Chapter

Physiological and Cellular Targets of Neurotrophic Anxiolytic Phytochemicals in Food and Dietary Supplements

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Abstract

Diet impacts anxiety in two main ways. First anxiety can be caused by deficiencies in antioxidants, neurotransmitter precursors, amino acids, cations and vitamins and other cofactors. Second, anxiety can be reduced by anxiolytic nutraceuticals which are food molecules that bind to molecular targets of the amygdala and the hypothalamus-pituitary–adrenal axis (HPA-axis). Anxiety is a feeling of fear that arises from a perceived threat and can be a beneficial coping mechanism to threats and stressors. However excessive anxiety is a disorder that interferes with healthy responses to stressors. The amygdala is responsible for assigning value to a threat or stressor and triggering the HPA-axis to support the body wide system responses to the threat. The amygdala also communicates with the neuroplastic learning and memory centers of the hippocampus to fix or set a learned value to the threat. Interestingly, many anxiolytic nutraceuticals that show benefits in human clinical trials have neurotrophic activity and increase neuronal plasticity. Moreover, anxiolytic nutraceuticals either act like the neurotrophins, nerve growth factor (NGF), brain derived neurotrophic factor (BDNF and neurotrophin-3 (NT3) by either directly binding to or potentiating the tyrosine receptor kinase (TRK) family of receptors (TRKA, TRKB and TRKC) and activating the ERK1/2 signal transduction pathway associated with neurite outgrowth and neural plasticity. This chapter will explore the neuritogenic activity of clinically proven plant-based anxiolytic nutraceuticals and examine the commonality of TRKA-C receptors and the ERK1/2 signaling pathway in the pharmacological and nutraceutical treatment of anxiety disorders.

Keywords: Anxiety, Anxiolytic, Nutraceutical, Cannabidiol, Neurotrophin, Neurite Outgrowth, NGF, BDNF, NT3, TRKA, TRKB, TRKC, ERK1/2

1. Introduction

In humans, the appropriate and measured behavioral responses to environmental cues are under control of the limbic nervous system which is composed primarily of the amygdala, hippocampus, thalamus, and hypothalamus [1]. In order for

sensory inputs to the cerebral cortex to result in the appropriate responses in the body, sensory inputs relay from the cerebrum, to the limbic system and then from the limbic system to the body either through the brainstem or through the pituitary gland. It is when relaying sensory inputs from the cerebral cortex to the body that the limbic system also assigns emotional value to sensory input and sets or fixes that value by learning and remembering the rewards and punishments associated with specific environmental cues. The amygdala is known for assigning a scaled value to negative threats and stressors which the amygdala then communicates to learning and memory centers in the hippocampus so that human behavioral responses to negative cues can be consistent and appropriate. The amygdala also stimulates the hypothalamus to secrete corticotrophin-releasing hormone (CRH) which in turn stimulates the pituitary to release adrenocorticotropin hormone (ACTH), which in turn stimulates the adrenal cortex to secrete glucocorticoids including primarily, cortisol in what is known as the HPA-axis [2–4]. The hypothalamus can also send signals through the brainstem and activate the adrenal medulla to secrete epinephrine and norepinephrine. Cortisol, epinephrine and norepinephrine are hormones that can signal body wide changes in metabolic rates, breathing, heart rate, blood pressure and a variety of other appropriate body responses to the presence of an environmental threat or stressor [2, 3]. Anxiety is the feeling of fear or worry that arises from the neurochemistry of the amygdala in response to negative environmental cues and the activation of the HPA-axis and the overall preparation of the body to meet the challenges of a threat or stressor and while anxiety is a negative feeling, when it is in proportion to the actual threat a stressor presents, anxiety can be a normal and even healthy part of an adequate response to the stressor [5–7]. However, excessive and prolonged anxiety that is unwarranted by the environmental cue and exaggerated in proportion to the actual threat level leads to inappropriate and prolonged activation of the HPA-axis and cortisol release which is associated with inflammatory damage and other pathophysiologies that further stresses the human body system [2–4]. In these cases anxiety interferes with normal and health everyday life and is considered an anxiety disorder [8, 9].

People suffer from five different types of anxiety disorders; generalized anxiety disorder (GAD, obsessive compulsive disorder (OCD), panic disorder (PD), social anxiety disorder (SAD), and posttraumatic stress disorder (PTSD) [8, 9]. Each of these anxiety disorders can be described by the level of synaptic neurotransmitters and cell surface neurotransmitter receptors in the amygdala [1, 8, 9]. For example, GAD is associated with decreased activity of the inhibitory neurotransmitter, GABA. GABA acts on $GABA_A$ receptors on neurons within the amygdala to inhibit signals and help to assign lower threat values to certain stressor. Down regulation of the $GABA_A$ receptor and the subsequent reduction of GABA signaling in the amygdala leads GAD through elevated valuation of threats [10]. Similarly, PD is also associated with decreased GABAnergic transmission and subsequent over stimulation of neural pathways, however in PD the decrease GABAnergic signaling may be due to reduced level of the GABA neurotransmitter itself and not due to decreased $GABA_A$ receptors as seen in GAD [9, 11, 12]. While GABAnergic pathways in the amygdala are inhibitory and stress reducing, glutamate, the major stimulatory neurotransmitter, when over active in the amygdala enhances stress and can lead to OCD. Pharmacological enhancement of glutaminergic signals in the frontolimbic regions of the brain enhance anxiety and imaging studies have shown increased glutaminergic activity in various structures of the limbic system in the brain [13–15]. PTSD and SAD also appear involve increased glutaminergic activity in the amygdala [9, 16]. GABA and glutamate influence the feeling of anxiety be reducing and enhancing the perceived threats, while the neurotransmitters, serotonin and dopamine are associated with the reward and pleasure pathways of the limbic

system and can influence the overall perception of environmental stressors generally reducing anxiety. For example, SAD is associated with both decreased activity at serotonin receptors and also decreased dopamine levels in limbic neurocircuitry [9, 16, 17]. Taken together, anxiety disorders involve irregularities in the levels of neurotransmitters and neurotransmitter receptors in the neurocircuitry of the limbic system. The inappropriate levels of neurotransmitters and their receptors can lead to hyper activity in regions of the limbic system such as the amygdale and lead to incorrect and unhealthy assessment of the risks and threats associated with stressors or lack of stressors and lead to anxiety and fear potentially even in the absence of threat. Activation of the HPA-axis can contribute to both the clinical signs and symptoms of anxiety and also lead to chronic glucocorticoid induced pathologies which serve and further internal stressors and add to anxiety. Treatments for anxiety disorders have therefore focused on developing drugs that correct and manage the levels of neurotransmitters and neurotransmitters receptors and signaling in the limbic system pathways and particularly in the amygdala.

GABAnergic benzodiazepines are the favored class of anxiolytic medications [10, 11, 18]. The diazepine ring is a seven membered ring structure containing two nitrogens and this diazepine ring and when fuses with a benzene ring forms a benzodiazepine that can bind to GABAA receptors on neurons in the brain [18]. Benzodiazepines are favored due to their lesser side-effects compared to other anxiolytic drugs, although side effects are still concerns [18]. The mechanism of benzodiazepine signaling is binding to either $GABA_A$ or $GABA_B$ receptors and allowing either chlorine ions into the cell at the synapse or stimulating the release of potassium from the cell into the synapse respectively [10, 11, 18]. In the cells of the amygdala, the chlorine influx inhibits the signaling of the pathway and diminishes the level of potential threat assigned to a sensory input or any external or internal stressor. People with GAD and PD express low levels of $GABA_A$ and produce less GABA respectively thereby limiting the patient's ability diminish the signals from stressors is associate with a heightened sense of fear and worry. By being GABAnergic the benzodiazepines help to restore or boost the GABAnergic pathway and the therefore the reduction of anxiety. Alternatively to drugs that act in a GABAnergic fashion, serotonin and dopamine uptake inhibitors, often used for depression, reduce anxiety and fear by increasing levels of these "feel good" neurotransmitters in the limbic neurocircuitry. Low synaptic serotonin and dopamine in the amygdale and nucleus accumbens is associated SAD. Serotonin uptake inhibitors (SSRIs) and noradrenalin and dopamine reuptake inhibitors (NDRIs) increased the level of serotonin and dopamine in the synapse and have been used to treat depression and also provide relief from anxiety and anxiety disorders. [18–22].

In addition to the development of new drugs that interact with the amygdala and HPA-axis, anxiety can also be addressed by diet. The diet can be associated with anxiety in two main ways. First, if a diet is deficient in nutrients such as selenium, lysine, magnesium and inositol, changes in food consumptions or dietary supplementation can replace the deficient nutrient, balance the diet and alleviate anxiety [23]. Further, dietary deficiencies in antioxidants can lead to the buildup of reactive oxygen species (ROSs) that form as a part of normal metabolism and are reactive chemicals that can bind to DNA, lipids and proteins leading to DNA and membrane damage and cellular toxicity. This cellular damage serves as a stress signal and is associated with anxiety [24, 25]. Therefore, increasing dietary antioxidant intake can help with anxiety. Second, food nutrients can directly affect the neurochemistry of the limbic system by either directly boosting GABA or Serotonin levels or by binding to neurotransmitter receptors. For example, GABA is an amino acid is available directly in the diet. Further the amino acid,

5-hydroxytryptophan is a serotonin precursor and is a popular dietary supplement taken to easy feelings of anxiety and stress. While it is not clear if increasing oral consumption of GABA and 5-hydroxytrptophan can increase brain GABA and serotonin levels, clinical studies have shown and relaxing effect of GABA and 5-HTP supplementation [23]. The neurochemistry of the brain can also be altered by food chemicals eaten from bacteria, fungi and plants that have nutraceutical effects by acting in a drug-like fashion as cell signaling molecules and alerting cellular behavior. In this chapter we focus on food nutraceuticals that are anxiolytic in humans and alter the neurochemistry and the amygdala and other limbic structures in the brain. Of particular interest are anxiolytic phytochemicals that in addition to changing the brain neurotransmitter physiology also stimulate neuronal plasticity through the activation and or potentiating of neurotrophin receptors and signal transduction pathways.

Recent studies have revealed that numerous anxiolytic substances, including endogenous neurotransmitters, anxiolytic drugs, and nutraceuticals, are also neurotrophic in that they also activate the brain derived neurotrophic factor (BDNF) pathway, the neurotrophin-3 (NT-3) pathways and the nerve growth factor (NGF) pathway by binding to or potentiating the TRKA – C neurotrophin receptors and directly activating the ERK1/2 signaling pathway leading to neuroplasticity [26–37]. This is important because neurotrophins can regulate neuroplasticity not only during development but also during learning and the establishment of memories [35–37]. Neurotrophins are small soluble signaling molecules that can diffuse between cells to play a role in cell–cell communication [35–37]. These neurotrophic factors include BDNF, NGF and NT3 bind to cell surface molecules on neuronal cells known as the tropomyosin receptor kinases (TRK) A – C respectively [35–37]. Neurotrophin signaling is associated with neuritogenesis or new neurite formation in neuronal cells. The changes in cell shape associated with the establishment of new neurites and therefore potentially new connections is known as neuroplasticity [35–37]. Recent attention has been brought to the idea that in so far as anxiety is related to the memories of trauma and the establishment of a learned threat level in the perception of stressors through neuroplasticity, perhaps anxiolytic phytochemicals with neurotrophic activity can be used to reduce anxiety not only through changes neurotransmitter activity, but also by providing the plasticity required to relearn and reduce the emotional value ascribed to a stressor thereby also facilitating the reduction in anxiety [26–34]. Therefore anxiolytic phytochemical neurotrophins are important because they offer a new area of research into not simply adjusting neurotransmitter activity, but to the development of natural treatments and drugs that can actually reverse the neurocircuitry associated with anxiety through neuroplasticity and relearning. It is important to note however, not all anxiolytic phytochemicals are capable of stimulating neuroplasticity. The following section of this chapter will present all nutraceutical phytochemicals that are anxiolytic in human clinical trials that also show potential for stimulating neuroplasticity either by directly stimulating neuritogenesis or neurite outgrowth neuronal cells or by binding to the TRKA-C neurotrophin receptors and or by the activation of the neurotrophin ERK1/2 signal transduction pathway and others associated with neurite formation.

2. Clinically relevant anxiolytic phytochemicals with neurotrophic activity

In this chapter we present only plants and plant extracts that contain phytochemicals that are both shown to be anxiolytic in human clinical trials and also

possess neuroplastic properties (**Table 1**). The specific anxiolytic nutraceutical or phytochemical in the plant is in most cases not known, in part because neuroactive plants usually contain many nervine agents. Often however there is a suspected phytochemical or group of phytochemicals thought to be responsible for the anxiolytic activity. In some cases the anxiolytic nutraceutical in the plant extract is the same phytochemical that has the neurotrophic activity, while in other cases it may be a different phytochemical in the plant extract. Anxiolytic drugs adjust neurotransmitter and neurotransmitter receptors levels which leads to increased drug insensitivity, extreme withdrawal effects and a return to imbalance neurotransmitter and neurotransmitter receptor levels when and if the drug is removed. In addition to altering neurotransmitter and receptor levels neuroplastic anxiolytics also stimulate the new neurite connections associated with learning and remembering appropriate responses to stressors. If a new response to a threat is learned, then treatment of the anxiety disorder may not require dosage increases and the newly learned healthy perceptions of threats could remain with the patient even if the drug or treatment is removed or reduced. This would represent a tremendous advancement in the treatment of anxiety disorders. **Table 1** is a list of the fourteen clinically supported anxiolytic plants that also have neuroplastic properties.

2.1 Theanine

Theanine is an amino acid that when taken as a green tea extract or in a purified form is able to reduce anxiety in clinical trials [38–40]. When administered in a double blind placebo controlled study, theanine was shown to reduce stress-induced salivary cortisol levels [39]. However in other studies, while theanine did improve the sleep in people with GAD, theanine did not reduced anxiety scores on the HAMA scale [41]. Both animal and in vitro studies have suggested that theanine supplementation increases brain serotonin, dopamine and GABA levels and that the cellular target for theanine includes glutamate receptors to which theanine binds and antagonizes the stimulating action of glutamate on neurons [42–44]. With regard to neuroplasticity, theanine facilitates neuritogenesis in the developing rat hippocampus and enhances object learning memory [45]. Further, dietary theanine increases nerve growth factor (NGF) levels in the developing rat brain [46]. Theanine is not the only green tea molecule that can affect neurotrophin activity. The catechins from green tea have been show to potentiate BDNF binding to TRKB receptors in PC12 cells and enhance neurite outgrowth [47], and potentiate NGF signaling through TRKA receptors and enhance neurite outgrowth also in PC12 cells [48]. Further, the green tea catechin, green tea polyphenol (−)-epigallocatechin-3-gallate (EGCG) also stimulates neurite outgrowth in cultured PC12 neurons [49].

2.2 Chamomile

In clinical trials, chamomile has been shown to decrease the symptoms of general anxiety disorder [50, 51], in part by exerting an effect on diurnal cortisol changes [52]. While not yet known, apigenin is a plant flavone component of chamomile which is thought to contribute to the anxiolytic effects of chamomile [53]. Interestingly, apigenin increases neurite formation in murine N2a cells [54] and reverses PTZ induced behavioral impairments in mice by increasing hippocampal levels of brain derived neurotrophic factor (BDNF) [55]. Apigenin also has been shown to increase hippocampal BDNF levels in a chronic corticosteroid treatment model of depression in mice [56]. Apigenin also activates the ERK1/2 pathway in PC12 cells and while not sufficient to stimulate differentiation in PC12 cells [57], apigenin does increases neurite outgrowth in estrogen receptor

Anxiolytic	Neurotrophic Activities.		
Plant/Nutraceutical	**Neurotrophin Pathways**	**Neurite Outgrowth**	**References**
Green Tea/theanine	Increases brain BDNF levels	hippocampal cells	[45, 46]
	Increases brain NGF synthesis	Neural stem cells	
	Potentiates NGF at TRKA		
Chamomile/apigenin	Increases hippocampal BDNF	N2a cells	[55–58]
	ERK1/2 kinase activation		
Lavender/N.D.	Increases brain NGFR	neuronal cells	[67, 68]
Ashwagandha/N.D.	Increases brain GDNF	hippocampal cells	[75]
Passion Flower/ apigenin	Potentiates NGF	PC12 cells	[80]
Cannabis/CBD	Binds TRKA	PC12 cells	[88]
	Activates ERK1/2		
Valerian/ Sesquiterpenes	Increases BDNF secretion	PC12 cells	[102, 103]
	NGF potentiation		
Citrus/limolene	ERK1/2 activation	PC12 cells	[110–113]
Saffron/N.D.	Increases BDNF and GDNF	N.D.	[116, 117]
Bacopa Monieri/ saponins	Increases brain NGF and	N.D.	[120, 121]
	BDNF levels		
Skullcap/baicalin	Increases brain BDNF	N.D.	[126, 127]
	ERK1/2 activation		
Rhodiola rosea/ salidroside	Increases NT-3, BDNF	stem cells	[130, 131]
	and NGF; ERK1/2 activation		
Hops/prenylflavinoids	TRKA signaling	PC12 cells	[135, 136]
		dorsal root ganglia	
Nigella sativa/ thymoquinone	N.D.	hippocampal cells	[141, 142]
		dorsal root ganglia	

All plants listed above have been shown to be anxiolytic in human clinical trials. In some cases the anxiolytic molecules and neurotrophic activities have not been determined (N.D.). Neurotrophic activities are those associated with activating neurotrophin signaling pathways by increasing levels of neurotrophin (NGF, BDNF, NT3, GDBF) synthesis, or by directly binding to neurotrophin receptors (TRKA, TRKB, TRKC and NGFR) or by activating the ERK1/2 signaling pathway. Another neurotrophic activity is the induction of neurite outgrowth or neuritogenesis in neuronal cell cultures and in. In these cases the names of the cells or tissues showing a neuroplastic response is provided.

Table 1.
The neurotrophic activities of anxiolytic plant extracts and phytochemical nutraceuticals.

expressing PC12 cells [58] again linking dietary phytochemicals that are anxiolytic to neural plasticity.

2.3 Lavender

Lavender oil also has anxiolytic effects in clinical trials in which it can both reduce anxiety associated with stressful event such as surgeries and recovery and also reducing anxiety in anxiety disorders [49–63]. Targets for lavender oil include the 5-HT1A serotonin receptor, the NMDA receptor and the serotonin transporter (SERT) [64, 65]. Linalool, a lavender oil terpene in specific can bind to SERT [64]. In a clinical trial where subjects were subjected to stress, linalool helped to reduce stress as measured by salivary cortisol levels, blood pressure and heart rate [66]. While linalool may be responsible for much of the anxiolytic effects of lavender oil, linalool has not been shown to have neurotrophic activity, however, lavender oil has be shown to increase neurite outgrowth and synapse formation in neuronal cell cultures [67] and increase both BDNF and nerve growth factor receptor (NGFR) levels in mouse brain [68]. Activation of NGFR is associated with enhanced TRKA receptor activity in neurons which triggers neurite outgrowth in response to NGF signaling [69, 70].

2.4 Ashwagandha

Ashwagandha is a plant used in Ayeurvic medicine from which the roots and berries have been used as adaptagens and also to relieve stress. In double blind placebo controlled clinical trials Ashwagandha supplementation has been shown to reduce anxiety based reducing both scores on the Hamiltion-Anxiety (HAMA) scale and morning salivary cortisol [71] and reduce anxiety in a variety of other contexts including schizophrenia and sleep disorders [71–73]. With regard to brain neurochemistry, Ashwagandha does not appear to affect sertotonergic, GABAnergic, or glutaminergic pathways but instead increases cholinergic signaling in the cortical and basal forebrain [74]. While the specific bioactive molecule(s) in Ashwagandha that are anxiolytic have not been specifically identified, sominone, an aglycone derivative of Withanoside IV when injected into mice stimulated neurite outgrowth in the hippocampus and increased production of the neurotrophin, Glial Derived neurotrophic Factor (GDNF) [75]. Further injection of sominone into mice enhances spatial memory, again suggesting that anxioloytic phytochemicals that are neurotrophic may ease anxiety by providing signals to enhance neural plasticity and learning [75].

2.5 Passion flower

Passion flower also shows anxiolytic properties in clinical trials that are as effective as midazolam and oxezepam [76, 77] and can reduce anxiety associated with ambulatory surgery and dental extraction [77–79]. The anxiolytic molecule from passion flower has not been identified and the effects of passion flower on brain neurochemistry is not well studied. It is interesting to note however that C-dideoxyhexosyl flavones from passion flower have been shown to enhance NGF-induced neurite outgrowth in PC12 cells [80].

2.6 Cannabidiol

Cannabidiol (CBD) is anxiolytic and has been shown in clinical trials to reduce social stress [81] and reduces anxiety in social phobia patients [82]. CBD

also reduces anxiety associated with drug-craving during recovery from heroin addiction [83]. With regard to brain neurochemistry in clinical trials, CBD reduction in SAD was associated with increased blood flow in the limbic and paralimbic brain areas [84]. CBD is anxiolytic through direct binding of the $GABA_A$ receptor and activating the GABAnergic pathway [85–87]. CBD also bind to the NGF receptor, TRKA which signals the ERK1/2 signal transduction pathway and stimulates neurite outgrowth in PC12 cells [88]. Indeed the mechanism of action of CBD is recognized to help with the neuronal plasticity through autophagy and neuritogenesis and may help not only with anxiety, but also with other psychiatric disorders [88, 89]. Due to the lipophilicity of CBD, there is interest in developing emulsification techniques to increase CBD bioavailability when taken in the diet. For example, nanoemulsification [90] and lipid extractions [91] and lipid-vehicles [92] and piperine nanoliposhperes of CBD [93, 94] have been investigated for better oral absorption and better bioavailability for cellular targeting. Nanoemulsified, versus lipid emulsified CBD were tested for their ability to stimulate neurite outgrowth in PC12 cells (**Figure 1**). Continuous lipid extracted CBD shows greater bioavailability and activity compared to nanoemulsification and piperine nanoliposheperes (**Figure 1**).

2.7 Valerian root

The anxiolytic activity seen in patients supplementing with Valerian root extract [95], is known to be due to the sesquiterpene, valerenic acid [96]. While there is evidence to suggest that valerenic acid activates the GABAnergic pathway [97, 98],

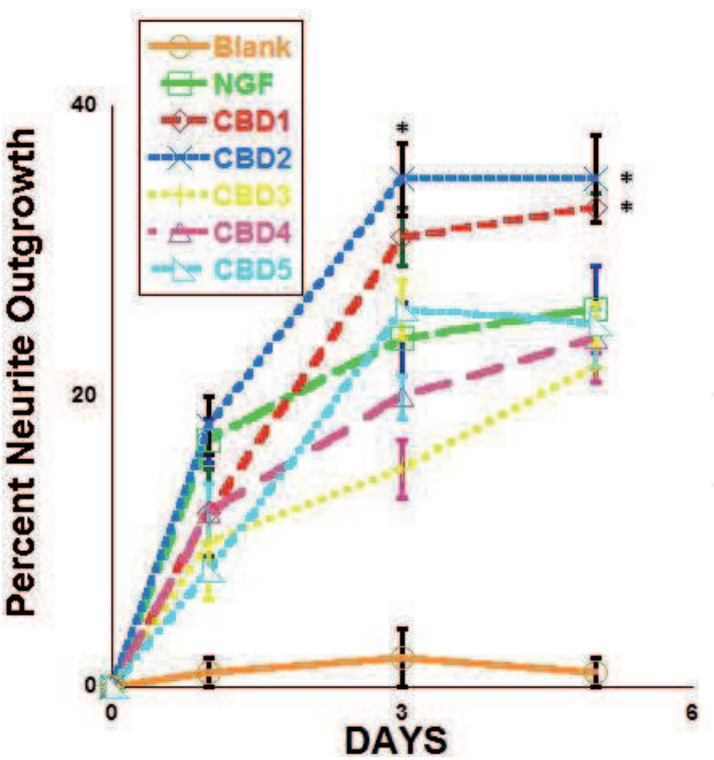

Figure 1.
The effects of CBD on PC12 cell neurite outgrowth. PC12 cells were seeded on tissue culture plastic in a serum free defined medium and the percentage of cells that formed neurites were counted by visual inspection over a five day period. Cells were either untreated (blank) or treated with 100 ng/ml nerve growth factor (NGF) or with 10 uM of five different CBD formulations (CBD1-CBD5). CBD designations are as follows: CBD isolated by continuous lipid extraction, (CBD1), CBD1 + vitamin C (CBD2), nano-emulsion CBD (CBD3), liposomal-emulsion CBD (CBD 4) and piperine nanoliposhpere preparation CBD (CBD5). CBD1 and CBD2 were statistically significantly more neuritogenic at 95% confidence (*) on days three and five ($p < 0.05$, t-test) compared to any of the other CBD formulations.

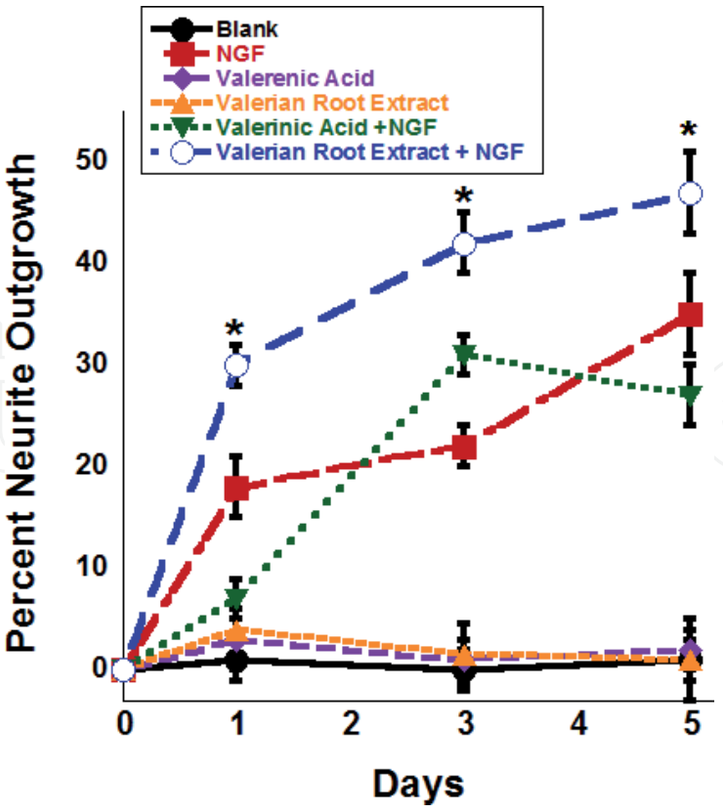

Figure 2.
The effect of valerian root extract and valerenic acid on neurite outgrowth in PC12 cell cultures. PC12 cells were seeded in serum free defined medium and treated as indicated with 10 ng/ml NGF, 100 μM valerenic acid and 50 mg/ml of 4:1 aqueous valerian root extract. The percentage of cells that formed neurites was counted by visual inspection over a five day period. More neurites were seen in PC12 cells treated with the valerian root extract and NGF when compared to NGF alone and these differences were statistically significantly at 95% confidence (*) on days one, three and five ($p < 0.05$, t-test). These data suggest valerian root extract phytochemical can potentiate NGF activity.

growing evidence suggests that valerenic acid mediates anxiolytic effects also by both antagonizing glutaminegric pathways [99, 100] and agonizing the serotonin receptor [101]. Valerenic acid also activates secretion of BDNF in cultured SH-SY5Y neurons [102]. Interestingly, germacrane, another sesquiterpene extracted from Valerian root, while not associated with an anxiolytic activity, has been shown to potentiate NGF and TRKA signaling and neurite outgrowth in PC12 cells [103]. An aqueous extract of valerian root enhances NGF-mediated neurite outgrowth and neuroplasticity but unlike CBD, the valerian extract is not neurotrophic in PC12 cells in the absence of NGF stimulation (**Figure 2**).

2.8 Citrus

Citrus plant extracts, including those from lemon, bitter orange, and bergamot relieve anxiety in clinical trials. For example, lemon inhalation reduced anxiety in myocardial infarction patients [104] and bergamot aromatherapy reduced preoperative anxiety [105] and bitter orange aroma therapy relieves anxiety in patients with acute coronary syndrome [106] and chronic myeloid leukemia [107] and preoperative anxiety [108]. Bitter orange extract contains primarily limolene and b-myrcrene, appear to act on the 5-HT serotonergic pathway [109]. When tested in PC12 cells, citrus phytochemicals such as nobilitin, gardenin A and auraptene all stimulate neurite outgrowth [110–112] and 5-Hydroxy-3,6,7,8,3′,4′-hexamethoxyflavone from sweet orange peel stimulates neurite outgrowth in and NGF-like fashion activating the ERK1/2 signaling pathway suggesting binding to TRKA [113].

2.9 Saffron

Saffron has been shown to be anxiolytic in two double blind placebo controlled clinical trials. [114, 115] and while the active anxiolytic molecule in Saffron has not been identified, crocin, a carotinoid in Saffron, has been shown to increase BDNF and GDNF expression in neuronal stem cells [116] and also increase hippocampal BDNF and protect the murine brain from methamphetamine toxicity [117].

2.10 *Bacopa monnieri*

In double blind placebo controlled trials, *Bacopa monnieri*, an adaptagen of Ayruvic medical tradition, has been show to enhance cognition and reduce anxiety [118, 119]. This anxiolytic adaptagen has been shown to increase nerve NGF expression in rats [120]. A saponin isolated from *Bacopa monnieri*, Bacopacide I, has been shown to have antidepressant activity in mice by modulating the HPA axis and enhancing BDNF mRNA expression in the hippocampus and prefrontal cortex of mice [121].

2.11 Skullcap

Skuttleria is a genus of plants known as the skullcaps that include scutellaria Radix and *Scutellaria lateriflora* (American skullcap). American skullcap has been shown to be anxiolytic in humans as shown by a reduction in anxiety in healthy volunteers using the Beck Anxiety Inventory (BAI) [122, 123]. The Skullcap flavone, baicalin which is found in american sckullcap and other members of the skuttleria genus has been shown to be anxiolytic by binding to $GABA_A$ receptors in mice [124, 125]. Interestingly biacalin increases hippocampal BDNF expression and in doing so protects the hippocampus from corticosterone induced depression in mice [126]. In addition biacalin stimulates neurite outgrowth in C17.2 neuronal stem cells by signaling through the ERK1/2 pathways, the known signal transduction triggered by NGF binding to TRKA [127].

2.12 Rhodeola Rosea

Rhodeola Rosea has been shown to reduce GAD in small pilot and self reporting clinical trials [128, 129]. While the effects of Rhodeola on brain neurochemistry has not been well studied, salidroside, a glycoside from Rhodiola has been shown to increase stem cells expression of neurotrophin-3 (NT-3), BDNF, NGF mRNA and induce differentiation into neurons [130] and also activates the ERK1/2 pathway in NGF treated PC12 cells [131].

2.13 Hops

One study shows Hops to be anxiolytic in clinical trials [132]. Prenylflavinoids from extracted from Hops can both bind to the benzodiazepine binding site on $GABA_A$ receptors [133, 134] and stimulate neurite outgrowth through TRKA a signaling in PC12 cells and cultures of dorsal root ganglia neurons [135, 136].

2.14 *Nigella sativa*

One clinical study shows that *Nigella sativa* seeds are anxiolytic in clinical trials [137]. Oral administration of Nigella extracts increase brain serotonin level in rats [138, 139] and thymoquinone, a terpine from Nigella is anxiolytic through

a GABAnergic pathway when orally administered to mice [140]. Thymoquinone promotes neurite outgrowth in rat hippocampal neurons and dorsal root ganglion neurons [141, 142].

3. Conclusion

Phytochemical nutrients that are used to reduce anxiety may have this affect in part by stimulating neuroplasticity and altering the brain neurocircuitry associated with learned responses to stressors and threats. Fourteen of the roughly forty-five plant and plant extracts proven to reduce anxiety in humans in clinical trials are also able to act like neurotrophins; endogenous molecules that stimulate neuroplasticity in the human brain. Anxiolytic drugs are harsh and symptoms return when the drug is removed because the neurotransmitter chemistry returns to an imbalance. Neuroplasticity offers an opportunity to use food phytochemicals along with drugs or in their place to learn to establish more appropriate responses to perceived threats by reworking neural connections. These new neural connections may not be lost even when the anxiolytic treatment is removed. Using neuroplastic drugs and foods to not only alter brain chemistry, but also the circuitry, would be a tremendous advancement in the treatment of anxiety.

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Acronyms and abbreviations

BDNF	Brain derive neurotrophic factor
NGF	Nerve growth factor
NT3	Neurotrophin-3
GDNF	Glial derived neurotrophic factor
TRK	Tropomyosin receptor kinase
GABA	Gama miniobutyric acid
CBD	Cannabidiol
GAD	Generalized anxiety disorder
PTSD	Post traumatic stress disorder
OCD	Obsessive compulsive disorder
PD	Panic disorder
SAD	Social anxiety disorder

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