish. *Int J Mol Med*. 2011; 27(2): 195-203. doi:10.3892/ijmm.2010.571.
84. Bournival J, Plouffe M, Renaud J, Provencher C, Martinoli M-G. Quercetin and sesamin protect dopaminergic cells from MPP+-induced neuroinflammation in a microglial (N9)-neuronal (PC12) coculture system. *Oxid Med Cell Longev*. 2012; 2012: 921941. doi:10.1155/2012/921941.
85. Haleagrahara N, Siew CJ, Ponnusamy K. Effect of quercetin and desferrioxamine on 6-hydroxydopamine (6-OHDA) induced neurotoxicity in striatum of rats. *J Toxicol Sci*. 2013; 38(1): 25-33.
86. Magalingam KB, Radhakrishnan A, Haleagrahara N. Protective effects of flavonol isoquercitrin, against 6-hydroxy dopamine (6-OHDA)-induced toxicity in PC12 cells. *BMC Res Notes*. 2014; 7: 49. doi:10.1186/1756-0500-7-49.
87. Magalingam KB, Radhakrishnan A, Ramdas P, Haleagrahara N. Quercetin glycosides induced neuroprotection by changes in the gene expression in a cellular model of Parkinson's disease. *J Mol Neurosci*. 2015; 55(3): 609-617. doi:10.1007/s12031-014-0400-x.
88. Magalingam KB, Radhakrishnan A, Haleagrahara N. Protective effects of quercetin glycosides, rutin, and isoquercetrin against 6- hydroxydopamine (6-OHDA)- induced neurotoxicity in rat pheochromocytoma (PC-12) cells. *Int J Immunopathol Pharmacol*. 2016; 29(1): 30-39. doi:10.1177/0394632015613039.
89. Ross CA, Tabrizi SJ. Huntington's disease: from molecular pathogenesis to clinical treatment. *Lancet Neurol*. 2011; 10(1): 83-98. doi:10.1016/S1474-4422(10)70245-3.
90. Novak MJU, Tabrizi SJ. Huntington's disease. *BMJ*. 2010; 340 (jun30 4): c3109-c3109. doi:10.1136/bmj.c3109.
91. Bates GP, Dorsey R, Gusella JF, et al. Huntington disease. *Nat Rev Dis Prim*. 2015: 15005. doi:10.1038/nrdp.2015.5.
92. Sandhir R, Mehrotra A. Quercetin supplementation is effective in improving mitochondrial dysfunctions induced by 3-nitropropionic acid: implications in Huntington's disease. *Biochim Biophys Acta*. 2013; 1832(3): 421-430. doi: 10.1016/ j.bbadis.2012.11.01 8.
93. Jain D, Gangshettiwar A. Combination of lycopene, quercetin and poloxamer 188 alleviates anxiety and depression in 3-nitropropionic acid-induced Huntington's disease in rats. *J Intercult Ethnopharmacol*. 2014; 3(4): 186-191. doi:10.5455/jice.20140903012921.
94. Chakraborty J, Singh R, Dutta D, Naskar A, Rajamma U, Mohanakumar KP. Quercetin improves behavioral deficiencies, restores astrocytes and microglia, and reduces serotonin metabolism in 3-nitropropionic acid-induced rat model of Huntington's Disease. *CNS Neurosci Ther*. 2014; 20(1): 10-19. doi:10.1111/cns.12189.