# Herbal Products and GABA Receptors

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## Introduction

This article profiles ten examples of chemicals derived from herbal preparations that influence ionotropic receptors for the brain's major inhibitory neurotransmitter γ-aminobutyric acid (GABA). Many herbal medicines are used to influence brain function in order to treat anxiety, cognitive disorders, and insomnia. Such disorders are considered likely to involve GABA receptors, particularly ionotropic receptors that are responsible for fast synaptic transmission via ion channels. The evidence of efficacy, or otherwise, in the use of herbal medicines to treat central nervous system (CNS) disorders remains to be established unequivocally in many cases. There is no doubt, however, that these products contain chemicals that in their pure form can be shown to influence brain function in specific ways. The structural diversity of these chemicals provides valuable leads for the development of new therapeutic agents and tools with which to investigate nervous system function.

Herbal medicines are usually complex mixtures of chemicals. The combined actions of these chemicals may contribute to the overall effects of the medicines. Thus, it is important to study interactions among the individual chemicals. With respect to ionotropic GABA receptors, there are examples of synergistic interactions between two chemicals and examples of what has been termed second-order modulation, in which a second modulator enhances the action of a first-order modulator. This may represent a new form of drug action and lead to the identification of new drug targets. Investigating the complex ways in which these chemicals interact could lead to a better understanding of the function of ionotropic GABA receptors.

GABA itself is an important plant constituent, and thus it should not be surprising that plants contain a range of other chemicals that can influence GABA function. In addition, there are examples of plant-derived GABA mediating some form of communication among animals, bacteria, fungi, and plants. Thus, GABA may represent a conserved and ubiquitous biological signaling molecule.

Quality control of herbal medicines is a significant problem. Identification of key active ingredients in herbal medicines and the interactions among them may provide the means to better quality control using functional assays in addition to the currently used chemical assays. Many of the advances in studies of herbal products and GABA receptors have come from ligand binding studies, for example, from the binding of benzodiazepines to brain membranes and, more recently, from functional studies in which GABA receptors of known subunit composition are expressed in cells that do not usually express such receptors (e.g., frog oocytes).

## Ionotropic GABA Receptors

The discovery that the plant alkaloid bicuculline could antagonize certain inhibitory actions of GABA in the CNS provided a vital pharmacological agent with which to probe GABA-mediated inhibition. This observation was a key component of the pharmacological basis for the original classification of GABA receptors into $GABA_A$ and $GABA_B$, with $GABA_A$ receptors being sensitive to bicuculline antagonism and insensitive to the $GABA_B$ agonist baclofen and $GABA_B$ receptors being insensitive to bicuculline and sensitive to baclofen. Subsequently, a third class of GABA receptors was described, $GABA_C$ receptors, that were insensitive to both bicuculline and baclofen. $GABA_A$ and $GABA_C$ receptors are ionotropic receptors, being ligand-gated ion channels, whereas $GABA_B$ receptors are metabotropic G-protein-coupled receptors.

Ionotropic receptors for the inhibitory neurotransmitter GABA are found on most, if not all, neurons in the CNS. They mediate fast neurotransmission via a central ion pore constituted by five surrounding protein subunits that, on activation by GABA, are permeant to chloride ions. They belong to the nicotinicoid superfamily of ligand-gated ion channels (also called cys-loop receptors) that includes nicotinic acetylcholine, strychnine-sensitive glycine, and serotonin 5-$HT_3$ receptors. The family of ionotropic GABA receptors may be divided into two subfamilies, $GABA_A$ and $GABA_C$ receptors, on the basis of their ability to form endogenous functional heteromeric and homomeric receptors, respectively, and of significant differences in their physiological and pharmacological properties.

The heteromeric $GABA_A$ receptors are made up of different protein subunits (e.g., a common makeup involves two α1, two β2, and one γ2 subunits). There are 16 different subunits making up the $GABA_A$ receptor family in human brain: α1–6, β1–3, γ1–3, δ, ε, π, and θ. In addition, there are splice variants of many of these subunits. Although the potential combinatorial diversity of $GABA_A$ receptors is huge,

studies of native $GABA_A$ receptors suggest that there may be fewer than 20 widely occurring $GABA_A$ receptor subtype combinations. There is less diversity in the homomeric $GABA_C$ receptors in that they are made up exclusively of either $\rho 1$, $\rho 2$, or $\rho 3$ subunits, although pseudoheteromeric $GABA_C$ receptors have been described *in vitro*.

Enhancing the action of GABA on $GABA_A$ receptors is a key property of several classes of important therapeutic agents, including the benzodiazepines, barbiturates, and many general anesthetics. With advances in our understanding of the molecular diversity of $GABA_A$ receptors, there is an urgent need for the development of agents acting selectively on subtypes of these receptors.

Agents that enhance the action of GABA on $GABA_A$ receptors are known as positive modulators and are generally considered to involve action at allosteric sites on $GABA_A$ receptors remote from the GABA recognition sites (orthosteric sites). Such allosteric sites are highly valued as targets for the development of subtype-specific drugs because there is generally greater diversity between receptor subtypes in amino acid sequence at allosteric sites than at orthosteric sites. Agents that reduce the action of GABA on $GABA_A$ receptors are known as negative allosteric modulators (once known as inverse agonists because they have the opposite actions to those of the classical benzodiazepines). Agents that block the actions of both positive and negative modulators are known as neutralizing allosteric modulators (e.g., the classical benzodiazepine antagonist flumazenil).

$GABA_C$ receptors are much less widely distributed than $GABA_A$ receptors. There is evidence for functional $GABA_C$ receptors in the retina, spinal cord, superior colliculus, pituitary, and gastrointestinal tract. Given the lower abundance and less widespread distribution of $GABA_C$ receptors compared to $GABA_A$ receptors, $GABA_C$ receptors may be a more selective drug target than $GABA_A$ receptors. The major indications for drugs acting on $GABA_C$ receptors are in the treatment of visual, sleep, and cognitive disorders. Agents acting on $GABA_C$ receptors may be useful for the treatment of myopia. A recent study has linked $GABA_C$ receptors to the possible treatment of Alzheimer's disease by providing evidence that the stimulation of $GABA_C$ receptors has a neuroprotective action against $\beta$-amyloid protein.

## GABA Receptor Antagonists/Negative Modulators

Many of the chemicals currently used to study ionotropic GABA receptors, including the antagonists bicuculline and picrotoxin, are of plant origin. Bicuculline is used extensively to characterize $GABA_A$ receptor function, but has no known therapeutic use. Picrotoxin was used as an analeptic to treat barbiturate overdose and may have therapeutic use in the treatment of vertigo. Other plant-derived antagonists include bilobalide and the ginkgolides from *Ginkgo biloba*, extracts of which are used to treat mild cognitive impairment, and $\alpha$-thujone, one of the active constituents of the psychotropic beverage absinthe prepared from *Artemisia absinthium*.

### Bicuculline

Bicuculline (**Figure 1(a)**) is a phthalide isoquinoline alkaloid first isolated from the plant *Dicentra cucullaria* and subsequently from a variety of *Corydalis, Dicentra,* and *Adlumia* species. The discovery of the GABA antagonist action of bicuculline came from a systematic study of convulsant alkaloids following the discovery of the glycine antagonist action of strychnine. Extensive studies of convulsant alkaloids showed that, whereas many isoquinoline alkaloids are convulsants, most are glycine antagonists, with GABA antagonism being restricted to the phthalide isoquinoline alkaloids that have the 1S,9R configuration (i.e., bicuculline, corlumine and (+)-hydrastine).

Bicuculline is a competitive antagonist of $GABA_A$ receptor activation. There is substantial evidence that bicuculline interacts with the same binding sites as agonists. Point mutations of $\alpha 1$ subunits of $GABA_A$ receptors alter both agonist and competitive antagonist properties, suggesting a close structural association of $\alpha 1$ Phe64 with agonist/antagonist binding sites.

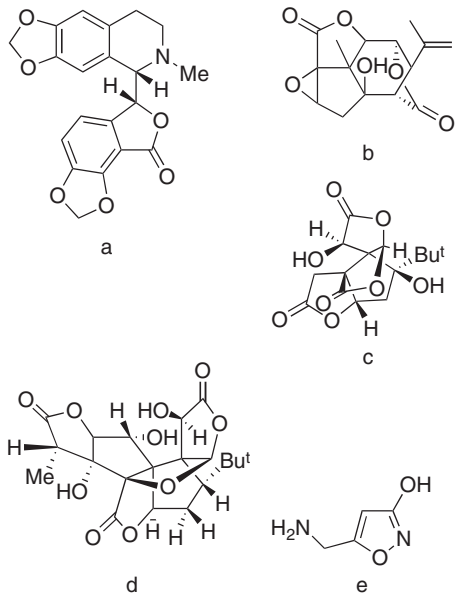


**Figure 1** Plant-derived agonists and antagonists of ionotropic GABA receptors: (a) bicuculline; (b) picrotoxinin; (c) bilobalide; (d) ginkgolide B; (e) muscimol.

Substitution of this Phe by Leu results in a large decrease in the apparent affinity for both GABA and bicuculline.

Quaternary salts of bicuculline, such as bicuculline methochloride and bicuculline methiodide, are easier to use than the ternary salts such as the hydrochloride in that they are more water soluble and more stable; however, the lactone ring can still open in these derivatives, rendering them inactive as GABA antagonists. Unlike bicuculline, the quaternary salts do not pass the blood–brain barrier on systemic administration. The quaternary salts are more potent inhibitors of acetylcholinesterase than the hydrochlorides. Other actions of bicuculline and its derivatives include effects on 5-$HT_3$, nicotinic, and *N*-methyl-D-aspartate (NMDA) receptors, as well as voltage-activated potassium channels.

### Picrotoxin

Picrotoxin is an equimolar mixture of picrotoxinin and picrotin isolated from *Anamirta cocculus* and related poisonous plants of the moonseed family. Picrotoxinin (**Figure 1(b)**) is a relatively potent convulsant and a noncompetitive $GABA_A$ receptor antagonist, whereas picrotin is some 50 times less active than picrotoxinin. Picrotoxinin is one of a number of structurally related convulsants of plant origin, including coriamyrtin and tutin, that act as $GABA_A$ receptor antagonists. Most of the development of picrotoxinin-related compounds has been directed toward the discovery of new insecticides, exploiting differences between insect and mammalian ionotropic GABA receptors.

Picrotoxinin is relatively nonspecific in that it is a potent antagonist at $GABA_A$ and $GABA_C$, moderate at glycine, and weak at $5HT_3$ receptors. Interestingly, picrotin is equipotent with picrotoxinin as an antagonist of glycine receptors, indicating substantial differences in the picrotoxinin binding sites of ionotropic GABA and glycine receptors.

Unlike bicuculline, picrotoxinin can act as a GABA antagonist when administered intracellularly, probably by entering open chloride channels. It is clear that bicuculline and picrotoxinin act at different sites to antagonize GABA. The actual mechanisms by which picrotoxinin blocks ionotropic GABA receptors are complex. Site directed mutagenesis studies show that picrotoxinin acts at sites within the chloride channels of $GABA_A$, $GABA_C$, and glycine receptors.

Because the antagonist action of picrotoxinin is directed toward the GABA-activated chloride channels rather than the GABA recognition sites on ionotropic GABA receptors, picrotoxinin should perhaps be classified more correctly as a negative allosteric modulator, even though it is traditionally described as an antagonist.

### Bilobalide and Ginkgolides

Bilobalide (**Figure 1(c)**), a sesquiterpenoid lactone from *Ginkgo biloba* that bears some structural similarities to picrotoxinin, including a lipophilic side chain and a hydrophilic cage, acts as an antagonist at $GABA_A$ and $GABA_C$ receptors. Like picrotoxinin, bilobalide appears to act at sites in the chloride channels of ionotropic GABA receptors and is thus more appropriately considered as a negative allosteric modulator.

There is, however, a major difference between the effects of bilobalide and picrotoxinin *in vivo*. Whereas picrotoxinin is a convulsant, bilobalide is an anticonvulsant. The lack of convulsant action in an agent that reduces GABA action may be important for the enhancement of cognition. The lack of convulsant action of bilobalide may result from inhibition of glutamate release, thus reducing synaptic excitation and outweighing reductions in synaptic inhibition. Bilobalide has a neuroprotective action in a variety of animal models.

The structurally related ginkgolides, especially ginkgolide B (**Figure 1(d)**), also act as negative modulators at $GABA_A$ receptors. They also inhibit strychnine-sensitive glycine receptors and platelet-activating factor. Bilobalide and the ginkgolides reduce barbiturate-induced sleeping time in mice, an effect perhaps relevant to the clinically observed vigilance-enhancing and antidepressant-like actions of *Ginkgo* extracts.

There are extensive, well-controlled clinical trials on *Ginkgo* extracts, particularly the standardized leaf extract code named EGb761, for the treatment of cognitive and vascular disorders. This extract contains 24% flavonoids and 6% terpenoids (including bilobalide and ginkgolides). It exerts multiple effects including reducing apoptosis, lipid peroxidation, inflammation, and $\beta$-amyloid aggregation, in addition to effects on GABA receptors and glutamate release. The cognition-enhancing effects of *Ginkgo* extracts may be partly mediated by bilobalide acting to enhance hippocampal pyridamidal neuronal excitability.

## GABA Receptor Agonists

Two well-known agonists at ionotropic GABA receptors are found in plants and fungi: GABA itself, which occurs in all plants, and muscimol, a psychoactive agent from the mushroom *Amanita muscaria*.

### GABA

As an important plant constituent, GABA is part of our regular diet. In recent years, chocolate, tea, wine, and yoghurt rich in GABA have been developed and promoted as being beneficial to health. Such products are reported to reduce blood pressure and increase mental alertness. Pure GABA is promoted for body building, acting via human growth hormone to increase muscle and reduce fat.

Because GABA is not considered to pass the blood–brain barrier on systemic administration, dietary GABA may be acting on GABA systems in the periphery. $GABA_A$ and $GABA_B$ receptors have been described on peripheral smooth muscle, cerebral blood vessels, and the sinus node of the heart. Alternate transcripts of the $\varepsilon$ subunit of $GABA_A$ receptors have been found in human and rat heart; the effects of GABA and related compounds on blood pressure may be mediated in part through $GABA_A$ receptors in the heart containing $\varepsilon$ subunits that have not been studied extensively to date. Bicuculline methiodide, a quaternary salt of bicuculline that does not cross the blood–brain barrier, increases mean arterial pressure and heart rate in rats on intravenous administration, thus providing functional evidence for bicuculline-sensitive GABA receptors outside the CNS that influence blood pressure.

Plant-derived GABA is a confounding factor in the bioactivity-guided fractionation of extracts of herbal preparations using ligand binding or functional assays of GABA receptors.

### Muscimol

Muscimol (**Figure 1(e)**) is one of the most widely used agonists in the investigation of ionotropic GABA receptors. It is a more potent agonist at $GABA_C$ receptors than at $GABA_A$ receptors. The agonist action of muscimol at $GABA_C$ receptors is not blocked by bicuculline but is sensitive to picrotoxinin.

Muscimol is a conformationally restricted analog of GABA in which a hydroxyisoxazole moiety replaces the carboxyl group of GABA. The 3-hydroxyisoxazole is recognized as a carboxyl group equivalent by $GABA_A$ and $GABA_C$ receptors but not by $GABA_B$ receptors. The neuronal GABA uptake system recognizes the 3-hydroxyisoxazole moiety, in that muscimol is a weak inhibitor of GABA uptake but is neither an inhibitor of nor a substrate for GABA aminotransferase, indicating that this enzyme does not interact with the 3-hydroxyisoxazole moiety.

Muscimol became a prototype substance for the design and development of a range of isoxazoles with varying activities on GABA systems. THIP (Gaboxadol) is a conformationally restricted analog of muscimol with analgesic and sleep-promoting properties. It was investigated for the treatment of insomnia, but was recently withdrawn from phase III clinical trials due to efficacy and side-effect problems. THIP shows selectivity for $GABA_A$ receptors that contain a $\delta$ subunit, at which it is a more efficacious agonist than GABA. At similar concentrations, THIP is a $GABA_C$ receptor antagonist.

## GABA Receptor Modulators

Flavonoids are responsible for many of the brilliant colors of fruits and vegetables and are important constituents of red wine, green tea, and many herbal preparations. More than 5000 different flavonoids have been identified. It has been estimated that the average daily intake of flavonoids is 1–2 g. Their antioxidant properties are regarded as important in our diet. They have a wide variety of biological activities and are being studied intensively as anticancer agents and for their effects on the vascular system.

Flavonoids have a range of activities on $GABA_A$ receptors and have been described as a new family of benzodiazepine receptor ligands. They were first linked to $GABA_A$ receptors when three isoflavans isolated from bovine urine were shown to inhibit diazepam binding to brain membranes. These isoflavans were most probably derived from plant sources in the bovine diet. Subsequently, many flavonoids directly isolated from plants were shown to influence benzodiazepine binding. A low-affinity benzodiazepine site has emerged as a possible target for the modulatory action of some flavonoids. This site is insensitive to the classical benzodiazepine antagonist flumazenil and has been described on a wide range of $GABA_A$ receptors, including those made up of only $\alpha 1\beta 2$ subunits. Flumazenil-insensitive effects of flavonoids on $GABA_A$ receptors have been extensively described.

Terpenoids are widespread in plants, especially in what are known as essential oils that can be extracted from plants. They have a wide range of uses from perfume constituents to paint thinners. Terpenoids are oxygenated products formally derived from C5 units and are classified by the number of C5 units in their structure. The sesquiterpenoid lactone picrotoxinin is the most widely used terpenoid in studies on ionotropic GABA receptors. However, a number of other terpenoids are of interest for their actions as positive modulators of $GABA_A$ receptors.

Another important class of herbal products that act as positive modulators of $GABA_A$ receptor function are $\alpha$-pyrones, especially the kava lactones found in kava-kava (*Piper methysticum*). There are many random controlled trials of extracts of kava-kava that show a reduction in anxiety relative to placebo.

### Apigenin

Apigenin (Figure 2(a)) is a common flavonoid found in a range of plants, including chamomile (*Matricaria recutita*). The traditional use of chamomile tea as a treatment for insomnia and anxiety led to investigations of its active constituents, including apigenin. Apigenin was found to have anxiolytic properties, and it competitively inhibited the binding of flunitrazepam to brain membranes without influencing the binding of muscimol to $GABA_A$ receptors.

Recent studies on human recombinant receptors in oocytes have shown that apigenin inhibited the activation of $GABA_A$ receptors in a flumazenil-insensitive manner and had a similar effect on $GABA_C$ receptors. Other studies on recombinant $GABA_A$ receptors also found an inhibitory effect of apigenin on GABA responses and, in addition, an enhancement of the diazepam-induced positive allosteric modulation of GABA responses by lower concentrations of apigenin (1 μM), described as a second-order modulation by apigenin of the first-order modulation by diazepam.

The flumazenil-sensitive anxiolytic effects of apigenin may be the result of apigenin enhancing a subthreshold effect of an endogenous benzodiazepine system. Evidence for physiologically relevant endozepines has come from the discovery of a mutant $GABA_A$ receptor in childhood absence epilepsy and febrile seizures that has diminished sensitivity to benzodiazepines with no other apparent alteration in function.

Overall, it seems that the effects of apigenin on $GABA_A$ receptors are complex and involve both flumazenil-sensitive and flumazenil-insensitive components, and that other receptors could be involved in the behavioral effects of apigenin. Like most flavonoids, apigenin is known to have a wide variety of biological actions, including effects on adenosine receptors and progestational activity. Of particular interest are the findings that apigenin, at concentrations that inhibit $GABA_A$ and $GABA_C$ receptors, also inhibits NMDA receptors. Such an action could contribute to the flumazenil-insensitive sedative actions of apigenin.

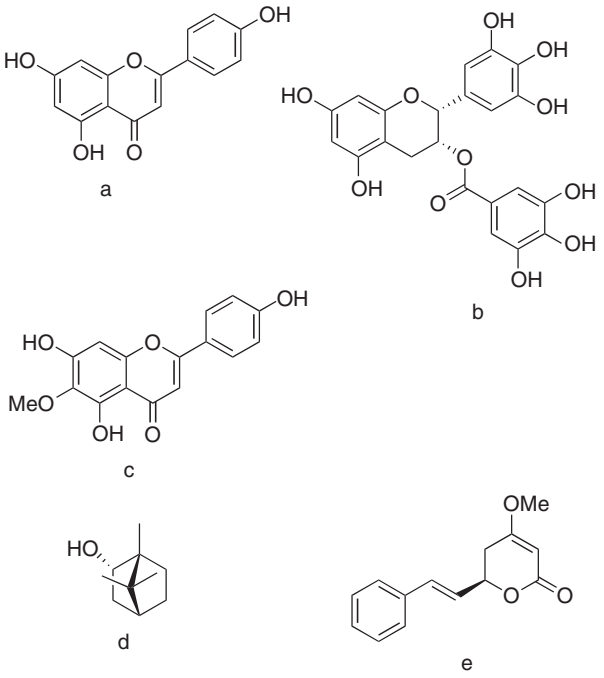


**Figure 2** Plant-derived positive modulators of the activation of $GABA_A$ receptors by GABA: (a) apigenin; (b) epigallocatechin gallate; (c) hispidulin; (d) (+)-borneol; (e) kawain.

### Epigallocatechin Gallate

Green tea polyphenols are being considered as therapeutic agents in well-controlled epidemiological studies aimed to alter brain-aging processes and to serve as possible neuroprotective agents in progressive neurodegenerative disorders such as Parkinson's and Alzheimer's diseases.

Epigallocatechin gallate (Figure 2(b)) (EGCG) is the major polyphenol in green tea (*Camellia sinensis*). Studies on $\alpha 1\beta 2\gamma 2L$ $GABA_A$ receptors showed that EGCG at low concentrations (0.1 μM) has a potent second-order modulatory action on the first-order modulation by diazepam but inhibits the action of GABA at higher concentrations (>1 μM). EGCG was an order of magnitude more potent than apigenin in acting as a second-order modulator. In addition, it has been found that EGCG, at concentrations that have no influence on the activation of $GABA_A$ receptors by GABA, was able to reverse the negative modulation of such receptors by methyl $\beta$-carboline. This indicates that EGCG may act as a second-order modulator with respect to the first-order modulation by both positive and negative modulators that act on benzodiazepines sites on $GABA_A$ receptors.

The novel second-order modulation by EGCG and apigenin of the maximum first-order modulatory action of diazepam of the activation by GABA of $GABA_A$ receptors observed in these studies may result from altering the coupling of the benzodiazepine allosteric sites with the orthosteric GABA sites on $GABA_A$ receptors. There is evidence from binding studies that the nexus between the benzodiazepine and GABA sites on $GABA_A$ receptors is complex and involves other factors such as phospholipids. EGCG and apigenin may serve as lead compounds for the development of more selective agents for the second-order modulation of benzodiazepine enhancement of the action of GABA on $GABA_A$ receptors.

There is much interest in the anticancer and antitumor properties of EGCG associated with the consumption of green tea. EGCG has anticancer effects on ovarian carcinoma cell lines, is a selective inhibitor of COX-2 expression, and induces apoptosis in monocytes. Little is known about the CNS actions of this flavan. However, it is found in the brain after gastric administration to mice and is neuroprotective in rats on intraperitoneal (IP) injection after focal ischemia and in a mouse model of Parkinson's disease. This neuroprotective action may be associated with its antioxidant properties, but enhancement of $GABA_A$-mediated synaptic inhibition could also contribute. In addition, EGCG is known to reduce glutamate-induced cytotoxicity via intracellular $Ca^{2+}$ ion modulation, suggesting that other neurotransmitter systems may be involved.

### Hispidulin and Related Flavonoids

Hispidulin (Figure 2(c)) (the 6-methoxy derivative of apigenin) was isolated together with apigenin from *Salvia officinalis* (sage) recently using a benzodiazepine binding assay-guided fractionation. Hispidulin was some 30 times more potent than apigenin in displacing flumazenil binding. Preparations of sage have been used in herbal medicine to assist memory, and an extract of *Salvia lavandulaefolia* (Spanish sage) has been shown to enhance memory in healthy young volunteers. Unlike apigenin, hispidulin has been shown to act as a positive allosteric modulator of $GABA_A$ receptors. The positive modulatory action of 10 μM hispidulin at $\alpha1\beta2\gamma2S$ receptors was reduced from 47% to 17% by flumazenil, indicating that sites other than classical flumazenil-sensitive benzodiazepine sites were involved in the action of hispidulin. Because hispidulin did not influence the action of GABA on $\alpha1\beta2$ $GABA_A$ receptors, this indicates that it did not interact with low-affinity flumazenil-insensitive benzodiazepine sites. Hispidulin was shown to have an anticonvulsant action in seizure-prone Mongolian gerbils and to pass the blood–brain barrier.

Flavonoids structurally related to hispidulin that influence benzodiazepine binding have been isolated from *Scutellaria baicalensis*, an important herb in traditional Chinese medicine. Oroxylin A (5,7-dihydroxy-6-methoxyflavone, i.e., hispidulin lacking the 4′-hydroxy group) inhibits flunitrazepam binding at 1 μM, and on oral administration acts as a neutralizing modulator blocking the anxiolytic, myorelaxant, and motor incoordination effects but not the sedative and anticonvulsant effects elicited by diazepam. 6-Methylapigenin isolated from *Valeriana wallichii,* a known sedative herb, influences benzodiazepine binding at 0.5 μM in manner suggesting it may be a positive modulator of $GABA_A$ receptors. 6-Methylapigenin has anxiolytic properties, and it can act synergistically to potentiate the sleep-enhancing properties of hespiridin, a flavanone glycoside from *Valeriana.*

Thus, flavones substituted in the 6 position with a methoxy or methyl substituent have interesting effects on $GABA_A$ receptor function and may contribute to the properties of some herbal preparations. Several flavonoid glycosides, including goodyerin, linarin, and hesperidin, are also being studied as sedative and anticonvulsant agents likely to interact with $GABA_A$ receptors.

### (+)-Borneol

(+)-Borneol (Figure 2(d)) is a bicyclic monoterpenoid found in high concentrations in extracts of *Valeriana officinalis* that are widely used to reduce the latency of sleep onset and to increase the depth of sleep and the perception of well-being. Extracts of *Valeriana* are known to contain a large number of constituents, including flavonoids and terpenoids, many of which are considered to be active at $GABA_A$ receptors.

(+)-Borneol was shown to be a flumazenil-insensitive positive allosteric modulator of human recombinant $\alpha1\beta2\gamma2L$ $GABA_A$ receptors of low potency (median effective concentration ($EC_{50}$) 250 μM) but very high efficacy, producing a ten-fold enhancement of the action of 10 μM GABA at a concentration of 450 μM. (−)-Borneol showed similar positive modulatory properties to (+)-borneol, whereas isoborneol, (−)-bornyl acetate, and camphor (a known convulsant) were much less active. The relatively rigid cage structure of these bicyclic monoterpenes and their high efficacy may aid in a greater understanding of the molecular aspects of positive modulation.

### Kava Lactones

Kava lactones (α-pyrones) have been recognized as the active constituents responsible for the reported anxiolytic and sedative effects of kava-kava. The six major kava lactones are kawain (Figure 2(e)), dihydrokawain, methysticin, dihydromethysticin, yangonin, and demethoxyyangonin, and they occur in varying proportions in different cultivars.

There is evidence to indicate that kava lactones may mediate their anxiolytic and sedative effects via positive modulation of $GABA_A$ receptor function acting at sites different from the classical benzodiazepine binding sites. Individual kava lactones have been shown to enhance the binding of bicuculline methochloride and muscimol to brain membranes without

influencing the binding of diazepam or flunitrazepam, indicating that they interact with the agonist/competitive antagonist binding sites on $GABA_A$ receptors. The enhancement of muscimol binding showed regional selectivity to be highest in the hippocampus, amygdala, and medulla, with minimal effect on the cerebellum and frontal cortex, suggesting that the kava lactones may show subtype specificity for particular $GABA_A$ receptors. Functional studies of recombinant $\alpha1\beta2\gamma2L$ $GABA_A$ receptors have shown that kava lactones do act as positive modulators, but such studies need to be extended to investigate different subunit combinations.

*See also:* Anxiety Disorders; GABA Synthesis and Metabolism; GABA-A Receptors: Developmental Roles; GABA-A Receptor Synaptic Functions; GABA-A Receptors: Molecular Biology, Cell Biology and Pharmacology; GABA-A Receptors and Disease; Gamma-Aminobutyric Acid (GABA); Glial Glutamate and GABA Metabolism.

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