Received: 17 July 2017 | Revised: 7 September 2017 | Accepted: 7 September 2017

DOI: 10.1002/ptr.5940

WILEY

REVIEW

GABA-modulating phytomedicines for anxiety: A systematic review of preclinical and clinical evidence

Karen Savage[1,2] | Joseph Firth[3,4] | Con Stough[2] | Jerome Sarris[1,4]

[1] ARCADIA Mental Health Research Group, Professorial Unit, The Melbourne Clinic, Department of Psychiatry, The University of Melbourne, 2 Salisbury Street, Richmond, Victoria 3121, Australia

[2] Centre for Human Psychopharmacology, Swinburne University of Technology, John St, Hawthorn, Victoria 3122, Australia

[3] Division of Psychology and Mental Health, University of Manchester, Oxford Rd, Manchester M13 9PL, UK

[4] NICM, School of Science and Health, Western Sydney University, Sydney, NSW, Australia

Correspondence
Ms Karen Savage, Department of Psychiatry and The Melbourne Clinic, The University of Melbourne, 2 Salisbury Street, Richmond, Victoria 3121, Australia.
Email: ksavage@unimelb.edu.au

Anxiety disorders are chronic and functionally disabling conditions with high psychological stress, characterised by cognitive symptoms of excessive worry and focus difficulties and physiological symptoms such as muscle tension and insomnia. Gamma-aminobutyric acid (GABA) is an inhibitory neurotransmitter within the central nervous system and is a key target of pharmacotherapies in the treatment of anxiety. Although current pharmaceutical treatments are often efficacious, they may cause undesirable side effects including cognitive decrements and withdrawal symptoms. Plant-based "phytomedicines" may provide novel treatment options, to act as an adjunctive or alternative to existing anxiolytic medications. As such, we conducted a systematic review to assess the current body of literature on anxiolytic phytomedicines and/or phytoconstituents. An open-ended search to 5 July 2017 was conducted using MEDLINE (PubMed), Scopus, and Cochrane library online databases and performed in a stepped format from preclinical to clinical investigations. Eligible studies must have had (a) *in vitro* evidence of GABA-modulating activity, (b) animal studies using anxiety models to test an anxiolytic effect, and (c) human clinical trials. Ten phytomedicines were identified as having preclinical investigations showing interaction with the GABA system, in addition to human clinical trials: kava, valerian, pennywort, hops, chamomile, *Ginkgo biloba*, passionflower, ashwagandha, skullcap, and lemon balm. Collectively, the literature reveals preclinical and clinical evidence for various phytomedicines modulating GABA-pathways, with comparative anxiolytic effect to the current array of pharmaceuticals, along with good safety and tolerability profiles.

KEYWORDS

anxiety, anxiolytic, GABA, herbal, phytomedicine, phytotherapy

1 | INTRODUCTION

Anxiety disorders are ubiquitous and persistent conditions following a chronic course with high comorbidity (Baxter, Scott, Vos, & Whiteford, 2013; Bruce et al., 2005). It is estimated over 15% of the population in developed nations will experience anxiety symptoms (which are subsumed under affective disorder diagnostic categories), yet they remain under-diagnosed and under-treated (American Psychiatric Association, 2013; Wittchen et al., 2011). The cognitive and neurophysiological dysfunction that characterises anxiety disorders has been linked to regional dysregulation of excitatory/inhibitory neurobiological pathways, notably in the limbic and prefrontal brain regions (Etkin & Wager, 2007; Martin, Ressler, Binder, & Nemeroff, 2010; Schienle, Hettema, Caceda, & Nemeroff, 2011).

Gamma-aminobutyric acid (GABA) is a nonstandard amino acid that acts as the principal inhibitory neurotransmitter in central nervous system (CNS) function. Up to 40% of all synapses in the CNS operate for GABA, and GABA receptors are found in every region of the human brain. Thus, GABA systems are implicit in a number of neurophysiological processes, including motor function, pain, sleep, brain development, and importantly for the current review, anxiety. Furthermore, impairments in GABA-mediated inhibition are seen in various neurological and psychological conditions, such as movement disorders, epilepsy, schizophrenia, insomnia, and anxiety disorders (Mohler, 2001).

Preclinical investigations examining the role of GABA substrates in symptoms of anxiety have uncovered several pharmacological targets for treatment of these disorders. These include GABA transporters type 1 or 2, GABA transaminase (GABA-T) or glutamic acid decarboxylase (GAD), and more recently, novel allosteric binding sites on GABA-A receptors (Nuss, 2015; Puthenkalam et al., 2016; Stahl & Moore, 2013). The classes of GABA receptors identified within the human brain include GABA-A, GABA-B, and GABA A-rho (formerly considered

GABA-C; Olsen & Sieghart, 2008). The GABA-A receptor exerts an inhibitory effect when activated, hyperpolarising the neuron and thus reducing the likelihood of an action potential occurring. Therefore, the active site of the GABA-A receptor, along with its various allosteric binding sites (which influence GABA-A receptor activity separately) and receptor subtype units, provides the main target for various anxiolytic, analgesic, and sedative drugs (Ramachandran & Shekhar, 2011).

However, current pharmacotherapeutic treatments that exert anxiolytic effects primarily via the aforementioned GABA-ergic pathways (such as benzodiazepines [BDZs]) provide only a modest benefit for alleviating symptoms in substantial numbers of patients and are noted for negative side effects (such as concentration and memory impairment), along with addiction/abuse issues (Baldwin, Ajel, & Garner, 2010; Hoffman & Mathew, 2008). As such, novel anxiolytic treatments that provide efficacious adjunctive or first-line treatment for anxiety, with fewer adverse consequences from long-term use, would prove highly valuable for patients with these disorders.

Various "phytomedicines" (i.e., whole-plant or plant-extract compounds, thought to play a preventative or therapeutic role in health/disease) have been shown in both preclinical in vitro and animal studies to possess certain pharmacodynamic properties that may confer anxiolytic effects. The pharmacodynamic effects of phytomedicines could be attributable to constituents that may include alkaloids, terpenoids/saponins, and polyphenols, as these molecules have various hypothesised actions within the CNS including binding with BDZ receptor sites (e.g., GABA-α allosteric sites); inducing ionic channel transmission by voltage-gated blockage via alteration of membrane structures; and/or modulating enzymatic processes such as GABA-T or GAD (Awad et al., 2007; Johnston, 2015). Flavonoids in particular are well-known positive modulators of GABA-A receptors (Hanrahan, Chebib, & Johnston, 2011).

Although existing reviews have examined phytomedicines for the treatment of anxiety, the literature thus far has limitations; for instance, covering affective disorders in general, or focusing on several neurotransmitter systems involved in anxiety symptomatology (Sarris, 2007; Sarris, McIntyre, & Camfield, 2013a). Previous reviews have also detailed only preclinical evidence for GABA-ergic effects from phytomedicines, thereby omitting important human studies, thus reducing the clinical applicability of their findings (Awad et al., 2007; Johnston et al., 2009). Others have reviewed both preclinical and clinical evidence for GABA-ergic effects of phytomedicines but focused on isolated conditions (such as insomnia; Shi, Dong, Zhao, Tang, & Zhang, 2014) or lastly, have included combination herbal formulas and/or other nutritional/dietary compounds (Boonstra et al., 2015; Liu et al., 2015), making it difficult to delineate the effects of phytomedicines specifically. Thus, we conducted the first systematic review combining both preclinical and clinical evidence to evaluate how phytomedicines (and/or their constituents) may act within the GABA system to produce anxiolytic effects, while assessing the current level of human evidence.

2 | METHODS

An electronic search was conducted of MEDLINE (PubMed), Scopus, and Cochrane library online databases from inception to 5 July 2017. The search was not restricted to English, where translation via online journal or digital translation of the publication was adequate. A three-level search strategy was employed to identify phytomedicines with all of the following attributes (a) in vitro studies examining GABA-ergic activity, (b) preclinical animal studies of any GABA-mediated behavioural effects, and (c) human trials reporting anxiety as a primary outcome. To identify in vitro/in vivo studies, the initial primary search terms were "PLANT," "HERBAL," "HERB," "PHYTOMEDICINE" and "GABA," "gamma-aminobutyric acid" and "ANXIOLYTIC," "ANXIOLYSIS," "STRESS," or "ANXIETY." Additionally, individual herbal medicines with both common names and Latin binomial names (where relevant) plus their known bioactive constituents, were specifically searched for. A forward search of papers included in the review was also conducted, using Web of Science and MEDLINE cited reference. A further search of grey literature was conducted to identify unpublished data.

Eligible in vitro studies were those that applied preclinical methodologies towards assessing GABA-related actions of phytomedicines, for example, *Xenopus laevis* oocyte cell line models or ex vivo brain tissue investigations. Phytomedicines with any indicated GABA-ergic activity were eligible at this stage, including those that demonstrated direct GABA-A or GABA-B receptor binding (at either active or allosteric sites), ionic channel or cell membrane modulation, GABA transaminase or GAD inhibition, and/or phytomedicines examined in interactions with known GABA receptor antagonists, such as flumazenil.

We then sought for preclinical studies that used established animal models of anxiety, along with related behavioural paradigms (e.g., grooming behaviour, maze models, or light/dark exposure models) to examine in vivo anxiolytic effects of phytomedicines found to have evidence of GABA-ergic activity revealed in Stage 1.

In the final stage of the search, to identify human trials of relevant phytomedicines, we conducted a further search using the terms: "Anxiety," "Generalised Anxiety Disorder," or "Anxiety Disorder," in addition to anxiety disorders as defined by Diagnostic and Statistical Manual of Mental Disorders, Edition 5 (American Psychiatric Association, 2013), for example, panic/agoraphobia, social phobia, and post-traumatic stress disorder, along with "AND" terms for a list of the phytomedicines (using both common names and Latin binomial names) that had been identified by the earlier search stages to have GABA-mediated anxiolytic effects in in vitro/in vivo studies. Human trials with any design were eligible (including randomised, open-label, or single arm studies), provided that the total sample size was ten or more participants (case studies were also excluded). In cases where a recent (i.e. since 2010) review (and meta-analysis of human trials) had already been conducted on the three levels of evidence for a specific phytomedicine included here (i.e., *Piper methysticum* and *Withania somnifera*), the review was used preferentially over detailing all the individual studies (although updated with a subsequent relevant research).

"Traditional" use of selected phytomedicines was cross-referenced with British Herbal Medicine Association Pharmacopoeia (British Herbal Medicine British Herbal Medicine Association, 1996) and Martindale Complete Drug Reference (Brayfield, 2014). The dosing regimen of phytomedicine products consisted mainly of a single dose unless otherwise specified as multiple doses.

3 | RESULTS

3.1 | Overview

The literature search found 1,328 articles relevant to initial search parameters, for which 1,212 were excluded following a check against criteria for preclinical or clinical phytomedicine studies. Ten identified phytomedicines (key constituents or whole plant preparations) met inclusion criteria and were reviewed. Three phytomedicines found to be lacking one of the three areas of evidence (preclinical tissue and animal and clinical human studies) are later discussed in brief in Section 4. Refer to Figure 1 for the selection processes flowchart.

Of the animal models employed, most being rodent type, the elevated-plus maze (EPM) was most typical, consisting of open and closed trajectories along with a timed measure of arm entry exploration, which reflect animal anxiety levels (Lister, 1987; Walf & Frye, 2007). Other paradigms included timing durations spent in light/dark and open field environments, and measuring grooming, conflict avoidance, and social behaviours. Anxiety measures used in the clinical studies were Hamilton Anxiety Scale (HAM-A; Hamilton, 1959); Spielberger State–Trait Anxiety Inventory (Spielberger, Gorsuch, Lushene, Vagg, & Jacobs, 1983); Erlangen anxiety, Tension and Aggression Scale (Galster & Spörl, 1979); the Befindlichkeitsskala Well-being Self-rating Scale (von Zerssen, 1976), Anxiety Sensitivity Index (Reiss, Peterson, Gursky, & McNally, 1986), and the Clinical Global Impressions Scale (Guy, 1976). Various biomarkers were also used as proxy measures of physiological anxiety, including hormones (adrenaline, cortisol, adrenocorticotropic hormone, and noradrenaline levels) as well as neurophysiological data—skin conductance, heart rate, and blood pressure.

3.2 | Anxiolytic phytomedicines with preclinical and human evidence

3.2.1 | *Centella asiatica* (Gotu cola/kola, pennywort)

Centella asiatica is a native Asian herbaceous species used commonly as a whole plant extract in Indian Ayurvedic and Chinese Traditional Medicine (Diwan, Karwande, & Singh, 1991; Gohil, Patel, & Gajjar, 2010). Modern indications are for dermatotic, antimicrobial, nociceptive, cognition-enhancing, anxiolytic, and anti-depressive purposes (Anukunwithaya, Tantisira, Tantisira, & Khemawoot, 2016; Lokanathan, Omar, Ahmad Puzi, Saim, & Hj Idrus, 2016). The chief constituents are triterpenoid glycogens (saponins), sterols, flavonoids, tannins, and stearic acids. Investigations of constituents have chiefly involved the saponins brahmoside and brahminoside and the asiatic and ursolic acids (Brinkhaus, Lindner, Schuppan, & Hahn, 2000; James & Dubery, 2009).

GABA-ergic effects of *C. asiatica* (and constituents) were revealed in studies analysing GABA-T and GAD activity. One study (Awad et al., 2007) reported *C. asiatica* significantly increased GAD activity in vitro by more than 50% ($p < .001$) in male Sprague–Dawley rat brain assays. In a further in vitro study of GABA-A subtype receptor modulation (Hamid et al., 2016), the asiatic acid constituent was found to be negative modulator of GABA-induced currents for α1 β2 γ2L, α2 β2 γ2L, and α5 β3 γ2L receptors in *Xenopus* oocyte tissue. Further, in an ex vivo study using Charles Foster rats (Chatterjee, Chakraborty, Pathak, & Sengupta, 1992), *C. asiatica* extract was found to increase whole brain levels of GABA.

In rodent models, the constituent triterpenoid extracts of *C. asiatica* (madecassoside and asiaticoside) have been shown to be as effective as diazepam for alleviating behavioural symptoms of

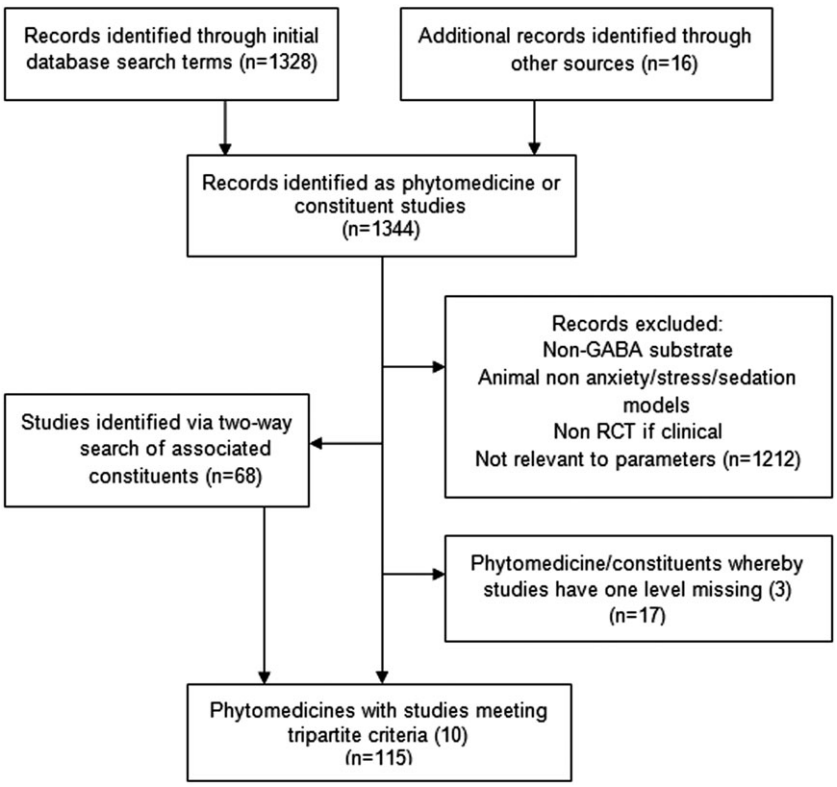

FIGURE 1 PRISMA flow chart showing the process of literature search and studies selection

anxiety in chronic stress settings of EPM, open-field, and dark–light box tests, as well as acute stress models (i.e., immobilisation stress test) in adult male imprinting control region mice (Wanasuntronwong, Tantisira, Tantisira, & Watanabe, 2012). Another study (Wijeweera, Arnason, Koszycki, & Merali, 2006) reported significant anxiolytic effects in EPM tests from both methanol and ethyl acetate extracts of *C. asiatica*, along with the isolated asiaticoside constituent in male Sprague–Dawley rats. A final animal study (Ceremuga et al., 2015) investigated the anxiolytic effects of an injected peritoneally asiatic acid extract in four male Sprague–Dawley rat comparison groups. Asiatic acid significantly increased EPM measures of open-arm latency and mobility speed and latency (all indicating reduced anxiety). Further, the investigation reported that administration of GABA receptor antagonist flumazenil attenuated the anxiolytic effects of asiatic acid, thus indicating that the anxiolytic effects of asiatic acid are due to GABA-ergic benzodiazepine receptor activity.

A human study examined the effects of an acute dose of *C. asiatica* extract (12 g) on measures of stress and anxiety, including acoustic startle response, against a placebo comparator in 40 healthy adults (Bradwejn, Zhou, Koszycki, & Shlik, 2000). The results showed significant attenuation of acoustic startle response in the treatment group. Another clinical investigation conducted an open-label study of 500 mg of the phytomedicine twice daily for 2 months in a sample of 33 patients with GAD (Jana, Sur, Maity, Debnath, & Bhattacharyya, 2010). The study found significant improvements in anxiety following 60 days of treatment ($p < .01$) on the HAM-A scale, along with significant reductions in secondary measures of stress and depression.

3.2.2 | *Humulus lupulus* (Hops)

The flowering cones of *Humulus lupulus* (Cannabaceae) have sedative/hypnotic, antibacterial, anti-inflammatory traditional indications; however, the plant is chiefly cultivated worldwide for alcoholic beverages derived from malted grain, namely "beer" (Katsiotis, Langezaal, & Scheffer, 1989). Modern medicinal indications exist for anxiety and insomnia (Blumenthal, 2009; Martin, McCallum, Stofer, & Eichinger, 2002), mostly associated with principal terpenoid and flavonoid alpha and beta acid constituents, such as humulene and myrcene.

In vitro studies of myrcenol (found in the *H. lupulus* plant) have demonstrated GABA-A receptor response potentiation in *Xenopus* oocytes (Aoshima et al., 2006). Additionally, a combination study (Muller, Schumacher, Brattstrom, Abourashed, & Koetter, 2002) investigated how hops extract and *Valeriana officinalis* influenced binding at adenosine receptors within rat cortical and striatum membranes and found that although *V. officinalis* in combination with hops affected this process, hops alone did not—indicating that the mechanism through which hops produces sedative effects may be through GABA receptor activity instead. Preclinical research further supported GABA-ergic effects of *H. lupulus* (extract 0.11–0.65 mg/mL) in male Sprague–Dawley rat brains (Awad et al., 2007), through inhibiting GAD activity.

Animal models examining effects on sedation and anxiety have also been conducted: one investigation (Franco, Sanchez, Bravo, Rodriguez, Barriga, & Juanez, 2012a) found that *H. lupulus* extract reduced nocturnal activity in an avian model (common quail) and preserved an activity/rest cycle that matches regular circadian rhythms. Similarly, an investigation using female U.S. Naval Medical Research Institute (NMRI) mice found that *H. lupulus* extract reduced locomotor activity and reduced body temperature consistent with sedating properties (Schiller et al., 2006). However, a preclinical study of *H. lupulus* specific constituent xanthohumol reported no evidence of anxiolysis via GABA-ergic mechanisms of this particular component using male Sprague–Dawley rats, suggesting effects occur by a different constituent or pathway (Ceremuga, Johnson, Adams-Henderson, McCall, & Johnson, 2013).

In human studies, one investigation (Franco, Sanchez, Bravo, Rodriguez, Barriga, Romero, et al., 2012b) measured sleep quality, circadian rhythms, and anxiety/stress levels of 17 "work stressed" shift workers for a regular seven nights (control period) and then following 14 days of nightly ingestion of a 330 ml alcohol-free beer. The study found significant reductions in state anxiety versus the control condition. Aside from this study, the anxiolytic effects of *H. lupulus* have not been widely examined, with the majority of studies focusing on sedating and hypnotic properties instead. As such, the anxiolytic potential of *H. lupulus* in anxiety symptom alleviation is yet to be substantiated through clinical research, despite clear indications of GABA-ergic mechanisms from preclinical research.

3.2.3 | *Ginkgo biloba* (Maiden hair)

The ginkgo tree (*Ginkgo biloba*) is one of the oldest angiosperm species, with the nuts and leaves being used for a range of therapeutic effects (Shizhen, 1990). Active constituents are mainly flavonoids quercetin and catechin, unique terpenoids ginkgolides and bilobalides, and proanthocyanidins (van Beek & Montoro, 2009). Tissue studies of human endothelial cell lines indicate that constituents from the leaves as standardised extract have potent neuroprotective and anti-apoptotic properties, as well as vasculatory benefits via nitric oxide (NO) enhancement (Koltermann et al., 2007). Modulations to cholinergic and monoaminergic pathways have also been evidenced in female Wistar rats and NMRI mice models ex vivo, thereby offering benefit for cognition and mood (Fehske, Leuner, & Muller, 2009).

Preclinical studies were located investigating GABA-ergic mechanisms of action; however, to date the findings are not entirely clear. The ginkgolides A, B, C and bilobalide were reported to operate as antagonists at recombinant α1 β2 γ2L GABA-A receptors expressed in *Xenopus* oocytes in tissue investigations, and to exact mixed antagonist action involving similar models with recombinant human ρ1 GABA-C receptors (Huang et al., 2003, 2004, 2012). Additionally, an increase of GABA in hippocampal, cortical, and striatum tissue in male ddY strain mice assayed ex vivo was reported in the presence of bicuculline and pictrotoxin, thought to occur via potentiating GAD and thus operating as an anticonvulsant (Sasaki, Hatta, Haga, & Ohshika, 1999; Sasaki, Hatta, Wada, Ohshika, & Haga, 2000). A tissue study also reported that both the ginkgolides and bilobalide selectively inhibit GABA-A receptors to the same extent as the pictrotoxin comparator in Sprague–Dawley rat brain tissue ex vivo when exposed to ginkgolides A, B, C, and J and bilobalide (Ivic et al., 2003). The study discussed structural similarities of *G. biloba* constituents to the GABA-A receptor antagonist, picrotoxinin. One investigation examined further the GABA-A mechanisms of action for bilobalide in rat

hippocampal tissue and reported a blocking of GABA-induced chloride uptake in these cells at low potency. In turn, GABA was found to partially antagonise bilobalide's inhibitory actions (Kiewert et al., 2007). One recent study reported that both bilobalide and ginkgolide B selectively inhibited structurally-varied GABA-A positive modulator agent actions when tested in *Xenopus* oocytes (Ng, Duke, Hinton, & Johnston, 2017).

In anxiety rodent models, ginkgolides A, B, C, and bilobalide (individually and combined) were tested using the EPM paradigm with mice (Kuribara, Weintraub, Yoshihama, & Maruyama, 2003). The study found significant dose-dependent relationships between ginkgolide A with reductions in anxiety (i.e., increased open arm time). However, these beneficial effects were not blocked by flumazenil, suggesting that these constituents exert anxiolytic effects through alternate routes to GABA receptors. A further preclinical study of bilobalide treatment found increased locomotor activity in open field and EPM tests and reduced latency in Morris water maze, using male Kunming mice, with glucocorticoid receptor modulation as a hypothesised mechanism of action (Ma et al., 2012). One human study testing *G. biloba* extract at doses of either 480 or 240 mg or placebo in 170 patients with GAD reported significant improvements on HAM-A scale in the treatment groups after 4 weeks, with greatest effects in the high-dose condition (Woelk, Arnoldt, Kieser, & Hoerr, 2007).

3.2.4 | *Matricaria recutita/Matricaria chamomilla* (Chamomile, German chamomile)

Matricaria recutita, an Asteraceae (daisy) family flowering plant, is plentiful throughout Eurasia and has been utilised for millennia, chiefly as tea, for calming/sedative effects (McKay & Blumberg, 2006). The bioactive constituents include sesquiterpenes, coumarins (herniarin, umberlliferone, among others), and flavonoids including apigenin, luteolin and rutin, and phenylpropanoids (chlorogenic and caffeic acids; Srivastava, Shankar, & Gupta, 2010).

Tissue studies report that certain *M. recutita* constituents modulate the function of GABA receptors via ligand effects on the central BDZ site, with a reduction of GABA-activated Cl- currents, with evidence of action in other (possibly monoamine) pathways in Sprague–Dawley rat brain assays (Avallone, Zanoli, Corsi, Cannazza, & Baraldi, 1996; Avallone et al., 2000). Principally, the flavonoid apigenin has been investigated—present also in other plant species—with anxiolytic effects as an inverse BDZ agonist (Johnston, 2015; Viola et al., 1995), potentially biphasic action via receptor subunits similar to BDZs (Campbell et al., 2004; Hanrahan et al., 2011) as the activity on BDZ receptors is somewhat equivocal (aforementioned Avallone et al., 2000). Animal models have supported this, reporting sedative and anxiolytic effects. For instance one study (Shinomiya et al., 2005) found administration of the extract improved sleep latency in male Wistar rats and that these effects were attenuated from co-administration of flumazenil, thus implicating GABA-ergic BDZ receptor modulation as the likely mechanism when measured ex vivo. Another study further reported significant reductions in anxiety behaviours in male Balb/c mice via in various testing paradigms, including open field, EPM, and social interaction paradigms (Can, Demir, Kiyan, & Demirci, 2012).

In humans, a double blind randomized controlled trial (RCT) with 57 GAD patients showed significantly greater reductions in anxiety (HAM-A) among those undergoing *M. recutita* treatment (220 mg, one to four times daily) than those in the control condition, after 8 weeks (Amsterdam et al., 2009). A post hoc analysis of the same sample found that *M. recutita* may also reduce co-morbid depression (HAM-D) in anxious patients (Amsterdam et al., 2012).

Three further human studies reported short- and long-term (i.e., 8 and 38 weeks) effects of treatment for anxiety with 1500 mg (500 mg capsule 3 times daily) in a sample of 179 patients with GAD. At 8 weeks, 58% of patients met the criteria for "clinical response" to *M. recutita* treatment, with significant reductions in mean anxiety (GAD-7) across the entire sample (Keefe, Mao, Soeller, Li, & Amsterdam, 2016). For the long-term study, those who were "responders" were randomly assigned to continued active or placebo treatment for a further 26 weeks. GAD relapse rates were 25.5% in the placebo group and only 15.2% in the continued treatment group (Mao et al., 2016). Although the difference in relapse rates did not reach statistical significance, GAD-7 ratings were significantly improved in the continuation versus placebo conditions. Another clinical investigation in 34 patients with DSM-IV primary insomnia found significant improvements over placebo from 28 days of 270 mg *M. recutita* twice daily to ratings of fatigue, sleep quality and latency, but with no significantly relevant changes in symptoms of anxiety (Zick, Wright, Sen, & Arnedt, 2011).

3.2.5 | *Melissa officinalis* (Lemon Balm)

Lemon balm is reported in Theophastrus' Historia Plantarum (350–c. 287 BC), thus holding longstanding use for millennia. It is native to Eurasia as a member of the Lamiaceae (mint) family, with a number of phytochemicals including phenolic acids, terpenes, rosmarinic and caffeic acids, eugenol acetate, and tannins. Indications are for antibacterial, anti-stress, hypnotic, and gastrointestinal symptom benefit (Shakeri, Sahebkar, & Javadi, 2016; Ulbricht et al., 2005).

In vitro studies have indicated various GABA substrate activity for *M. officinalis* constituents such as elevation of GABA levels, owing to inhibition of GABA-T, as observed in male Sprague–Dawley rat brain homogenised tissue samples (Awad et al., 2007; Awad, Muhammad, Durst, Trudeau, & Arnason, 2009). An aqueous extract of *M. officinalis* (rosmarinic acid) inhibited GABA-T activity (IC50 = 0.35 mg/ml) in the former study (Awad et al., 2007) and replicated in the latter study (Awad et al., 2009). An additional investigation of the triterpenoids, ursolic and oleanolic acids found that these constituents also increase GABA-A BDZ receptor affinity in rat brain tissue (Salah & Jager, 2005). A neurotropic tissue study (Yoo et al., 2011) reported decreased GABA-T levels in the dentate gyrus of C57BL/6 J mice, along with an increase in total GABA levels, following extract 50/200 mg/kg daily for 3 weeks. Finally, in radio-ligand/electrophysiological studies using male Wistar rat cortical cultures in vitro, *M. officinalis* extract and selected constituents have shown trans-ocimene inhibited [35S] (TBPS) binding (set radio ligand) at GABA-A receptor level, in a dose-dependent fashion (Mahita et al., 2014), building upon previous similar findings in preclinical investigations (Abuhamdah et al., 2008; Huang et al., 2008).

Rodent model studies have reported a dose-dependent response on the EPM with C57 Bl/6 Jico mice using the extract "Cyracos" (Naturex, *M. officinalis* L. Extract), with principal constituents

rosmarinic acid and triterpenoids oleanolic and ursolic acids (Ibarra, Feuillere, Roller, Lesburgere, & Beracochea, 2010). Similarly, a second study (Taiwo et al., 2012) used EPM and open field measures to show significant anxiolytic effects of *M. officinalis* extract in male and female Wistar rats, which matched those of the diazepam comparator.

In contrast, one preclinical study (Raines et al., 2009) reported the constituent luteolin confers additive or synergist effects in the presence of a GABA-A agonist but not alone; reporting no differences in anxiety behaviours using an EPM model with male Sprague–Dawley rats between the constituent and a flumazenil group, both administered with a GABA-A BDZ agonist, suggesting that anxiolysis also occurs via different mechanisms.

In human investigations, an open-label study (Cases, Ibarra, Feuillere, Roller, & Sukkar, 2011) in a self-reported "stressed" sample, showed a 15–42% reduction in insomnia and mild-to-moderate anxiety symptoms over 15 days from 600 mg per day of Cyracos (standardised to 7% rosmarinic acid and greater than 15% hydroxycinnamic acid derivatives). In a healthy sample study (Kennedy, Scholey, Tildesley, Perry, & Wesnes, 2002) a laboratory stress-induction paradigm showed improvements in mood and attention from *M. officinalis* extract. Several similar studies with other laboratory-based paradigms have also reported improvements from *M. officinalis*, which are consistent with anxiolytic-like effects (Kennedy et al., 2003; Kennedy, Little, & Scholey, 2004; Scholey et al., 2014). Finally, a double-blind RCT in 100 school age females (Akbarzadeh et al., 2015) reported efficacy of 1200 mg of *M. officinalis* extract daily over 3 months for reducing stress and tension associated with premenstrual dysphoric disorder.

3.2.6 | *Passiflora incarnata* (Passion flower)

Passiflora incarnata is native to the Americas having been utilised for millennia for conditions of nervousness, anxiety, and sleep issues (Krenn, 2002; Meier, 1995; Miroddi, Calapai, Navarra, Minciullo, & Gangemi, 2013). The chief identified constituents include alkaloids such as chrysin, flavonoids such as schaftoside, isoschaftoside and swertisina, and phenolic compounds (Wohlmuth, Penman, Pearson, & Lehmann, 2010). GABA-ergic modulation could feasibly occur through number of processes, including affinity with GABA-A and GABA-B receptor subtypes and GABA reuptake inhibition, and also via positive allosteric modulation of the GABA-A receptor complex through BDZ sites (Appel et al., 2011; Nassiri-Asl, Zamansoltani, & Shariati-Rad, 2008).

A combined in vitro/in vivo study (Elsas et al., 2010) used a pentelenetrazol-induced seizure procedure in a CF-1 mice model to examine effects of *P. incarnata* extract, which was reported to induce direct GABA-A currents in CA1 hippocampal pyramidal neurons. Another combined in vivo/in vitro study (Grundmann, Wang, McGregor, & Butterweck, 2008) utilised a *P. incarnata* extract (as homoorientin, orientin, vitexin, and isovitexin) in male BL6/C57 J mice using an EPM model to show behavioural anxiolytic effects matching those of diazepam from moderate-strength doses. Furthermore, the findings that flumazenil acted as an antagonist against *P. incarnata* demonstrates GABA-mediated pathways for the anxiolytic effects. A related study (Grundmann, Wahling, Staiger, & Butterweck, 2009) examined effects in a similar rodent EPM model using three strengths of *P. incarnata* with diazepam comparator, finding a U-shape trend dose–response efficacy, in favour of the middle dose (375 mg/kg).

Several in vivo studies investigated both dose and constituent optimisation; the first (Dhawan, Kumar, & Sharma, 2001) used a Swiss mice (male and female) EPM model to delineate potency of different plant components, reinforcing use of the flower for maximal anxiolytic effects. A second study (Sampath, Holbik, Krenn, & Butterweck, 2011) used differential EPM performances in male C57BL/6 J mice to ascertain optimal fractionalisation methods, and a third (Wolfman, Viola, Paladini, Dajas, & Medina, 1994) used anxiolytic and sedative paradigms (EPM, hole board and horizontal wire) in male CF1 mice to examine the constituent chrysin (5,7-dihydroxyflavone) with a diazepam comparator, reporting no significant differences in anxiolytic effect, but an increase to head-dipping and myo-relaxant properties via horizontal wire test in the BDZ group. Finally, a recent study (Aman et al., 2016) observed anxiolytic and sedative effects of *P. incarnata* in staircase and open field tests, with BALB/c mice and female Sprague Dawley rats, which were antagonised by pentylenetetrazole, indicating the behavioural effects of *P. incarnata* occurred through GABA-ergic mechanisms; however, precise mechanisms were again unclear.

Seven studies involving human populations were found; four examined effects on preoperative anxiety in RCTs (Aslanargun, Cuvas, Dikmen, Aslan, & Yuksel, 2012; Dantas, de Oliveira-Ribeiro, de Almeida-Souza, & Groppo, 2017; Kaviani, Tavakoli, Tabanmehr, & Havaei, 2013; Movafegh, Alizadeh, Hajimohamadi, Esfehani, & Nejatfar, 2008) all reporting significant benefits from *P.incarnata* treatment in comparison to placebo conditions and/or non-inferiority to BDZ comparators. Most recently, this effect was observed in a double-blind RCT of 40 dental surgery patients (Dantas et al., 2017), showing 260 mg of *P. incarnata* orally administered 30 min before dental surgery reduced subjective anxiety along with physiological indicators (i.e., blood pressure and heart rate) to the same extent as midazolam, with fewer cognitively-impairing side effects.

The BDZ-like efficacy has also been observed for psychiatric disorders. First, a double-blind RCT in 36 patients with GAD (Akhondzadeh et al., 2001) found no significant difference between *P. incarnata* extract (administered as liquid drops) to oxazepam comparator after 4 weeks. Second, another investigation (Mori, Hasegawa, Murasaki, Yamaguchi, & Ito, 1993) found no difference between extract ("Passiflamin" 90 mg) and mexazolam comparator groups for anxiety measures in patients with "neurosis."

3.2.7 | *Piper methysticum* (Kava)

Kava (*Piper methysticum*) is a plant native to the South Pacific, where the roots have been used in traditional medicine for a range of conditions via its anxiolytic, nootropic and neuroprotective, nociceptive, and dysphoric actions (Singh, 1992; Singh & Singh, 2002; Tzeng & Lee, 2015). The chief bioactive constituents are the six lipophilic kavalactones, of which kawain and dihydrokawain are evidenced to exert the strongest anxiolytic activity. Mechanisms evidenced in preclinical models primarily involve modulation of GABA receptors, and although the exact mechanisms are not clear, these models suggest several avenues beyond an allosteric mode of action in the GABA substrate, such as rapid upregulation of

GABA-A receptor function, blockade of voltage-gated sodium ion channels, enhanced ligand binding across GABA-A receptor subtypes, and reduced excitatory neurotransmitter release via blockade of calcium ion channels (Gleitz, Beile, & Peters, 1995; He, Lin, & Lian, 1997; Jussofie, Schmiz, & Hiemke, 1994; Magura, Kopanitsa, Gleitz, Peters, & Krishtal, 1997; Mathews et al., 2005). Table 1 details these studies further. Abundant animal anxiety model investigations exist (e.g., Davies, Drew, Duffield, Johnston, & Jamieson, 1992; Garrett, Basmadjian, Khan, Schaneberg, & Seale, 2003; Rex, Morgenstern, & Fink, 2002), reporting anxiolytic effects (such as reduced entry latencies and increased time in unfamiliar environments) in rodent EPM and other paradigms.

A number of clinical reviews published have systematically examined the mechanisms and efficacy of P. methysticum in the clinical treatment of anxiety, including an early Cochrane review (Pittler & Ernst, 2003), along with more recent reviews of preclinical and clinical investigations (LaPorte, Sarris, Stough, & Scholey, 2011; Sarris et al., 2013a; Sarris, McIntyre, & Camfield, 2013b). From human clinical studies these reviews indicate *P. methysticum* is superior to placebo in the treatment of anxiety symptoms. For instance, a pooled effect size calculated for HAM-A scores across 6 RCTs found large, significant reductions in anxiety in favour of the extract (Cohen's d = 1.1) (Sarris, LaPorte, & Schweitzer, 2011). A subsequent RCT has also confirmed these findings (Sarris et al., 2013c) observing moderate effects on reducing anxiety reported with the HAM-A in a GAD sample (n = 75) treated with either P. methysticum (120/240 mg kavalactones), compared to placebo, for 6 weeks.

Through combining human evidence alongside preclinical (in vitro and in vivo) studies, reviews have also found strong indication that the anxiolytic effects of P. methysticum are due (at least in part) to the extract's multiple GABA-ergic actions (Sarris et al., 2013a, 2013c) Again, the conclusions drawn from these earlier reviews have been supported by recent studies, whereby GABA-ergic mechanisms are investigated in more detail; in a recent tissue study, examining actions of a specific *P. methysticum* phytoconstituent "kavain" on expressed *Xenopus* oocytes (Chua et al., 2016) found that positive allosteric modulation occurred across all GABA-A receptor subtypes examined (α1β2, β2γ2L, αxβ2γ2L (x = 1,2,3, and 5), α1βxγ2L (x = 1, 2, and 3), and α4β2δ), and where this action was not affected by administration of flumazenil, implicating sites other than BDZ receptors.

3.2.8 | *Scutellaria lateriflora* (Scullcap, Blue Skullcap)

Scutellaria lateriflora is native to America and Europe with use in Native American practice and Western medicine for anti-anxiety, relaxant, and antispasmodic effects (Mills & Bone, 2000). Identified chief bioactive constituents include a number of flavonoids: scutelaterin A, baicalin, bacalein, apigenin, oroxylin A, and wogonin (Li, Wang, Smillie, & Khan, 2012; Zhang, Lian, Li, & Stringer, 2009). Preclinical studies have found several constituents of *S. lateriflora* have GABA-ergic properties, including baicalin and baicalein, wogonin, and apigenin, as these constituents act at the BDZ site of GABA-A receptor via positive allosteric modulation (Awad et al., 2003; Wang, Xu, Ren, Tsang, & Xue, 2008; Xu et al., 2006). Animal studies have also shown behavioural changes that reflect these GABA-mediated anxiolytic properties. For instance, one investigation (Awad et al., 2003) observed *S. lateriflora* extract significantly increased open field entries, head dips, and time spent in open arms in an EPM paradigm with male Sprague–Dawley rats.

Two human studies using *S. lateriflora* extract were found, both in non-clinical samples. One double-blind crossover study (n = 43; Brock, Whitehouse, Tewfik, & Towell, 2014) reported significantly greater mood improvement after 2 weeks from 350 mg extract (three times per day) than placebo comparator. Although there were no differences in anxiety outcomes between groups; this was ascribed to a "floor effect" as these healthy volunteers were mostly non-anxious at baseline. Similarly, an earlier briefly-detailed pilot investigation reported clinically significant reductions in anxiety and stress from various strengths of *S. lateriflora* in 19 healthy volunteers (Wolfson & Hoffmann, 2003).

3.2.9 | *Valeriana officinalis* (Valerian)

Valerian is a perennial plant native to mainly Europe, with the root extract utilised medicinally for millennia as a sedative and anxiolytic. The bioactive constituents include alkaloids, flavonones, and terpenes (specifically the sequiterpene valerenic acid; Houghton, 1988). The results of preclinical studies have indicated that *Valerian spp*. modulates the GABA-ergic system through potentiation of GABA release and inhibition of reuptake or degradation, via inhibited GABA-T activity in brain tissue, from male Sprague–Dawley rats, female Swiss-Webster mice, and male Wistar rats, respectively (Awad et al., 2007; Ortiz, Nieves-Natal, & Chavez, 1999; Santos et al., 1994).

Valerenic acid as the principle constituent was identified as a subunit-specific allosteric modulator of GABA-A receptors, specifically acting as an agonist at β2/3 subunits in male RjHan:WI rat cortical tissues and in *Xenopus* oocytes tissue investigations, likely to interact with the loreclezole binding pocket (Becker, Felgentreff, Schroder, Meier, & Brattstrom, 2014; Khom et al., 2007, 2016; Trauner et al., 2008).

The in vitro findings are reflected through in vivo studies; with animal models demonstrating anxiolytic properties of valerian extracts with female NMRI mice and male Sprague–Dawley rats on the EPM (and also anti-depressant effects via forced swim test model; Hattesohl et al., 2008), that it is equitable to BDZ comparators in female Hooded Wistar rats (Murphy, Kubin, Shepherd, & Ettinger, 2010). There is a strong indication that these behavioural effects are due to valerenic acid and valerenol constituents modulating GABA-A receptors, specifically β3 subtypes from one combined in vitro and in vivo study involving human embryonic kidney cell lines, and in β3 point-mutated mice in EPM and light–dark choice behavioural tests, using valerenic acid and valerenol extracts (Benke et al., 2009).

In human studies, one investigation (Kohnen & Oswald, 1988) showed that valerian extract reduced subjective feelings of anxiety among healthy adults in a stress-inducing scenario and that these effects occurred independently of the beta-blocker propranolol. Additionally, another study (Ahmadi, Khalili, Abbasian, & Ghaeli, 2017), in a double-blind RCT with 51 HIV-positive patients, found that 530 mg of V. officinalis extract daily for 4 weeks significantly reduced the anxiety that commonly occurs as a neuropsychiatric side effect of antiretroviral therapy.

In affective disorder samples, a placebo-controlled, 8 week clinical study of *V. officinalis* root extract (765 mg per day) among 31 adults with obsessive-compulsive disorder (DSM-IV diagnosed) found the extract significantly reduced symptoms of the disorder (Pakseresht, Boostani,

TABLE 1 List of selected phytomedicines with preclinical (in vitro and animal) and clinical evidence

Botanical name	Common name	Active constituents	GABA substrate mechanism	Preclinical animal anxiety models	Clinical group
Centella asiatica	Gotu kola, pennywort	Asiatic acid triterpenoids	Stimulation of glutamic acid decarboxylase; selective GABA-B agonist Awad et al. (2007); Chatterjee et al. (1992); Hamid et al. (2016)	EPM, open-field test, dark–light box Ceremuga et al. (2015); Wanasuntronwong et al. (2012); Wijeweera et al. (2006)	Acoustic startle response Bradwejn et al. (2000); Jana et al. (2010)
Humulus lupulus	Hops	Terpenoids and flavonoids: humulene, myrcene, and xanthohumol	Glutamic acid decarboxylase inhibition Awad et al., 2007	EPM, avian model of nocturnal behaviours Ceremuga et al., 2013; Franco, Sanchez, Bravo, Rodriguez, Barriga, and Juanez, (2012a); Schiller et al. (2006)	Menopausal females; "work stressed" healthy sample Franco, Sanchez, Bravo, Rodriguez, Barriga, Romero, et al. (2012b)
Ginkgo biloba	Maiden hair, *Ginkgo*	Terpenoids: bilobalides, gingkolides A, B, and C	GABA-A and GABA-C receptors anatagonist; increased GABA levels and glutamic acid decarboxylase enzyme activity Huang et al. (2003, 2004, 2006, 2012); Ivic et al., 2003; Kiewert et al., 2007; Ng et al., 2017; Sasaki et al., 1999, 2000	EPM, open field, water maze Kuribara et al. (2003); Ma et al. (2012)	Generalised anxiety disorder Woelk et al. (2007)
Matricaria recutita/Matricaria chamomilla	Chamomile, German chamomile	Flavonoids, Sesquiterpenoids, coumarins, phenylpropanoids: apigenin, luteolin, rutin, herbiarin, umberlliferone, chlorogenic and caffeic acids	GABA-BZD receptor ligand binding Avallone et al. (1996, 2000); Johnston (2015); Viola et al. (1995)	EPM, open field, social interaction Can et al. (2012); Shinomiya et al. (2005)	Generalised anxiety disorder, insomnia Amsterdam et al. (2009, 2012); Keefe et al. (2016); Mao et al. (2016); Zick et al. (2011)
Melissa officinalis	Lemon balm	Flavonoids, volatile oils, triterpenoids: rosmarinic acid, oleanolic acid, ursolic acid	GABA-T inhibition; GABA-A BDZ receptor affinity; elevation of brain GABA levels Awad et al. (2007); Awad et al. (2009); Abuhamdah et al. (2008); Huang et al. (2008); Mahita et al. (2014); Raines et al. (2009); Salah and Jager (2005); Yoo et al. (2011)	EPM, open field rodent models Ibarra et al. (2010); Taiwo et al. (2012)	Stress induction paradigm in healthy controls; general anxiety; sleep disorders; premenstrual dysphoria Akbarzadeh et al. (2015); Cases et al. (2011); Kennedy et al. (2002, 2003, 2004); Scholey et al. (2014)
Passiflora incarnata	Passionflower	Flavonoids: chrysin, b-carboline alkaloids	GABA-A BDZ positive allosteric modulation; GABA-A and GABA-B receptor subtype affinities	EPM, hole board, horizontal wire rodent models Aman et al. (2016); Dhawan et al. (2001); Elsas et al. (2010);	Generalised anxiety disorder; pre-surgery Anxiety Akhondzadeh et al. (2001); Aslanargun et al. (2012), Dantas

(Continues)

TABLE 1 (Continued)

Botanical name	Common name	Active constituents	GABA substrate mechanism	Preclinical animal anxiety models	Clinical group
			Elsas et al. (2010); Grundmann et al. (2008)	Grundmann et al. (2008); Sampath et al. (2011), Wolfman et al. (1994)	et al. (2017); Kaviani et al. (2013); Mori et al. (1993); Movafegh et al. (2008).
Piper methysticum	Kava	Kavalactones: kavain and dihydrokavain	GABA-A receptor positive modulators, ligand binding at all subtypes, also NaCl, Ca + gated ion channels	EPM	Generalised anxiety disorder; phobia; social phobia; nonpsychotic anxiety; stress in healthy samples; menopausal anxiety
			Baxter et al (2013); Boonen and Haberlein (1998); Chua et al. (2016); Davies et al. (1992); Gleitz et al. (1995); He et al. (1997); Jussofie et al. (1994); Magura et al. (1997); Martin et al. (2002); Mathews et al. (2005)	Garrett et al. (2003); Rex et al. (2002)	LaPorte et al. (2011); Pittler and Ernst (2003); Sarris (2007); Sarris et al. (2011, 2013a, b, c); Singh & Singh (2002)
Scutellaria lateriflora	Scullcap	Flavonoids: bacalin, baicalein, scutelaterin A, apigenin, oroxylin A, wogonin	GABA-A receptor positive allosteric modulation	EPM, open field	Healthy sample; general anxiety
			Awad et al. (2003); Wang et al. (2008); Xu et al. (2006)	Awad et al. (2003)	Brock et al. (2014); Wolfson and Hoffmann (2003)
Valeriana spp.	European or Mexican valerian	Sesquiterpenoids: valerenic acid, valepotriates, alkaloids, flavonones	Glutamic acid decarboxylase stimulatory effect; GABA-A receptor binding, specifically GABA-A chloride channel receptor complex: β2 or β3 subunit (N265 M); GABA-A receptor allosteric modulation	EPM, light/dark choice test, seizure model	Children with anxiety; HIV adults; healthy sample driving; OCD; generalised anxiety disorder
			Awad et al. (2007); Becker et al. (2014); Khom et al. (2007, 2016); Ortiz et al. (1999); Ramharter and Multzer (2009); Santos et al. (1994); Trauner et al. (2008)	Benke et al. (2009); Khom et al. (2016); Hattesohl et al. (2008); Murphy et al. (2010)	Ahmadi et al. (2017); Andreatini et al. (2002); Gutierrez et al. (2004); Pakseresht et al. (2011); Thomas et al. (2016);
Withania somnifera	Ashwaghanda, Winter Cherry, Indian Ginseng	Glycosides alkaloids and steroidal triterpenoid lactones: Glycowithanolides, withanone, withaferin A, withanolides A,D, and G, withanine, withasamnin, withanoloids	Ionotropic GABA-A receptors; GABAρ1 receptor agonists	EPM, social interaction; feeding latency, unfamiliar environment	Generalised anxiety disorder ; general anxiety; chronic stress; bipolar disorder
			Candelario et al. (2015); Yin et al. (2013)	Bhattacharya et al. (2000); Kaur et al. (2016)	Andrade, Aswath, Chaturvedi, Srinivasa, and Raguram (2012); Auddy et al. (2008); Cooley et al. (2009); Chandrasekhar, Kapoor, and Anishetty (2012); Chengappa et al. (2013); Khyati and Ayup (2013)

Note. BZD = benzodiazepine; EPM = elevated-plus maze; GABA = gamma/y amino-butyric acid; GABA-T = gamma-aminobutyric acid transaminase; GAT = gamma amino transferase; HIV = human immunodeficiency virus; OCD = obsessive–compulsive disorder.

& Sayyah, 2011). Another RCT compared a mixture of isolated valepotriates (from V. officinalis), diazepam, and placebo over 4 weeks in 36 patients with GAD (Andreatini, Sartori, Seabra, & Leite, 2002). Although there were no significant differences between groups on total symptom scores, valerian appeared to be relatively more effect for psychic symptoms of anxiety (according to HAM-A subscales), whereas diazepam performed better for State–Trait Anxiety Inventory trait anxiety.

Two further studies examined the anxiolytic and sedating effects in a safety context: The first investigated driving performance and sedating effects with placebo comparator, finding no detrimental effects from valerian (Thomas et al., 2016). The second in a similar design (n = 10), assessed subjective sedation and psychomotor/cognitive performance effects of *V. officinalis* extract, diazepam and placebo control, finding decrements only occurred from the BDZ comparator (Gutierrez, Ang-Lee, Walker, & Zacny, 2004).

3.2.10 | *Withania somnifera* (Ashwagandha, Indian ginseng, winter cherry)

Withania somnifera is an alkaloid-rich plant from the nightshade family, traditionally utilised in Indian Ayurvedic medicine for the adaptogenic or "rasayana" properties of the root extract (Mishra, Singh, & Dagenais, 2000). The constituents include glycosides (withanolides), alkaloids (withanine, ashwagandhine, ashwaganidhine, and somniferine), and steroidal triterpenoid lactones, which are related to the ginsenosides, hence the common name "Indian ginseng". Tissue studies investigating GABA-ergic actions of withaferin, withanolides-A, and methanol extract of *W. somnifera* have found these constituents are GABA-A and GABA-ρ1 receptor agonists via *Xenopus* cell lines that also included transplanted Sprague–Dawley rat brain GABA channels (Candelario et al., 2015; Yin, Cho, Park, & Han, 2013). In rodent models using EPM, social interaction and feeding latency/unfamiliar environment paradigms, withanolides have been shown to have equivalent anxiolytic effects to BDZ comparators (lorazepam) in male Charles Foster rats (Bhattacharya, Bhattacharya, Sairam, & Ghosal, 2000; Kaur et al., 2016).

In human studies, a recent systematic review (Pratte, Nanavati, Young, & Morley, 2014) captured all previous studies of *W. somnifera* with anxiety/stress outcomes (e.g., Auddy et al., 2008; Chandrasekhar et al., 2012; Chengappa et al., 2013). This identified five human RCTs with 400 participants (range = 39–130). All five trials reported at least one significant benefit of *W. somnifera* in comparison to control conditions for anxiety and/or stress related outcomes. Doses ranged from 125–1200 mg per day, over 6 to 16 weeks. Since this review, one further eligible study has been conducted: a double-blind RCT in 52 participants with chronic stress, comparing 600 mg of *W. somnifera* extract per day to placebo capsules over 8 weeks (Choudhary, Bhattacharyya, & Joshi, 2017). The study showed significantly greater reductions in stress outcomes from the extract, corresponding with significant decreases in salivary cortisol (a stress/anxiety biomarker).

4 | DISCUSSION

An increasing number of phytomedicines and their bioactive constituents with evidenced GABA mechanisms have been studied in the treatment of generalised anxiety symptoms in addition to clinically diagnosed anxiety disorders. Despite the increased use of phytomedicines, with up to 80% of the world's population using some form of herbal medicine (World Health Organization, 2003), most still lack rigorous investigations of clinical efficacy. This is a common observation from previous reviews of CNS mechanisms of phytomedicines for affective disorders (including GAD) (Ernst, 2006; Lakhan & Vieira, 2010; Liu et al., 2015; Sarris, 2007). In particular, there are a paucity of clinical investigations for phytomedicine use in the treatment of GAD, especially given the epidemiological prevalence of this particular disorder (Australian Bureau of Statistics, 2007; Wittchen et al., 2011).

In the current review, in vitro and in vivo animal studies both revealed GABA-mediated anxiolytic effects from various phytomedicines (as whole extracts and/or isolated constituents). This was demonstrated via a range of mechanisms, which include direct and allosteric interaction with GABA receptor, up-regulation of relevant agonists, and/or interactions with antagonist drugs such as flumazenil or pentylenetetrazole. However, there is also evidence that some of the phytomedicine constituents included in this review modulate ionotropic GABA receptors independently or only partially from flumazenil-sensitive BDZ sites such as that occurring with diazepam (cf. Johnston, 2005). For example, chrysin is known to be a partial agonist at the central BDZ binding site, but apigenin competitively binds to GABA-A BDZ site, also appears to act on both flumazenil-sensitive and -insensitive components, and the effects of Kavain are also not blocked by flumazenil, which suggest a different site of action (cf. Johnston, Hanrahan, Chebib, Duke, & Mewett, 2006; Hanrahan et al., 2011). In most cases, the precise sites of GABA-A receptor action are still to be ascertained, because subtype specificity for phytoconstituents has not been fully investigated in tissue and animal models.

Despite the need for preclinical research of GABA pathway mechanisms and subtype-selectivity behaviours, the next level of preclinical studies involving rodent models have demonstrated these GABA system effects result in reduced anxiety/stress related behaviours in established paradigms such as the EPM, light/dark box, and open fields. However, for some of these plant medicines there is currently a deficit of studies examining their anxiolytic potential for clinically diagnosed anxiety disorders.

Of the phytomedicines reviewed, *P. methysticum* has the largest evidence base for use in the anxiety disorders, with tripartite level of support from in vitro, in vivo, and human clinical studies. The number of positive findings from human studies of *P. methysticum* within randomised, well-controlled trials also supports its use as a treatment for various anxiety disorders and associated symptoms, demonstrating broad clinical utility (Sarris et al., 2011; Sarris et al., 2013b). Thus, *P. methysticum* remains the hallmark anxiolytic phytomedicine (minor concerns over liver toxicity notwithstanding). It is the most strongly evidenced for generalised anxiety, with clinical investigations now examining GABA metabolite changes via neuroimaging, and whether individual gene differences in GABA pathway polymorphisms influence treatment response (Savage et al., 2015).

V. officinalis, *C. asiatica*, *M. recutita*, *G. biloba*, *P. incarnata*, and *W. somnifera* offer the next most extensive evidence for reducing anxiety through GABA-related pathways; all with preclinical studies of GABA-ergic action in rodent animal models, with human studies

showing clinical efficacy of these extracts in people with diagnosed anxiety disorders. *V. officinalis*, particularly the constituent valerenic acid, has been identified as a potent allosteric modulator at GABA-A receptors. Interestingly, *V. officinalis* research involving dose–response effects suggest that mechanisms promote anxiolysis over sedation, with no decrements to cognitive function, even at high doses.

H. lupulus, *S. lateriflora*, and *M. officinalis* also have some promising preclinical evidence, along with human studies of anxiolytic effects in people experiencing stress and anxiety symptoms. However, the lack of evidence showing significant effects in patients with diagnosed anxiety disorders means that the clinical utility of these particular phytomedicines has yet to be shown.

With regard to phytochemical classes of the aforementioned plant medicines; the major GABA-modulating compounds were found to be terpenoids/sesquiterpenoids and saponins classes, such as kavalactones, bilobalide, humulene, valerenic acid, along with the withanones from *W. somnifera*, and the rosmarinic acids from *M. officinalis*. The flavonoids chrysin, apigenin (*M. recutita*), and bacalin (*scutellera*) are the next most evidenced for influencing GABA systems. As they have been extensively studied, flavonoids are well-known positive modulators of GABA-A receptors, operating in the CNS by competitive BDZ site binding and indirectly opening chloride channels; hyperpolarising cell membrane and increasing the threshold for activity, thus exerting an anxiolytic effect (Johnston, 2015; Wasowski & Marder, 2012). It should be noted that despite the broad range of preclinical studies exploring the actions of these phytoconstituents on GABA type A receptors and their subtypes, the mechanisms of anxiolytic effect are complex and warrant a great deal of further research in order to inform the development and evaluation of targeted phytomedicinal compounds for anxiety disorders.

One limitation recognised in this systematic review is the narrow eligibility criteria, which included only those phytomedicines with three levels evidence (in vitro, in vivo, and human studies) for GABA-mediated anxiolytic effects. Although this adds strength to the review by providing focused conclusions on anxiolytic efficacy linked to GABA-based mechanisms of action, the strict criteria forced omission of other phytomedicines, such as *Morinda citrifolia* and *Zizyphus jujuba*, which may also potentially act to reduce anxiety via these pathways (Gao, Wu, & Wang, 2013; Pawlus & Kinghorn, 2007) but lack sufficient investigation for inclusion in this review.

Most notably, *Magnolia officinalis* has preclinical studies showing positive allosteric modulation of GABA-A receptors from its bioactive constituents, resulting in animal behaviours consistent with reduced anxiety (Han et al., 2011). However, to our knowledge no current human studies of the plant for anxiety to date, other than investigations within a herbal combination formula. Although combination formulas of various phytomedicines have demonstrated efficacy for reducing anxiety in humans (e.g., "Relora"; Talbott, Talbott, & Pugh, 2013), the extent to which the observed anxiolytic effects are due to individual components cannot be established. Further, it is recognised that formulas that combine multiple active ingredients may be synergistically more effective—acting through independent but complimentary pathways to confer maximal effects (Bourin, Bougerol, Guitton, & Broutin, 1997).

Future research should aim to extend beyond animal models to investigate neurobiological mechanisms of anxiolytic action in human samples with technologies such as functional magnetic resonance imaging and 1H magnetic resonance spectroscopy to examine metabolite and neurobiological changes resulting from phytomedicine intervention. Specifically, investigating GABA profile level changes in regions of the brain associated with anxiety disorders would further elucidate the neurobiological activity of anxiolytic phytomedicines, especially regarding the interaction with GABA-ergic pathways and the resultant clinical activity. Given the existing evidence for GABA-modulating anxiolytic phytomedicines, continuing to distil the underlying operative pathways and further evaluating these compounds in rigorously designed trials (particularly in anxiety disorder samples) could ultimately lead to novel and evidence-based treatments for people with anxiety disorders; thus serving a major unmet need.

DISCLOSURES

J. S. has received either presentation honoraria, travel support, clinical trial grants, book royalties, or independent consultancy payments from Integria Healthcare & MediHerb (a company sponsoring kava research), Pfizer, Scius Health, Key Pharmaceuticals, Taki Mai (a kava selling company), Bioceuticals & Blackmores, Soho-Flordis, Healthworld, HealthEd, HealthMasters, Elsevier, Chaminade University, International Society for Affective Disorders, Complementary Medicines Australia, Terry White Chemists, ANS, Society for Medicinal Plant and Natural Product Research, Omega-3 Centre, the National Health and Medical Research Council, and CR Roper Fellowship.

ACKNOWLEDGEMENTS

K. S. is supported by Australian Government Research Training Program (RTP) Scholarship. J. F. is supported by a Blackmores Institute Fellowship. J. S. is supported by an NHMRC fellowship (APP1125000).

CONFLICT OF INTEREST

The authors have declared that there is no conflict of interest.

ORCID

Karen Savage http://orcid.org/0000-0002-8075-9222
Joseph Firth http://orcid.org/0000-0002-0618-2752
Con Stough http://orcid.org/0000-0002-3628-3220
Jerome Sarris http://orcid.org/0000-0001-9287-8854

REFERENCES

Abuhamdah, S., Huang, L., Elliott, M. S., Howes, M. J., Ballard, C., Holmes, C., … Chazot, P. L. (2008). Pharmacological profile of an essential oil derived from *Melissa officinalis* with anti-agitation properties: Focus on ligand-gated channels. *The Journal of Pharmacy and Pharmacology*, *60*(3), 377–384.

Ahmadi, M., Khalili, H., Abbasian, L., & Ghaeli, P. (2017). Effect of valerian in preventing neuropsychiatric adverse effects of efavirenz in HIV-positive patients: A pilot randomized, placebo-controlled clinical trial. *Annals of Pharmacotherapy*, *51*(6), 457–464.

Akbarzadeh, M., Dehghani, M., Moshfeghy, Z., Emamghoreishi, M., Tavakoli, P., & Zare, N. (2015). Effect of *Melissa officinalis* capsule on the intensity of premenstrual syndrome symptoms in high school girl students. *Nursing and Midwifery Studies*, *4*(2), e27001.

Akhondzadeh, S., Naghavi, H. R., Vazirian, M., Shayeganpour, A., Rashidi, H., & Khani, M. (2001). Passionflower in the treatment of generalized anxiety: A pilot double-blind randomized controlled trial with oxazepam. *Journal of Clinical Pharmacy and Therapeutics*, *26*(5), 363–367.

Aman, U., Subhan, F., Shahid, M., Akbar, S., Ahmad, N., Ali, G., … Sewell, R. D. E. (2016). *Passiflora incarnata* attenuation of neuropathic allodynia and vulvodynia apropos GABA-ergic and opioidergic antinociceptive and behavioural mechanisms. *BMC Complementary and Alternative Medicine*, *16*, 77.

American Psychiatric Association (2013). *Diagnostic and statistical manual of mental disorders* (5th ed.). Washington, DC: American Psychiatric Association.

Amsterdam, J. D., Li, Y., Soeller, I., Rockwell, K., Mao, J. J., & Shults, J. (2009). A randomized, double-blind, placebo-controlled trial of oral *Matricaria recutita* (chamomile) extract therapy for generalized anxiety disorder. *Journal of Clinical Psychopharmacology*, *29*(4), 378–382.

Amsterdam, J. D., Shults, J., Soeller, I., Mao, J. J., Rockwell, K., & Newberg, A. B. (2012). Chamomile (*Matricaria recutita*) may have antidepressant activity in anxious depressed humans—An exploratory study. *Alternative Therapies in Health and Medicine*, *18*(5), 44–49.

Andrade, C., Aswath, A., Chaturvedi, S. K., Srinivasa, M., & Raguram, R. (2012). A double-blind, placebo-controlled evaluation of the anxiolytic efficacy of an ethanolic extract of Withania somnifera. *Indian Journal of Psychiatry*, *42*(3), 295–301.

Andreatini, R., Sartori, V. A., Seabra, M. L. V., & Leite, J. R. (2002). Effect of valepotriates (valerian extract) in generalized anxiety disorder: A randomized placebo-controlled pilot study. *Phytotherapy Research*, *16*(7), 650–654.

Anukunwithaya, T., Tantisira, M. H., Tantisira, B., & Khemawoot, P. (2016). Pharmacokinetics of a standardized extract of *Centella asiatica* ECa 233 in rats. *Planta Medica*.

Aoshima, H., Takeda, K., Okita, Y., Hossain, S. J., Koda, H., & Kiso, Y. (2006). Effects of beer and hop on ionotropic gamma-aminobutyric acid receptors. *Journal of Agricultural and Food Chemistry*, *54*, 2514–2519.

Appel, K., Rose, T., Fiebich, B., Kammler, T., Hoffmann, C., & Weiss, G. (2011). Modulation of the gamma-aminobutyric acid (GABA) system by *Passiflora incarnata* L. *Phytotherapy Research*, *25*(6), 838–843.

Aslanargun, P., Cuvas, O., Dikmen, B., Aslan, E., & Yuksel, M. U. (2012). *Passiflora incarnata* Linneaus as an anxiolytic before spinal anesthesia. *Journal of Anesthesia*, *26*(1), 39–44.

Auddy, B, Hazra, J., A., M., & Abedon, B. S. G. (2008). A standardized withania somnifera extract significantly reduces stress-related parameters in chronically stressed humans: A double-blind, randomized, placebo-controlled study. *The Journal of the American Nutraceutical Association*, *11*(1), 50–56.

Australian Bureau of Statistics. (2007). National Survey of Mental Health and Wellbeing Australian Bureau of Statistics http://www.abs.gov.au/AUSSTATS/abs@.nsf/DetailsPage/4326.02007?OpenDocument, pp. 1–100.

Avallone, R., Zanoli, P., Corsi, L., Cannazza, G., & Baraldi, M. (1996). Benzodiazepine-like compounds and GABA in flower heads of *Matricaria chamomilla*. *Phytotherapy Research*, (10), S177–S179.

Avallone, R., Zanoli, P., Puia, G., Kleinschnitz, M., Schreier, P., & Baraldi, M. (2000). Pharmacological profile of apigenin, a flavonoid isolated from *Matricaria chamomilla*. *Biochemical Pharmacology*, *59*(11), 1387–1394.

Awad, R., Arnason, J. T., Trudeau, V., Bergeron, C., Budzinski, J. W., Foster, B. C., & Merali, Z. (2003). Phytochemical and biological analysis of skullcap (*Scutellaria lateriflora* L.): a medicinal plant with anxiolytic properties. *Phytomedicine*, *10*(8), 640–649.

Awad, R., Levac, D., Cybulska, P., Merali, Z., Trudeau, V. L., & Arnason, J. T. (2007). Effects of traditionally used anxiolytic botanicals on enzymes of the gamma-aminobutyric acid (GABA) system. *Canadian Journal of Physiology and Pharmacology*, *85*(9), 933–942.

Awad, R., Muhammad, A., Durst, T., Trudeau, V. L., & Arnason, J. T. (2009). Bioassay-guided fractionation of lemon balm (*Melissa officinalis* L.) using an in vitro measure of GABA transaminase activity. *Phytotherapy Research*, *23*(8), 1075–1081.

Baldwin, D., Ajel, K., & Garner, M. (2010). Pharmacological treatment of generalized anxiety disorder. *Current Topics in Behavioral Neurosciences*, (2), 453–467.

Baxter, A. J., Scott, K. M., Vos, T., & Whiteford, H. A. (2013). Global prevalence of anxiety disorders: a systematic review and meta-regression. *Psychological Medicine*, *43*(5), 897–910.

Becker, A., Felgentreff, F., Schroder, H., Meier, B., & Brattstrom, A. (2014). The anxiolytic effects of a valerian extract is based on valerenic acid. *BMC Complementary and Alternative Medicine*, *14*, 267.

Benke, D., Barberis, A., Kopp, S., Altmann, K. H., Schubiger, M., Vogt, K. E., … Möhler, H. (2009). GABA A receptors as in vivo substrate for the anxiolytic action of valerenic acid, a major constituent of valerian root extracts. *Neuropharmacology*, *56*(1), 174–181.

Bhattacharya, S. K., Bhattacharya, A., Sairam, K., & Ghosal, S. (2000). Anxiolytic-antidepressant activity of Withania somnifera glycowithanolides: An experimental study. *Phytomedicine*, *7*(6), 463–469.

Blumenthal, M. (2009). Systematic reviews and meta-analyses support the efficacy of numerous popular herbs and phytomedicines. *Alternative Therapies in Health and Medicine*, *15*(2), 14–15.

Boonen, G., & Haberlein, H. (1998). Influence of genuine kavapyrone enantiomers on the GABA-A binding site. *Planta Medica*, *64*(6), 504–506.

Boonstra, E., de Kleijn, R., Colzato, L. S., Alkemade, A., Forstmann, B. U., & Nieuwenhuis, S. (2015). Neurotransmitters as food supplements: the effects of GABA on brain and behavior. *Frontiers in Psychology*, *6*, 1520.

Bourin, M., Bougerol, T., Guitton, B., & Broutin, E. (1997). A combination of plant extracts in the treatment of outpatients with adjustment disorder with anxious mood: Controlled study versus placebo. *Fundamental & Clinical Pharmacology*, *11*(2), 127–132.

Bradwejn, J., Zhou, Y., Koszycki, D., & Shlik, J. (2000). A double-blind, placebo-controlled study on the effects of Gotu Kola (*Centella asiatica*) on acoustic startle response in healthy subjects. *Journal of Clinical Psychopharmacology*, *20*(6), 680–684.

Brayfield, A. (2014). *Martindale: The complete drug reference* (38th ed.). Pharmaceutical Press: London.

Brinkhaus, B., Lindner, M., Schuppan, D., & Hahn, E. G. (2000). Chemical, pharmacological and clinical profile of the East Asian medical plant *Centella asiatica*. *Phytomedicine*, *7*(5), 427–448.

British Herbal Medicine Association (1996). *British herbal pharmacopoeia*. UK: British Herbal Medicine Association Bristol.

Brock, C., Whitehouse, J., Tewfik, I., & Towell, T. (2014). American skullcap (*Scutellaria lateriflora*): A randomised, double-blind placebo-controlled crossover study of its effects on mood in healthy volunteers. *Phytotherapy Research*, *28*(5), 692–698.

Bruce, S. E., Yonkers, K. A., Otto, M. W., Eisen, J. L., Weisberg, R. B., Pagano, M., … Keller, M. B. (2005). Influence of psychiatric comorbidity on recovery and recurrence in generalized anxiety disorder, social phobia, and panic disorder: A 12-year prospective study. *The American Journal of Psychiatry*, *162*(6), 1179–1187.

Can, O. D., Demir, U., Kiyan, H. T., & Demirci, B. (2012). Psychopharmacological profile of chamomile (*Matricaria recutita* L.) essential oil in mice. *Phytomedicine*, *19*(3–4), 306–310.

Candelario, M., Cuellar, E., Reyes-Ruiz, J. M., Darabedian, N., Feimeng, Z., Miledi, R., … Limon, A. (2015). Direct evidence for GABAergic activity of *Withania somnifera* on mammalian ionotropic GABAA and GABArho receptors. *Journal of Ethnopharmacology*, *171*, 264–272.

Cases, J., Ibarra, A., Feuillere, N., Roller, M., & Sukkar, S. G. (2011). Pilot trial of *Melissa officinalis* L. leaf extract in the treatment of volunteers suffering from mild-to-moderate anxiety disorders and sleep disturbances. *Mediterranean Journal of Nutrition and Metabolism*, *4*(3), 211–218.

Ceremuga, T. E., Johnson, L. A., Adams-Henderson, J. M., McCall, S., & Johnson, D. (2013). Investigation of the anxiolytic effects of xanthohumol, a component of *Humulus lupulus* (Hops), in the male Sprague-Dawley rat. *AANA Journal*, *81*(3), 193–198.

Ceremuga, T. E., Valdivieso, D., Kenner, C., Lucia, A., Lathrop, K., Stailey, O., … Johnson, A. D. (2015). Evaluation of the anxiolytic and antidepressant effects of asiatic acid, a compound from Gotu kola or *Centella asiatica*, in the male Sprague Dawley rat. *AANA Journal*, *83*(2), 91–98.

Chatterjee, T. K., Chakraborty, A., Pathak, M., & Sengupta, G. C. (1992). Effects of plant extract *Centella asiatica* (Linn.) on cold restraint stress ulcer in rats. *Indian Journal of Experimental Biology*, *30*(10), 889–891.

Chandrasekhar, K., Kapoor, J., & Anishetty, S. (2012). A prospective, randomized double-blind, placebo-controlled study of safety and efficacy of a high-concentration full-spectrum extract of ashwagandha root in reducing stress and anxiety in adults. *Indian Journal of Psychological Medicine*, *34*(3), 255–262.

Chengappa, K. N., Bowie, C. R., Schlicht, P. J., Fleet, D., Brar, J. S., & Jindal, R. (2013). Randomized placebo-controlled adjunctive study of an extract of withania somnifera for cognitive dysfunction in bipolar disorder. *Journal of Clinical Psychiatry*, *74*(11), 1076–1083.

Choudhary, D., Bhattacharyya, S., & Joshi, K. (2017). Body weight management in adults under chronic stress through treatment with Ashwagandha root extract: A double-blind, randomized, placebo-controlled trial. *Journal of Evidence-Based Complementary & Alternative Medicine*, *22*(1), 96–106.

Chua, H. C., Christensen, E. T., Hoestgaard-Jensen, K., Hartiadi, L. Y., Ramzan, I., Jensen, A. A., … Chebib, M. (2016). Kavain, the major constituent of the anxiolytic kava extract, potentiates GABAA receptors: Functional characteristics and molecular mechanism. *PLoS One*, *11*(6), e0157700.

Cooley, K., Szczurko, O., Perri, D., Mills, E. J., Bernhardt, B., Zhou, Q., & Seely, D. (2009). Naturopathic care for anxiety: A randomized controlled trial ISRCTN78958974. *PLoS One*, *4*(8), e6628.

Dantas, L. P., de Oliveira-Ribeiro, A., de Almeida-Souza, L. M., & Groppo, F. C. (2017). Effects of *Passiflora incarnata* and *midazolam* for control of anxiety in patients undergoing dental extraction. *Medicina Oral, Patología Oral y Cirugía Bucal*, *22*(1), e95–e101.

Davies, L. P., Drew, C. A., Duffield, P., Johnston, G. A., & Jamieson, D. D. (1992). Kava pyrones and resin: Studies on GABAA, GABAB and benzodiazepine binding sites in rodent brain. *Pharmacology & toxicology*, *71*(2), 120–126.

Dhawan, K., Kumar, S., & Sharma, A. (2001). Anxiolytic activity of aerial and underground parts of *Passiflora incarnata*. *Fitoterapia*, *72*(8), 922–926.

Diwan, P., Karwande, I., & Singh, A. (1991). Anti-anxiety profile of maduk parni (Centella Asiatica) in animals. *Fitoterapia*, *62*(63), 253–257.

Elsas, S. M., Rossi, D. J., Raber, J., White, G., Seeley, C. A., Gregory, W. L., … Soumyanath, A. (2010). *Passiflora incarnata* L. (Passionflower) extracts elicit GABA currents in hippocampal neurons in vitro, and show anxiogenic and anticonvulsant effects in vivo, varying with extraction method. *Phytomedicine*, *17*(12), 940–949.

Ernst, E. (2006). Herbal remedies for anxiety – a systematic review of controlled clinical trials. *Phytomedicine*, *13*, 205–208.

Etkin, A., & Wager, T. D. (2007). Functional neuroimaging of anxiety: a meta-analysis of emotional processing in PTSD, social anxiety disorder, and specific phobia. *The American Journal of Psychiatry*, *164*(10), 1476–1488.

Fehske, C. J., Leuner, K., & Muller, W. E. (2009). Ginkgo biloba extract (EGb761) influences monoaminergic neurotransmission via inhibition of NE uptake, but not MAO activity after chronic treatment. *Pharmacological Research*, *60*(1), 68–73.

Franco, L., Sanchez, C., Bravo, R., Rodriguez, A., Barriga, C., & Juanez, J. C. (2012a). The sedative effects of hops (*Humulus lupulus*), a component of beer, on the activity/rest rhythm. *Acta Physiologica Hungarica*, *99*(2), 133–139.

Franco, L., Sanchez, C., Bravo, R., Rodriguez, A. B., Barriga, C., Romero, E., & Cubero, J. (2012b). The sedative effect of non-alcoholic beer in healthy female nurses. *PLoS One*, *7*(7), e37290.

Galster, J., & Spörl, G. (1979). Entwicklung einer skala zur quantifizierung transitorischer und habitueller Angstzustände. *Neurologie-Psychiatrie*, *5*, 223–226.

Gao, Q. H., Wu, C. S., & Wang, M. (2013). The jujube (Ziziphus jujuba Mill.) fruit: A review of current knowledge of fruit composition and health benefits. *Journal of Agricultural and Food Chemistry*, *61*(14), 3351–3363.

Garrett, K. M., Basmadjian, G., Khan, I. A., Schaneberg, B. T., & Seale, T. W. (2003). Extracts of kava (Piper methysticum) induce acute anxiolytic-like behavioral changes in mice. *Psychopharmacology*, *170*(1), 33–41.

Gleitz, J., Beile, A., & Peters, T. (1995). (+/−)-Kavain inhibits veratridine-activated voltagedependent Na(+)-channels in synaptosomes prepared from rat cerebral cortex. *Neuropharmacology*, *34*(9), 1133–1138.

Gohil, K. J., Patel, J. A., & Gajjar, A. K. (2010). Pharmacological review on *Centella asiatica*: A potential herbal cure-all. *Indian Journal of Pharmaceutical Sciences*, *72*(5), 546–556.

Grundmann, O., Wahling, C., Staiger, C., & Butterweck, V. (2009). Anxiolytic effects of a passion flower (*Passiflora incarnata* L.) extract in the elevated plus maze in mice. *Pharmazie*, *64*(1), 63–64.

Grundmann, O., Wang, J., McGregor, G. P., & Butterweck, V. (2008). Anxiolytic activity of a phytochemically characterized *Passiflora incarnata* extract is mediated via the GABAergic system. *Planta Medica*, *74*(15), 1769–1773.

Gutierrez, S., Ang-Lee, M. K., Walker, D. J., & Zacny, J. P. (2004). Assessing subjective and psychomotor effects of the herbal medication valerian in healthy volunteers. *Pharmacology, Biochemistry, and Behavior*, *78*(1), 57–64.

Guy, W. (1976). *Clinical global impressions*. Rockville, MD: USA: US Department of Heath, Education, and Welfare Public Health Service Alcohol, Drug Abuse, and Mental Health Administration.

Hamid, K., Ng, I., Tallapragada, V. J., Varadi, L., Hibbs, D. E., Hanrahan, J., & Groundwater, P. W. (2016). An investigation of the differential effects of ursane triterpenoids from *Centella asiatica*, and their semisynthetic analogues, on GABAA receptors. *Chemical Biology & Drug Design*, *88*(3), 386–397.

Hamilton, M. (1959). The assessment of anxiety states by rating. *British Journal of Medical Psychology*, *32*, 50–55.

Han, H., Jung, J. K., Han, S. B., Nam, S. Y., Oh, K. W., & Hong, J. T. (2011). Anxiolytic-like effects of 4-O-methylhonokiol isolated from Magnolia officinalis through enhancement of GABAergic transmission and chloride influx. *Journal of Medicinal Food*, *14*(7–8), 724–731.

Hanrahan, J. R., Chebib, M., & Johnston, G. A. R. (2011). Flavonoid modulation of GABA(A) receptors. *British Journal of Pharmacology*, *163*(2), 234–245.

Hattesohl, M., Feistel, B., Sievers, H., Lehnfeld, R., Hegger, M., & Winterhoff, H. (2008). Extracts of *Valeriana officinalis* L. s.l. show anxiolytic and antidepressant effects but neither sedative nor myorelaxant properties. *Phytomedicine*, *15*(1–2), 2–15.

He, X. G., Lin, L. Z., & Lian, L. Z. (1997). Electrospray high performance liquid chromatography-mass spectrometry in phytochemical analysis of kava (*Piper methysticum*) extract. *Planta Medica*, *63*(1), 70–74.

Hoffman, E. J., & Mathew, S. J. (2008). Anxiety disorders: A comprehensive review of pharmacotherapies. *Mount Sinai Journal of Medicine*, *75*(3), 248–262.

Houghton, P. J. (1988). The biological activity of valerian and related plants. *Journal of Ethnopharmacology*, *22*(2), 121–142.

Huang, L., Abuhamdah, S., Howes, M. J., Dixon, C. L., Elliot, M. S., Ballard, C., … Chazot, P.L. (2008). Pharmacological profile of essential oils derived from *Lavandula angustifolia* and *Melissa officinalis* with anti-agitation properties: Focus on ligand-gated channels. *The Journal of Pharmacy and Pharmacology*, *60*(11), 1515–1522.

Huang, S. H., Duke, R. K., Chebib, M., Sasaki, K., Wada, K., & Johnston, G. A. (2003). Bilobalide, a sesquiterpene trilactone from *Ginkgo biloba*, is an antagonist at recombinant alpha1beta2gamma2L GABA(A) receptors. *European Journal of Pharmacology*, *464*(1), 1–8.

Huang, S. H., Duke, R. K., Chebib, M., Sasaki, K., Wada, K., & Johnston, G. A. (2004). Ginkgolides, diterpene trilactones of *Ginkgo biloba*, as antagonists at recombinant alpha1beta2gamma2L GABAA receptors. *European Journal of Pharmacology*, *494*(2–3), 131–138.

Huang, S. H., Lewis, T. M., Lummis, S. C., Thompson, A. J., Chebib, M., Johnston, G. A., & Duke, R. K. (2012). Mixed antagonistic effects of the ginkgolides at recombinant human rho1 GABAC receptors. *Neuropharmacology*, *63*(6), 1127–1139.

Ibarra, A., Feuillere, N., Roller, M., Lesburgere, E., & Beracochea, D. (2010). Effects of chronic administration of *Melissa officinalis* L. extract on anxiety-like reactivity and on circadian and exploratory activities in mice. *Phytomedicine*, *17*(6), 397–403.

Ivic, L., Sands, T. T., Fishkin, N., Nakanishi, K., Kriegstein, A. R., & Stromgaard, K. (2003). Terpene trilactones from *Ginkgo biloba* are antagonists of cortical glycine and GABA(A) receptors. *The Journal of Biological Chemistry*, *278*(49), 49279–49285.

James, J. T., & Dubery, I. A. (2009). Pentacyclic triterpenoids from the medicinal herb, *Centella asiatica* (L.) Urban. *Molecules*, *14*(10), 3922–3941.

Jana, U., Sur, T. K., Maity, L. N., Debnath, P. K., & Bhattacharyya, D. (2010). A clinical study on the management of generalized anxiety disorder with *Centella asiatica*. *Nepal Medical College Journal*, *12*(1), 8–11.

Johnston, G., Chebib, M., Duke, R., Fernandez, S., Hanrahan, J., Hinton, T., & Mewett, K. N. (2009). Herbal products and GABA receptors. In L. Squire (Ed.), *Encyclopedia of Neuroscience* (pp. 1095–1101). London, UK: Academic Press.

Johnston, G. A. (2005). GABA(A) receptor channel pharmacology. *Current Pharmaceutical Design*, *11*(15), 1867–1885.

Johnston, G. A. (2015). Flavonoid nutraceuticals and ionotropic receptors for the inhibitory neurotransmitter GABA. *Neurochemistry International*, *89*, 120–125.

Johnston, G. A., Hanrahan, J. R., Chebib, M., Duke, R. K., & Mewett, K. N. (2006). Modulation of ionotropic GABA receptors by natural products of plant origin. *Advances in Pharmacology*, *54*, 285–316.

Jussofie, A., Schmiz, A., & Hiemke, C. (1994). Kavapyrone enriched extract from *Piper methysticum* as modulator of the GABA binding site in different regions of rat brain. *Psychopharmacology*, *116*(4), 469–474.

Katsiotis, S., Langezaal, C., & Scheffer, J. (1989). Analysis of the volatile compounds from cones of ten *Humulus lupulus* cultivars. *Planta Medica*, *55*(7), 634.

Kaur, T., Singh, H., Mishra, R., Manchanda, S., Gupta, M., Saini, V., … Kaur, G. (2016). *Withania somnifera* as a potential anxiolytic and immunomodulatory agent in acute sleep deprived female Wistar rats. *Molecular and Cellular Biochemistry*, *427*(1–2), 91–101.

Kaviani, N., Tavakoli, M., Tabanmehr, M., & Havaei, R. (2013). The efficacy of *Passiflora incarnata* Linnaeus in reducing dental anxiety in patients undergoing periodontal treatment. *Journal of dentistry (Shiraz, Iran)*, *14*(2), 68–72.

Keefe, J. R., Mao, J. J., Soeller, I., Li, Q. S., & Amsterdam, J. D. (2016). Short-term open-label chamomile (*Matricaria chamomilla* L.) therapy of moderate to severe generalized anxiety disorder. *Phytomedicine*, *23*(14), 1699–1705.

Kennedy, D. O., Little, W., & Scholey, A. B. (2004). Attenuation of laboratory-induced stress in humans after acute administration of *Melissa officinalis* (Lemon Balm). *Psychosomatic Medicine*, *66*(4), 607–613.

Kennedy, D. O., Scholey, A. B., Tildesley, N. T., Perry, E. K., & Wesnes, K. A. (2002). Modulation of mood and cognitive performance following acute administration of *Melissa officinalis* (lemon balm). *Pharmacology, Biochemistry, and Behavior*, *72*(4), 953–964.

Kennedy, D. O., Wake, G., Savelev, S., Tildesley, N. T., Perry, E. K., Wesnes, K. A., & Scholey, A. B. (2003). Modulation of mood and cognitive performance following acute administration of single doses of *Melissa officinalis* (Lemon balm) with human CNS nicotinic and muscarinic receptor-binding properties. *Neuropsychopharmacology*, *28*(10), 1871–1881.

Khom, S., Baburin, I., Timin, E., Hohaus, A., Trauner, G., Kopp, B., & Hering, S. (2007). Valerenic acid potentiates and inhibits GABA(A) receptors: molecular mechanism and subunit specificity. *Neuropharmacology*, *53*(1), 178–187.

Khom, S., Hintersteiner, J., Luger, D., Haider, M., Pototschnig, G., Mihovilovic, M. D., … Hering, S. (2016). Analysis of beta-subunit-dependent GABAA receptor modulation and behavioral effects of valerenic acid derivatives. *The Journal of Pharmacology and Experimental Therapeutics*, *357*(3), 580–590.

Khyati, S., & Anup, T. (2013). A randomized double blind placebo controlled study of Ashwagandha on Generalized Anxiety Disorder. *International Ayurvedic Medical Journal*, *1*(5), 4–7.

Kiewert, C., Kumar, V., Hildmann, O., Rueda, M., Hartmann, J., Naik, R. S., & Klein, J. (2007). Role of GABAergic antagonism in the neuroprotective effects of bilobalide. *Brain Research*, *1128*(1), 70–78.

Kohnen, R., & Oswald, W. D. (1988). The effects of valerian, propranolol, and their combination on activation, performance, and mood of healthy volunteers under social stress conditions. *Pharmacopsychiatry*, *21*(6), 447–448.

Koltermann, A., Hartkorn, A., Koch, E., Furst, R., Vollmar, A. M., & Zahler, S. (2007). *Ginkgo biloba* extract EGb 761 increases endothelial nitric oxide production in vitro and in vivo. *Cellular and Molecular Life Sciences*, *64*(13), 1715–1722.

Krenn, L. (2002). Passion Flower (*Passiflora incarnata* L.)—A reliable herbal sedative. *Wiener Medizinische Wochenschrift (1946)*, *152*(15–16), 404–406.

Kuribara, H., Weintraub, S. T., Yoshihama, T., & Maruyama, Y. (2003). An anxiolytic-like effect of *Ginkgo biloba* extract and its constituent, ginkgolide-A, in mice. *Journal of Natural Products*, *66*(10), 1333–1337.

Lakhan, S. E., & Vieira, K. F. (2010). Nutritional and herbal supplements for anxiety and anxiety-related disorders: Systematic review. *Nutrition Journal*, *9*, 42.

LaPorte, E., Sarris, J., Stough, C., & Scholey, A. (2011). Neurocognitive effects of kava (Piper methysticum): A systematic review. *Human psychopharmacology*, *26*(2), 102–111.

Li, J., Wang, Y. H., Smillie, T. J., & Khan, I. A. (2012). Identification of phenolic compounds from *Scutellaria lateriflora* by liquid chromatography with ultraviolet photodiode array and electrospray ionization tandem mass spectrometry. *Journal of Pharmaceutical and Biomedical Analysis*, *63*, 120–127.

Lister, R. (1987). The use of a plus-maze to measure anxiety in the mouse. *Psychopharmacology*, *92*(2), 180–185.

Liu, L., Liu, C., Wang, Y., Wang, P., Li, Y., & Li, B. (2015). Herbal medicine for anxiety, depression and insomnia. *Current Neuropharmacology*, *13*, 481–493.

Lokanathan, Y., Omar, N., Ahmad Puzi, N. N., Saim, A., & Hj Idrus, R. (2016). Recent updates in neuroprotective and neuroregenerative potential of *Centella asiatica*. *Malaysian Journal of Medical Sciences*, *23*(1), 4–14.

Ma, L., Wang, S., Tai, F., Yuan, G., Wu, R., Liu, X., … Yang, X. (2012). Effects of bilobalide on anxiety, spatial learning, memory and levels of hippocampal glucocorticoid receptors in male Kunming mice. *Phytomedicine*, *20*(1), 89–96.

Magura, E., Kopanitsa, M., Gleitz, J., Peters, T., & Krishtal, O. A. (1997). Kava extract ingredients (+)-methysticin and (+/−)-kavain inhibit voltage-operated Na(+)-channels in rat CA1 hippocampal neurons. *Neuroscience*, *81*(2), 345–351.

Mahita, M., Abuhamdeh, R., Howes, M., Ennaceur, A., Abuhamdah, S., & Chazot, P. (2014). Identification of a novel GABAA receptor channel ligand derived from *Melissa officinalis* and *Lavandula angustifolia* essential oils. *European Journal of Medicinal Plants*, *4*(7).

Mao, J. J., Xie, S. X., Keefe, J. R., Soeller, I., Li, Q. S., & Amsterdam, J. D. (2016). Long-term chamomile (*Matricaria chamomilla* L.) treatment for generalized anxiety disorder: A randomized clinical trial. *Phytomedicine*, *23*(14), 1735–1742.

Martin, E. I., Ressler, K. J., Binder, E., & Nemeroff, C. B. (2010). The neurobiology of anxiety disorders: Brain imaging, genetics, and psychoneuroendocrinology. *Clinics in Laboratory Medicine*, *30*(4), 865–891.

Martin, H. B., McCallum, M., Stofer, W. D., & Eichinger, M. R. (2002). Kavain attenuates vascular contractility through inhibition of calcium channels. *Planta Medica*, *68*(9), 784–789.

Mathews, J., Etheridge, A., Valentine, J., Black, S., Coleman, D., Patel, P., … Burka, L. T. (2005). Pharmacokinetics and disposition of the kavalactone kawain: Interaction with kava extract and kavalactones in vivo and in vitro. *Drug Metabolism and Disposition*, *33*, 1555–1563.

McKay, D. L., & Blumberg, J. B. (2006). A review of the bioactivity and potential health benefits of chamomile tea (*Matricaria recutita* L.). *Phytotherapy Research*, *20*(7), 519–530.

Meier, B. (1995). Passifloræ herba: Pharmazeutische qualitat. *Zeitschrift-fur-Phytotherapie*. 1995B; *16/2*: 90-99, *16*(2), 90–99.

Mills, S., & Bone, K. (2000). *Principles and practice of phytotherapy*. Edinburgh: Churchill Livingstone.

Miroddi, M., Calapai, G., Navarra, M., Minciullo, P. L., & Gangemi, S. (2013). *Passiflora incarnata* L.: ethnopharmacology, clinical application, safety and evaluation of clinical trials. *Journal of Ethnopharmacology*, *150*(3), 791–804.

Mishra, L. C., Singh, B. B., & Dagenais, S. (2000). Scientific basis for the therapeutic use of *Withania somnifera* (ashwagandha): A review. *Alternative Medicine Review*, *5*(4), 334–346.

Mohler, H. (2001). *Pharmacology of GABA and glycine neurotransmission*. Berlin Heidelberg: Springer.

Mori, A., Hasegawa, K., Murasaki, M., Yamaguchi, T., & Ito, S. (1993). Clinical evaluation of passiflamin (*Passiflora* extract) on neurosis - multicenter, double-blind study in comparison with mexazolam. *Rinsho Hyoka [Clinical Evaluation for Drugs]*, *21*(3), 383–440.

Movafegh, A., Alizadeh, R., Hajimohamadi, F., Esfehani, F., & Nejatfar, M. (2008). Preoperative oral *Passiflora incarnata* reduces anxiety in ambulatory surgery patients: A double-blind, placebo-controlled study. *Anesthesia and Analgesia*, *106*(6), 1728–1732.

Muller, C. E., Schumacher, B., Brattstrom, A., Abourashed, E. A., & Koetter, U. (2002). Interactions of valerian extracts and a fixed valerian–hop extract combination with adenosine receptors. *Life Sciences*, *71*, 1939–1949.

Murphy, K., Kubin, Z. J., Shepherd, J. N., & Ettinger, R. H. (2010). *Valeriana officinalis* root extracts have potent anxiolytic effects in laboratory rats. *Phytomedicine*, *17*(8–9), 674–678.

Nassiri-Asl, M., Zamansoltani, F., & Shariati-Rad, S. (2008). Possible role of GABAA-benzodiazepine receptor in anticonvulsant effects of Pasipay in rats. *Zhong xi yi jie he xue bao = Journal of Chinese Integrative Medicine*, *6*(11), 1170–1173.

Ng, C. C., Duke, R. K., Hinton, T., & Johnston, G. A. R. (2017). Effects of bilobalide, ginkgolide B and picrotoxinin on GABAA receptor modulation by structurally diverse positive modulators. *European Journal of Pharmacology*, *806*, 83–90.

Nuss, P. (2015). Anxiety disorders and GABA neurotransmission: A disturbance of modulation. *Neuropsychiatric Disease and Treatment*, *11*, 165–175.

Olsen, R. W., & Sieghart, W. (2008). International Union of Pharmacology. LXX. Subtypes of γ-aminobutyric acidA receptors: Classification on the basis of subunit composition, pharmacology, and function. Update. *Pharmacological Reviews*, *60*(3), 243–260.

Ortiz, J. G., Nieves-Natal, J., & Chavez, P. (1999). Effects of *Valeriana officinalis* extracts on [3H]flunitrazepam binding, synaptosomal [3H] GABA uptake, and hippocampal [3H]GABA release. *Neurochemical Research*, *24*(11), 1373–1378.

Pakseresht, S., Boostani, H., & Sayyah, M. (2011). Extract of valerian root (*Valeriana officinalis* L.) vs. placebo in treatment of obsessive-compulsive disorder: A randomized double-blind study. *Chinese Journal of Integrative Medicine*, *8*, 1553–3840.

Pawlus, A. D., & Kinghorn, D. A. (2007). Review of the ethnobotany, chemistry, biological activity and safety of the botanical dietary supplement *Morinda citrifolia* (noni). *The Journal of Pharmacy and Pharmacology*, *59*(12), 1587–1609.

Pittler, M. H., & Ernst, E. (2003). Kava extract for treating anxiety. *Cochrane Database of Systematic Reviews*, (1), Cd003383.

Pratte, M. A., Nanavati, K. B., Young, V., & Morley, C. P. (2014). An alternative treatment for anxiety: A systematic review of human trial results reported for the ayurvedic herb ashwagandha (*Withania somnifera*). *Journal of Alternative and Complementary Medicine*, *20*(12), 901–908.

Puthenkalam, R., Hieckel, M., Simeone, X., Suwattanasophon, C., Feldbauer, R. V., Ecker, G. F., & Ernst, M. (2016). Structural studies of GABA(A) receptor binding sites: Which experimental structure tells us what? *Frontiers in Molecular Neuroscience*, *9*, 44.

Raines, T., Jones, P., Moe, N., Duncan, R., McCall, S., & Ceremuga, T. E. (2009). Investigation of the anxiolytic effects of luteolin, a lemon balm flavonoid in the male Sprague-Dawley rat. *AANA Journal*, *77*(1), 33–36.

Ramachandran, P. V., & Shekhar, A. (2011). Welcome to 'GABAergic drugs'. *Future Medicinal Chemistry*, *3*(2), 139–140.

Ramharter, J., & Mulzer, J. (2009). Total synthesis of valerenic acid, a potent GABAA receptor modulator. *Organic Letters*, *11*(5), 1151–1153.

Reiss, S., Peterson, R. A., Gursky, D. M., & McNally, R. J. (1986). Anxiety sensitivity, anxiety frequency, and the prediction of fearfulness. *Behaviour Research and Therapy*, (24), 1–8.

Rex, A., Morgenstern, E., & Fink, H. (2002). Anxiolytic-like effects of kava-kava in the elevated plus maze test–A comparison with diazepam. *Progress in Neuro-Psychopharmacology & Biological Psychiatry*, *26*(5), 855–860.

Salah, S. M., & Jager, A. K. (2005). Screening of traditionally used Lebanese herbs for neurological activities. *Journal of Ethnopharmacology*, *97*(1), 145–149.

Sampath, C., Holbik, M., Krenn, L., & Butterweck, V. (2011). Anxiolytic effects of fractions obtained from *Passiflora incarnata* L. in the elevated plus maze in mice. *Phytotherapy Research*, *25*(6), 789–795.

Santos, M. S., Ferreira, F., Cunha, A. P., Carvalho, A. P., Ribeiro, C. F., & Macedo, T. (1994). Synaptosomal GABA release as influenced by valerian root extract–involvement of the GABA carrier. *Archives Internationales de Pharmacodynamie et de Thérapie*, *327*(2), 220–231.

Sarris, J. (2007). Herbal medicines in the treatment of anxiety disorders phytotherapy research

Sarris, J., LaPorte, E., & Schweitzer, I. (2011). Kava: A comprehensive review of efficacy, safety, and psychopharmacology. *The Australian and New Zealand Journal of Psychiatry*, *45*(1), 27–35.

Sarris, J, McIntyre, E, Camfield, D. (2013a). Plant-based medicines for anxiety disorders part 1

Sarris, J, McIntyre, E, Camfield, D. (2013b). Plant-based medicines for anxiety disorders, part 2: A review of clinical studies with supporting preclinical evidence.

Sarris, J., Stough, C., Bousman, C. A., Wahid, Z. T., Murray, G., Teschke, R., … Schweitzer, I. (2013c). Kava in the treatment of generalized anxiety disorder: a double-blind, randomized, placebo-controlled study. *Journal of Clinical Psychopharmacology*, *33*(5), 643–648.

Sasaki, K., Hatta, S., Haga, M., & Ohshika, H. (1999). Effects of bilobalide on gamma-aminobutyric acid levels and glutamic acid decarboxylase in mouse brain. *European Journal of Pharmacology*, *367*(2–3), 165–173.

Sasaki, K., Hatta, S., Wada, K., Ohshika, H., & Haga, M. (2000). Bilobalide prevents reduction of gamma-aminobutyric acid levels and glutamic acid decarboxylase activity induced by 4-O-methylpyridoxine in mouse hippocampus. *Life Sciences*, *67*(6), 709–715.

Savage, K., Stough, C., Byrne, G., Scholey, A., Bousman, C., Murphy, J., … Sarris, J. (2015). Kava for the treatment of generalised anxiety disorder (K-GAD): Study protocol for a randomised controlled trial. *Trials*, *16*, 493.

Schienle, A., Hettema, J., Caceda, R., & Nemeroff, C. (2011). Neurobiology and genetics of generalised anxiety disorder. *Psychiatric Annals*, *41*(2), 113–123.

Schiller, H., Forster, A., Vonhoff, C., Hegger, M., Biller, A., & Winterhoff, H. (2006). Sedating effects of *Humulus lupulus* L. extracts. *Phytomedicine*, *13*(8), 535–541.

Scholey, A., Gibbs, A., Neale, C., Perry, N., Ossoukhova, A., Bilog, V., … Buchwald-Werner, S. (2014). Anti-stress effects of lemon balm-containing foods. *Forum of Nutrition*, *6*(11), 4805–4821.

Shakeri, A., Sahebkar, A., & Javadi, B. (2016). *Melissa officinalis* L.-A review of its traditional uses, phytochemistry and pharmacology. *Journal of Ethnopharmacology*, *188*, 204–228.

Shi, Y., Dong, J., Zhao, J., Tang, L., & Zhang, J. (2014). Herbal insomnia medications that target GABAergic systems: A review of the psychopharmacological evidence. *Current Neuropharmacology*, *12*, 289–302.

Shinomiya, K., Inoue, T., Utsu, Y., Tokunaga, S., Masuoka, T., Ohmori, A., & Kamei, C. (2005). Hypnotic activities of chamomile and *Passiflora* extracts in sleep-disturbed rats. *Biological & Pharmaceutical Bulletin*, *28*(5), 808–810.

Shizhen, L. (1990). *Li Dongyuan's medicinal foods [Shiwu Bencao]*. Beijing: Zhongguo Yiyao Keji Chubansh.

Singh, Y. (1992). Kava: An overview. *Journal of Ethnopharmacology*, *37*(1), 13–45.

Singh, Y. N., & Singh, N. N. (2002). Therapeutic potential of kava in the treatment of anxiety disorders. *CNS Drugs*, *16*(11), 731–743.

Spielberger, C., Gorsuch, R., Lushene, R., Vagg, P., & Jacobs, G. (1983). *Manual for the State-Trait Anxiety Inventory*. Palo Alto, CA: Consulting Psychologists Press.

Srivastava, J. K., Shankar, E., & Gupta, S. (2010). Chamomile: A herbal medicine of the past with bright future. *Molecular Medicine Reports*, *3*(6), 895–901.

Stahl, S., & Moore, B. A. (2013). *Anxiety disorders: A guide for integrating psychopharmacology and psychotherapy*. London: Routledge.

Taiwo, A. E., Leite, F. B., Lucena, G. M., Barros, M., Silveira, D., Silva, M. V., & Ferreira, V. M. (2012). Anxiolytic and antidepressant-like effects of *Melissa officinalis* (lemon balm) extract in rats: Influence of administration and gender. *Indian Journal of Pharmacology*, *44*(2), 189–192.

Talbott, S. M., Talbott, J. A., & Pugh, M. (2013). Effect of *Magnolia officinalis* and *Phellodendron amurense* (Relora(R)) on cortisol and psychological mood state in moderately stressed subjects. *Journal of the International Society of Sports Nutrition*, *10*(1), 37.

Thomas, K., Canedo, J., Perry, P. J., Doroudgar, S., Lopes, I., Chuang, H. M., & Bohnert, K. (2016). Effects of valerian on subjective sedation, field sobriety testing and driving simulator performance. *Accident; Analysis and Prevention*, *92*, 240–244.

Trauner, G., Khom, S., Baburin, I., Benedek, B., Hering, S., & Kopp, B. (2008). Modulation of GABAA receptors by valerian extracts is related to the content of valerenic acid. *Planta Medica*, *74*(1), 19–24.

Tzeng, Y., & Lee, M. (2015). Neuroprotective properties of kavalactones. *Neural Regeneration Research*, *10*(6), 875–877.

Ulbricht, C., Brendler, T., Gruenwald, J., Kligler, B., Keifer, D., Abrams, T. R., … Lafferty, H. J. Natural Standard Research Collaboration. (2005). Lemon balm (*Melissa officinalis* L.): an evidence-based systematic review by the Natural Standard Research Collaboration. *Journal of Herbal Pharmacotherapy*, *5*(4), 71–114.

van Beek, T. A., & Montoro, P. (2009). Chemical analysis and quality control of *Ginkgo biloba* leaves, extracts, and phytopharmaceuticals. *Journal of Chromatography. A*, *1216*(11), 2002–2032.

Viola, H., Wasowski, C., Levi de Stein, M., Wolfman, C., Silveira, R., Dajas, F., … Paladini, A. C. (1995). Apigenin, a component of *Matricaria recutita* flowers, is a central benzodiazepine receptors-ligand with anxiolytic effects. *Planta Medica*, *61*(3), 213–216.

von Zerssen, D. (1976). Klinische Selbstbeurteilungsskalen (KSb-S) aus dem Münchner Psychiatrischen Informationssystem (PSYCHIS München). *Beltz: Weinheim: Beltz*.

Walf, A., & Frye, C. (2007). The use of the elevated plus maze as an assay of anxiety-related behavior in rodents. *Nature Protocols*, *2*(2), 322–328.

Wanasuntronwong, A., Tantisira, M. H., Tantisira, B., & Watanabe, H. (2012). Anxiolytic effects of standardized extract of *Centella asiatica* (ECa 233) after chronic immobilization stress in mice. *Journal of Ethnopharmacology*, *143*(2), 579–585.

Wang, F., Xu, Z., Ren, L., Tsang, S. Y., & Xue, H. (2008). GABA A receptor subtype selectivity underlying selective anxiolytic effect of baicalin. *Neuropharmacology*, *55*(7), 1231–1237.

Wasowski, C., & Marder, M. (2012). Flavonoids as GABAA receptor ligands: The whole story? *Journal of Experimental Pharmacology*, *4*, 9–24.

Wijeweera, P., Arnason, J. T., Koszycki, D., & Merali, Z. (2006). Evaluation of anxiolytic properties of Gotukola--(*Centella asiatica*) extracts and asiaticoside in rat behavioral models. *Phytomedicine*, *13*(9–10), 668–676.

Wittchen, H. U., Jacobi, F., Rehm, J., Gustavsson, A., Svensson, M., Jonsson, B., … Steinhausen, H. C. (2011). The size and burden of mental disorders and other disorders of the brain in Europe 2010. *European Neuropsychopharmacology*, *21*(9), 655–679.

Woelk, H., Arnoldt, K. H., Kieser, M., & Hoerr, R. (2007). Ginkgo biloba special extract EGb 761 in generalized anxiety disorder and adjustment disorder with anxious mood: a randomized, double-blind, placebo-controlled trial. *Journal of Psychiatric Research*, *41*(6), 472–480.

Wohlmuth, H., Penman, K. G., Pearson, T., & Lehmann, R. P. (2010). Pharmacognosy and chemotypes of passionflower (*Passiflora incarnata* L.). *Biological & Pharmaceutical Bulletin*, *33*(6), 1015–1018.

Wolfman, C., Viola, H., Paladini, A., Dajas, F., & Medina, J. H. (1994). Possible anxiolytic effects of chrysin, a central benzodiazepine receptor ligand isolated from Passiflora coerulea. *Pharmacology, Biochemistry, and Behavior*, *47*(1), 1–4.

Wolfson, P., & Hoffmann, D. L. (2003). An investigation into the efficacy of *Scutellaria lateriflora* in healthy volunteers. *Alternative Therapies in Health and Medicine*, *9*(2), 74–78.

World Health Organization. (2003). *Traditional medicine*. World Health Organization: Fact Sheet.

Xu, Z., Wang, F., Tsang, S. Y., Ho, K. H., Zheng, H., Yuen, C. T., … Xue, H. (2006). Anxiolytic-Like Effect of baicalin and its additivity with other anxiolytics. *Planta Medica*, *72*(2), 189–192.

Yin, H., Cho, D. H., Park, S. J., & Han, S. K. (2013). GABA-mimetic actions of *Withania somnifera* on *Substantia gelatinosa* neurons of the trigeminal subnucleus caudalis in mice. *The American Journal of Chinese Medicine*, *41*(5), 1043–1051.

Yoo, D. Y., Choi, J. H., Kim, W., Yoo, K. Y., Lee, C. H., Yoon, Y. S., … Hwang, I. K. (2011). Effects of *Melissa officinalis* L. (lemon balm) extract on neurogenesis associated with serum corticosterone and GABA in the mouse dentate gyrus. *Neurochemical Research*, *36*(2), 250–257.

Zhang, Z., Lian, X. Y., Li, S., & Stringer, J. L. (2009). Characterization of chemical ingredients and anticonvulsant activity of American skullcap (*Scutellaria lateriflora*). *Phytomedicine*, *16*(5), 485–493.

Zick, S. M., Wright, B. D., Sen, A., & Arnedt, J. T. (2011). Preliminary examination of the efficacy and safety of a standardized chamomile extract for chronic primary insomnia: A randomized placebo-controlled pilot study. *BMC Complementary and Alternative Medicine*, *11*, 78–78.

How to cite this article: Savage K, Firth J, Stough C, Sarris J. GABA-modulating phytomedicines for anxiety: A systematic review of preclinical and clinical evidence. *Phytotherapy Research*. 2018;32:3–18. https://doi.org/10.1002/ptr.5940