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The relevance of neuronal substrates of defense in the midbrain tectum to anxiety and stress: empirical and conceptual considerations

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Abstract

The medial hypothalamus, amygdala, and dorsal periaqueductal gray constitute the main neural substrates for the integration of aversive states in the brain. More recently, some regions of the mesencephalon, such as the superior and inferior colliculi have also been proposed as part of this system. In fact, fear-like behaviors often result when these sites are electrically or chemically stimulated. Both the behavioral and autonomic consequences of electrical stimulation of the mesencephalic tectum have been shown to be attenuated by minor tranquilizers, probably through enhancement of γ-aminobutyric acid (GABA)-mediated neurotransmission, which exerts a tonic inhibitory control on the neural circuits responsible for the so-called defense behavior repertoire. Besides GABA, also 5-hydroxy tryptamine serotonin (5-HT), opioids, neuropeptides, histaminergic and excitatory amino acids have all been implicated in the regulation of anxiety-related behaviors induced by stimulation of midbrain tectum. Efforts have been made to characterize how these neurotransmitters interact with each other in the organization of these reactions to aversive stimulation. In this review, we summarize the evidence linking the brain's defense response systems to the concept of fear–anxiety. Furthermore, a case is made for the consideration of the relevance of this body of data to the search for the physiological underpinnings of depression and its consequences.
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1. Introduction

A large body of literature has accrued over the last half century on the anatomy, physiology and pharmacology of areas or projection systems in the central nervous system, which, when stimulated electrically or chemically result in physiological and behavioral changes indicative of states of agony or displeasure as expressed by vegetative nervous system indicators, attempts to escape, and displays of fear and aggressive behavior. It is generally assumed that brain sites which mediate such behavioral changes integrate information of noxious valence into appropriate adaptive emotional and behavioral reactions and actions ranging from flight to fight, including fear-related behaviors. The search for the subcortical organization of fear through the use of electrical stimulation of the brain dates back to the work of Hess and Brügger (1943), showing that electrical stimulation of the hypothalamus of cats, results in defense-like behaviors accompanied by autonomic responses. Soon thereafter, a seminal work by Bard and Match (1951) showed that mesencephalic structures also have inherent neural circuits relevant to fear-related reactions. Other studies confirmed these findings and also added new structures to this list, since similar pattern of responses appeared upon electrical stimulation of the amygdala and the dorsal periaqueductal gray (Hunsperger, 1956; De Molina and Hunsperger, 1962; Olds and Olds, 1963).

The medial hypothalamus, amygdala and dorsal periaqueductal gray have been traditionally grouped together as a "brain aversion system". More recently, a continuous strip of midbrain structures composed of superior and inferior colliculi have also been proposed as parts of this "system". In this review, we will focus on the neural substrates of defensive behavior in the midbrain tectum (dorsal periaqueductal gray, superior and inferior colliculi), and their rele-

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vance for understanding fear and anxiety. The proposed link between defensive behavior, fear and anxiety is consistent with many behavioral, electrophysiological and immunohistochemical studies showing expressive activation of this region by threatening stimuli or conditions and also with the demonstrations of functional reciprocal connections of these midbrain structures with the medial hypothalamus and amygdala, two structures traditionally known to be involved with fear and anxiety (LeDoux et al., 1986; Graeff, 1990, 1997). Furthermore, many manipulations of the midbrain tectum structures which elicit the ''defensive'' behavior repertoire also induce fear and anxiety.

2. Electrical stimulation of the midbrain tectum

It has been shown that a gradual increase in the intensity of electrical stimulation of the dorsal periaqueductal gray, deep layers of the superior colliculus or of the inferior colliculus of rats induces, in a progressive manner, responses such as arousal, freezing and escape behaviors (Coimbra et al., 1992, 1996; Coimbra and Brandão, 1997; Melo et al., 1992; Melo and Brandão, 1995). That these responses are due to the aversive nature of the stimulation is attested by the fact that animals readily learn operant behaviors to interrupt or to turn this stimulation off (Brandão et al., 1982, 1997; Melo et al., 1992). In general, these responses are accompanied by analgesia (Fanselow, 1991; Coimbra et al., 1992, 1997) and changes in the mean arterial blood pressure, heart rate and respiration (Hilton and Redfern, 1986; Brandão et al., 1988; Carrive, 1991). Thus, these midbrain structures together with the periventricular gray substance of the mesencephalon diencephalic areas and the amygdala appear to be constituents of a neuronal system or systems that control defensive behavior in the brain (Graeff, 1990; Brandão et al., 1994, 1999).

The deep layers of the superior colliculus have been implicated in the control of orienting movements of the eyes and head, as well as in the mediation of defensive movements together with cardiovascular changes that would be appropriate for a sudden emergency such as the appearance of a predator, or of an object on collision course (Dean et al., 1989), and to tactile stimulation of the vibrassae, which induces reflexive biting under various pharmacological challenges (Welzl et al., 1984). Lesions of this area reduce defensive responding to threatening visual stimuli, for example, to unexpected overhead movement or to an approaching human (Blanchard et al., 1981). The periaqueductal gray matter participates in the expressions of at least five major brain functions, including pain processing and modulation, vocalization, autonomic regulation, fear and anxiety and lordosis (Behbehani, 1995). It has been proposed that neurons in the inferior colliculus are filters for sounds that require immediate action, such as certain sounds made by prey, predators and conspecifics (Casseday and Covey, 1996). Recent studies using c-*fos* immunoreactivity bring further support to the notion that amygdala, medial hypothalamus, dorsal periaqueductal gray, deep layers of superior colliculus and inferior colliculus constitute a ''brain aversion system'', by showing that these areas are labeled following either electrical or chemical stimulation of the dorsal periaqueductal gray or exposure of the animals to aversive environmental stimulation (Silveira et al., 1993, 1995, 2001; Sandner et al., 1993).

It must be emphasized that there exist some differences in responses depending on the midbrain structures stimulated. For example, turning is the predominant response following stimulation of the superior colliculus. Furthermore, the flight behavior induced by stimulation of the inferior colliculus is not as explosive as that observed after dorsal periaqueductal gray stimulation (Dean et al., 1989; Brandão et al., 1994, 1999). It has been proposed that γ-aminobutyric acid (GABA)-mediated mechanisms are involved in the gating of distinct sensory information of aversive nature depending on the midbrain structure which is activated; visual in the superior colliculus, tactile-nociceptive in the dorsal periaqueductal gray and auditory in the inferior colliculus, a region traditionally known to process high-pitched sounds (Rose et al., 1963; Merzenick and Reid, 1974; Schmitt et al., 1984, 1986; Brandão et al., 1983, 1990).

There are many reports showing that defensive behavior patterns are organized in a hierarchical series of responses (Blanchard et al., 1989; Fanselow, 1991). In general, animals show an increased arousal and attention to negative stimuli. In the face of potential threat, there is a conflict between approach and risk assessment while immediate threat leads to escape behavior. Overall, the organism has a tendency to evaluate and therefore approach novel and thus potentially dangerous stimuli and this evaluation is critical for defining the direction of approach to safety cues and retreat from aversive cues. These processes seem to involve a combination of conditioned and innate responses to proximal and distal aversive stimulation, such that aversive cues detected at a distance can serve to activate a fear-related system which guides the organism from danger. Interestingly, somehow the stimulation of the midbrain tectum may represent an animal model for all these situations, because a gradual increase in the intensity of the electrical stimulation of the midbrain tectum structures initially produces alertness, then freezing, and finally escape reactions. Furthermore, electrical stimulation of the dorsal periaqueductal gray and inferior colliculus has been consistently shown to induce aversive effects, because it elicits defensive behaviors, sustains learned operant escape responses and also supports learning of conditioned emotional responses (Jenck et al., 1983, 1995; Brandão et al., 1997). Recently, we have shown that the neural circuits of the dorsal periaqueductal gray underlying freezing behavior are unique and do not encompass the neural substrates for conditioned fear in the ventral regions of this structure (Vianna et al., 2001). Such elicited freezing behavior is, in general, accompanied by increases in mean arterial blood pressure, heart rate and respiration, and by

analgesia. Thus, the neural substrate of dorsal periaqueductal gray stimulation-induced freezing may process unconditioned fear responses to impending danger, which have been implicated in panic disorder. All evidence points to multicomponent processes underlying the defense reaction, which is understandable given the complexity of defensive behavior expressed by animals upon facing predators and other threatening stimuli.

Although the midbrain tectum seems to have local circuits for the generation and elaboration of defense reactions, higher brain structures are necessarily enlisted in the control of more complex fear-related behaviors. For example, it is well known that the dorsal periaqueductal gray and amygdala have reciprocal anatomical connections (Bandler and Shipley, 1994). Examination of functional connection of the inferior colliculus with the amygdala—an important filter for sensorial information of aversive nature—has shown that lesion of the basolateral nucleus enhances, whereas lesion of the central nucleus of the amygdala reduces the aversiveness of electrical stimulation of the inferior colliculus (Maisonnette et al., 1996). Lesion of the bed nucleus of the stria terminalis, part of the ''extended amygdala'', attenuates sound induced freezing behavior (Schulz and Canbeyli, 2000) and enhanced learned despair (Schulz and Canbeyli, 1999). The ascending anatomical connections between amygdala and inferior colliculus seem to indirectly relay the medial geniculate nucleus of the thalamus (LeDoux et al., 1986; Iwata et al., 1987).

3. Chemical stimulation of the midbrain tectum

Circumventing the main problem associated with the electrical stimulation procedure, which excites both neuronal cell bodies and axons of passage, intracerebral microinjections of excitatory amino acids selectively depolarize somatodendrites. Among their many important physiological functions, the excitatory amino acids also play a significant role in the expression of defensive behavior. Considerable evidence suggests that excitatory amino acids have a critical role in integrating somatic and autonomic reactions characteristic of defensive behaviors. For instance, injection of excitatory amino acids into the dorsolateral periaqueductal gray elicited the complete pattern of defensive behavior (Bandler, 1982; Bandler et al., 1986; Krieger and Graeff, 1985; Bandler and Carrive, 1988). Injections of these excitatory amino acids also mimic the effects of electrical stimulation when injected into the deep layers of superior colliculus of rats (Dean et al., 1988; Bandler et al., 1985). Injections of glutamate into the midbrain of the squirrel monkey elicited vocalizations, many of which were also characteristic of defense (Jurgens and Richter, 1986). Based on responses to injection of excitatory amino acids into the dorsal periaqueductal gray, it was possible to depict a columnar organization for this region (Carrive, 1991; Bandler and Shipley, 1994). These studies have indicated that the periaqueductal gray is organized in four columns: the dorsomedial, dorsolateral, lateral and ventromedial. Stimulation of the dorsomedial and dorsolateral columns produces an increase in blood pressure and blood flow to muscle, a decrease in blood flow to skin, and escape behavior, whereas stimulation of the lateral and ventromedial columns produces hypotension and freezing behavior (Carrive, 1991). In this review, we use the abbreviation dorsal periaqueductal gray to designate the dorsomedial and dorsolateral columns of the periaqueductal gray.

The same mechanisms are also apparent in the inferior colliculus, because injection of *N*-methyl D-aspartate (NMDA) produced a graded and dose-dependent pattern of defensive behavior very similar to that seen after gradual increases in the intensity of electrical current applied to this structure (Cardoso et al., 1994). Initially, the animals are alert; with intermediate doses, there is moderate immobility, which progresses to behavioral activation, and to jumping at high doses. These effects were antagonized by previous injection of 2-amino-7-phosphonoheptanoate (AP7), an NMDA receptor antagonist, into the inferior colliculus. This pattern of reactions suggests that the type of defensive behavior varies quantitatively and qualitatively with the increase in aversive stimulation (Brandão et al., 1994) as occurs with the prey's perception of the risk of predatory imminence (Fanselow, 1991). Congruent with this view are findings showing that injection of glutamate into the central nucleus of the inferior colliculus, which presumedly activates mainly AMPA/kainate receptors, induced freezing responses (Pandossio and Brandão, 1999). Behavioral activation was not obtained even with high doses of glutamate due, perhaps, to the quick uptake of this excitatory amino acid into both neurons and glia. More intense responses, such as escape responses, were only obtained with injections of NMDA into the inferior colliculus. Perhaps the activation of fast-acting AMPA/kainate receptors in this midbrain region is able to trigger the initial steps of the defense reaction without eliciting the explosive motor behavior, which is usually seen following the activation of NMDA receptors. In this context, it is worth mentioning that glutamate is an effective agonist at AMPA/kainate receptors, whereas aspartate is a poor agonist at these receptors (Watkins et al., 1990). In fact, while glutamic acid diethyl ester (GDEE), a specific antagonist of AMPA/kainate receptors, blocked the freezing response caused by glutamate microinjection into the inferior colliculus, AP7 did not interfere with the occurrence of this response. Therefore, probably in the inferior colliculus, the defense reaction starts with freezing behavior engendered by activation of AMPA/kainate receptors, which results in rapid excitation related to the opening of Na^+ and K^+ channels (Agarawal and Evans, 1986). This effect could be secondarily counteracted by the activation of NMDA receptors, which results in escape behavior that displaces the expression of freezing. The effects mediated by NMDA receptors should be longer acting, as they are the result of long-lasting changes in gene

Table 1
Similarities between dorsal periaqueductal gray-induced defensive reactions in laboratory animals and in man

	"Emotional" reactions	Autonomic reactions	References
Dorsal periaqueductal gray stimulation in laboratory animals	alertness; freezing; escape	Tachycardia; arterial hypertension; hyperventilation; piloerection	Schenberg et al., 1983; Graeff, 1990, 1997; Brandão et al., 1994, 1999; Lovick, 2000
Dorsal periaqueductal gray stimulation in man	Sudden anxiety; reports of feeling terrified	Tachycardia; hyperventilation; blushing; sweating; piloerection	Nashold et al., 1969; Amano et al., 1978
Panic attacks	Sudden apprehension and intense fear; fear of dying; fear of going crazy	Tachycardia; arterial hypertension; dyspnea; hyperventilation; sweating; flushes	American Psychiatric Association, 1994 (DSM-IV)

expression. Indeed, it has been reported that stimulation of NMDA receptors increases the expression of c-*fos*, whereas non-NMDA receptors seem to have only a permissive or no role in these events (Vaccarino et al., 1992; Condorelli et al., 1994; Hou et al., 1997). Also, injection of $GABA_A$ receptor antagonists, such as bicuculline, or inhibitors of the synthesis of GABA, such as semicarbazide, into the midbrain tectum also produces the characteristic patterns of behavioral and autonomic responses related to fear (Brandão et al., 1982, 1986, 1988). Therefore, the existence of neural substrates of defensive behavior, and perhaps of fear in the midbrain tectum is evidenced not only by the aversive properties of its electrical stimulation but also by the fact that chemical stimulation with bicuculline or by excitatory amino acids has similar effects as electrical stimulation.

4. Neural substrates of anxiety in midbrain tectum

Because electrical stimulation of the dorsal periaqueductal gray induces defensive behavior and a host of autonomic and behavioral responses that resemble a panic attack in human beings, it has been suggested that this structure could be involved in panic disorder (Graeff, 1990; Graeff et al., 1986; Jenck et al., 1995; Vianna et al., 2001). In fact, stimulation of the dorsal periaqueductal gray in man has been reported to produce very unpleasant and fear-like sensations in human subjects (Nashold et al., 1969; Amano et al., 1978). Table 1 summarizes the similarities between various aspects of the symptomatic expression of panic attacks in man and the responses induced by dorsal periaqueductal gray stimulation in animals.

On the basis of pharmacological evidence showing antiaversive effects of benzodiazepines in animals, aversive dorsal periaqueductal gray stimulation, which animals learn to turn off, has been proposed as a model of anxiety (Brandão et al., 1982; Graeff, 1990, 1996). Local injections of benzodiazepines into the midbrain tectum depress both unlearned and learned escape behaviors induced by electrical stimulation of this region (Brandão et al., 1982; Melo et al., 1992; Bovier et al., 1982; Castilho and Brandão, 2001). That these effects are mediated by benzodiazepine

Table 2
Summary of various neurotransmitters which have been found to be implicated in the neural substrates of the defense reaction in the midbrain tectum

	Superior colliculus	Dorsal periaqueductal gray	Inferior colliculus
5-HT	Coimbra and Brandão, 1997	Graeff et al., 1986; Beckett et al., 1992; Lovick, 1993; Nogueira and Graeff, 1995; Graeff, 1997	Melo and Brandão, 1995; Castilho and Brandão, 2001
GABA-BZD	Imperato and DiChiara, 1981; Redgrave et al., 1981; DiScala et al., 1983; DePaulis and Vergnes, 1986; Dean et al., 1989	Brandão et al., 1982, 1986, 1990; Bovier et al., 1982; Audi and Graeff, 1987; Schenberg et al., 1983; Schmitt et al., 1985; DePaulis and Vergnes, 1986; DiScala et al., 1989; DiScala and Sandner, 1989; Motta et al., 1993	Brandão et al., 1988; Bagri et al., 1989; Melo et al., 1992; Pandossio et al., 1999
Substance P		Aguiar and Brandão, 1996; Mongeau et al., 1998; De Araújo et al., 2001a,b	
Cholecystokinin		Guimarães et al., 1992; Jenck et al., 1996; Luo et al., 1998	
Histamine	Santos et al., 2001, 2002	Santos et al., 2001, 2002	Santos et al., 2001, 2002
Dopamine			Cuadra et al., 2000
Neurotensin		Da-Silva et al., 1989	
Excitatory amino acids	Dean et al., 1988, 1989	Bandler, 1982; Bandler et al., 1985; Krieger et al., 1985; Carrive et al., 1989; Zhang et al., 1990; Guimarães et al., 1991	Cardoso et al., 1994; Pandossio et al., 1999; Maisonnette et al., 2000
Nitric oxide		De Oliveira et al., 2000	
Opioids	Coimbra et al., 2000; Eichenberger et al., 2002	Jacquet and Lajtha, 1974; Sharpe et al., 1974; Jenck et al., 1983; Motta et al., 1995	Cardoso et al., 1992; Coimbra et al., 2000
Glycine		Schmitt et al., 1997; Carobrez et al., 2001	
Chondroitin sulfate		De Araújo et al., 2001a,b	

The list is not complete in terms of papers published on a particular variable.

receptors is supported by the fact that chlordiazepoxide or midazolam raise the threshold of aversive electrical stimulation of the midbrain tectum and that the effects of both drugs were blocked by flumazenil, a competitive benzodiazepine receptor antagonist (Audi and Graeff, 1987).

The biochemical bases of the defense reaction have been examined for decades and the advances achieved in this area of research now provide a clearer picture of what happens in the brain during these processes. Although a wide range of transmitters are likely to be involved in these processes, strong agreement exists regarding a tonic inhibitory control exerted by GABAergic interneurons on neural circuits responsible for the tectally induced defense-like behavior, including fear-like responses. Interestingly, some obvious neurotransmitters, which might be expected to interact with the control of fear/defensive-related behavior, such as noradrenaline and adrenaline, have not been shown to be active modulators of tectal-defense mechanisms. However, in addition to GABA, also 5-hydroxy tryptamine serotonin (5-HT), opioids, neuropeptides, histaminergic and excitatory amino acids have also been implicated in the regulation of anxiety-related behaviors induced by stimulation of these midbrain tectum (Table 2). Efforts have been made to characterize how these neurotransmitters interact with each other to characterize a given stimulus as aversive and to trigger signals for the induction of defense reactions. In this context, we now know that there are also specificities in the neurochemical mechanisms involved in the modulation of the defense reactions, depending on the region studied. For example, dopaminergic mechanisms seem to be involved in the inferior colliculus, but not the dorsal periaqueductal gray, in the expression of adaptive responses to threatening situations (Cuadra et al., 2000; Troncoso et al., in press).

5. Theoretical considerations

Over the years, the focus of research in this area has changed in terms of the prevalent anatomy as well as conceptualization of the behavioral relevance. The earliest studies focused on hypothalamic structures, employing electrical stimulation thereof, to induce aggression-like behavior and flight-like responses or simply escape responses, as indexed by the animals' learning a response to terminate the stimulation. The sophistication of the theoretical constructs employed to account for the effects of electrically induced behaviors in this early work, for example, the induction of changes in receptive fields, responsive to particular stimulation, such as of visual or tactile importance (MacDonne and Flynn, 1966; Huston et al., 1980, 1990; Welzl et al., 1984; Schmitt et al., 1984), has been lost in recent years, with the current emphasis on pharmacological, anatomical and electrophysiological characterization of the systems under study. The notion of changes in sensorimotor fields should be reconsidered in attempting to account also for elements of the defense behavior repertoire that is elicitable via midbrain tectum stimulation (dorsal periaqueductal gray or inferior colliculus). Such complex behavioral patterns are unlikely to be strictly "in vacuo" responses, and are probably, as in the case of hypothalamic aggression (MacDonne and Flynn, 1966) and the central activation of the perioral biting reflex (Huston et al., 1980; Welzl et al., 1984) linked to changes in sensorimotor gating processes. For example, the inferior colliculus-induced defense behavior repertoire is accompanied by changes in auditory-evoked potentials in this structure, indicative of a modification of sensory input channels by the stimulation (Brandão et al., 2001). It is likely that various neurochemical (Cuadra et al., 2000) and molecular (Lamprea et al., 2002) concomitants of the midbrain tectum-induced defense behavior repertoire are potentially markers of such changes in sensorimotor thresholds or "fields" which influence the probability of responses.

The research emphasis has also shifted from the hypothalamus to other areas, namely, the midbrain tectum, including the periaqueductal gray matter, superior and inferior colliculus on the one hand, and the amygdala and its extensions such as bed nucleus of the stria terminalis (LeDoux et al., 1986; Schulz and Canbeyli, 1999, 2000) along with its afferents and efferents, on the other. Each of these areas of focus has emphasized somewhat different aspects of so-called "defensive behavior". Whereas different forms of the hypothalamic stimulation-induced "defensive"-like behaviors were described in terms of "rage", "aversion", "species-specific aggression", and "defensive–aggression", the amygdala and its extensions are generally conceptualized as a system that processes aversive inputs of all types and as a decision making unit for information related to aversive experiences. The midbrain tectal sites are often considered to contain substrates of "defensive behavior", "aversion", and "fear". Obviously, these systems must eventually be considered in relation to one another, although little attempt has been made at integrative research and/or theory in this regard, and this endeavor will be a challenge for the future.

In the present review, we emphasized work relevant to the midbrain tectum structures. Conceptually, these areas are considered to "mediate", "organize", "modulate", "represent", "be substrates of", or "contain mechanism underlying" defensive behavior, as defined by an array of behavioral and physiological responses, including attempts at escape from the situation (jumping), defensive posture (crouching), and fear (sympathetic responses, freezing, vocalization, etc.).

A large number of studies have looked at the effects of various neurotransmitter agonists or antagonists given systemically or directly into these structures, with the goal of characterizing the relevant neurochemical parameters involved in the control of these behaviors. Thus, the thrust of the empirical work in this area has been to delineate the midbrain tectum areas in the organization of elicited defen-

sive-like behaviors. Little effort has been made to relate this behavioral complex to other obviously related concepts of fundamental importance for behavioral neuropharmacology, such as the search for animal models for anxiety, depression, stress and schizophrenia. Such a synthesis must be attempted, lest this research area may become conceptually isolated. For example, it is clear that, let us say, electrical and chemical stimulation of dorsal periaqueductal gray and other areas like the inferior colliculus elicits the behavioral repertoire that operationalizes "defensive behavior" and that, by inference, these neural structures somehow "experience" and process aversive input and transduce it into behavioral and vegetative nervous system reactions. We can also infer that such induced states and behaviors must be either a precondition for the expression of "fear" as well as "anxiety" or a consequence thereof. We can hypothesize (I) that any manipulation of the dorsal periaqueductal gray and the inferior colliculus, which induces the "defensive behavior repertoire", will also be anxiogenic as measured by tests of anxiety, such as the elevated plus-maze, social interaction test, open-field avoidance, and dark preference, and that any manipulation which diminishes the "defensive behavior repertoire" in magnitude or quality is likely to have anxiolytic properties. In fact, a number of studies exist in which midbrain tectum manipulations have been measured in terms of the "defensive behavior repertoire", as well as in tests for fear such as the elevated plus-maze, and we have compiled a number of the existing studies in terms of the extent of the concordance between tectally elicited behaviors and anxiogenicity (Table 2). As hypothesized, it has been shown that there is indeed a very high correspondence between propensity of electrical or pharmacological manipulations that elicit the defense behavior repertoire also to have anxiogenic properties, and vice versa. Thus, we can conclude that the midbrain tectum "defense–aversion systems" as they are called, also comprise a substrate for, or source of information for state properties that decide the animals' behavior on the scale of anxiety as defined by performance in anxiety tests. It follows that the research in this area can be considered in terms not merely of "defensive–aversive behavior" as is often the case, but also in terms of animal models of fear and anxiety. Conversely, it follows that our understanding of neurochemical, anatomical and genetic substrates of fear and anxiety must take into account the neuropharmacology and chemistry of the midbrain tectum "defense system".

As a further step, we can hypothesize (II) that continuous inescapable induction of an aversive state, as by activation of this midbrain "defense behavior repertoire", should, in addition to inducing fear and anxiety, have further consequences which are known to occur upon otherwise chronically administered aversive stimulation. For one, chronic induction of the "defensive-response repertoire" may be expected to induce *stress* and the whole complex of neurohumoral events which define this condition.

Furthermore, (III) stress and inescapable aversive stimulation is considered to be a major source of certain kinds of depression, as defined by the variety of animal models for depression, such as the forced swimming task, etc. This link between midbrain tectum "defense system" and the concept of depression has been totally neglected, and deserves attention. For example, would chronic induction of the "defense-system repertoire" (and its accompanying state of fear and anxiety) by activation of the dorsal periaqueductal gray or the colliculi lead to depressive-like behavior in appropriate animal models? (We are currently engaged in testing this hypothesis experimentally.)

Our proposal here is that the very active area of investigation of the midbrain tectum structures in terms of electrically and chemically elicited aversive-like states and defensive-like behaviors has neglected to spell out and to examine its potential significance for understanding a cascade of behaviors and behavioral deficits that are known to result form aversive stimulation. Perhaps this field of study has been conceptually isolated partially due to the adherence either to restricted empirical descriptions of the elicited behaviors, or to global terms such as "brain-defense-aversion system", etc. The same criticisms hold for the research foci on hypothalamic rage and aggression and, to same degree, for the massive work on amygdala-related information processing of noxious information. We have tried to point out the potential relevance of this field of research for understanding aversive event-induced processes, including fear and anxiety, stress, and depression, and perhaps other related consequent problems, such as stress and depression-induced cognitive and immune deficits (Fig. 1). The challenge will be to establish a neural and behavioral systems approach that can integrate for one, the neurochemical systems underlying the processes of aversive stimulation which have been treated in relative isolation, such as the hypothalamus, midbrain tectum and amygdala, and secondly, the neurochemical and anatomical systems that subserve the consequences of aversive information to the organism, including (a) the basic responses to aversive stimulation, such as fight, flight, fear and anxiety, and (b) the adverse consequences of excessive or chronic aversive stimulation, such as stress and depressive behavior and their physiological, immunological and behavioral concomitants.

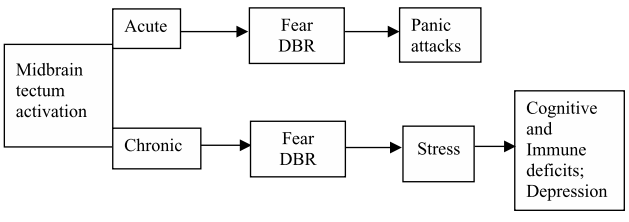

Fig. 1. Hypothetical scheme suggesting the involvement of midbrain tectum in fear. Acute fear stimuli activate midbrain tectum structures and produce the defense behavior repertoire, characterized by behavioral and autonomic responses accompanied by sensorimotor alterations that may underlie panic attacks. Chronic fear stimuli cause stress, which may lead to cognitive and immune deficits and depression. DBR = defense behavior repertoire.

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References

American Psychiatric Association, 1994. Diagnostic and Statistical Manual of Mental Disorders, 4th ed. American Psychiatric Association, Washington, DC.

Agarawal, S.G., Evans, R.H., 1986. The primary afferent depolarizing action of kainate in the rat. Br. J. Pharmacol. 87, 345–355.

Aguiar, M.S., Brandão, M.L., 1996. Effects of microinjections of the neuropeptide Substance P in the dorsal periaqueductal gray of rats submitted to the plus maze test. Physiol. Behav. 60 (4), 1183–1186.

Amano, K., Tanikama, T., Iseki, H., Kawabatake, H., Notani, M., Kawamura, H., Kitamura, K., 1978. Single neuron analysis of the human midbrain tegmentum: rostral mesencephalic reticulotomy for pain relief. Appl. Neurophysiol. 41, 66–78.

Audi, E.A., Graeff, F.G., 1987. $GABA_A$ receptors in the midbrain central grey mediate the antiaversive action of GABA. Eur. J. Pharmacol. 135, 225–229.

Bagri, A., Sandner, G., DiScala, G., 1989. Effects of unilateral microinjections of Gabaergic drugs into the inferior colliculus on auditory evoked potentials. Exp. Neurol. 104, 82–87.

Bandler, R., 1982. Induction of "rage" following microinjections of glutamate into midbrain but not hypothalamus of cats. Neurosci. Lett. 30, 183–188.

Bandler, R., Carrive, P., 1988. Integrated defense reaction elicited by excitatory amino acid microinjection in the midbrain periaqueductal grey region of the unrestrained cat. Brain Res. 439, 95–106.

Bandler, R., Shipley, M.T., 1994. Columnar organization in the midbrain periaqueductal gray: modules for emotional expression. Trends Neurosci. 17, 379–389.

Bandler, R., De Paulis, A., Vernes, M., 1985. Identification of midbrain neurones mediating defensive behaviour in the rat by microinjections of excitatory amino acids. Behav. Brain Res. 15, 107–109.

Bandler, R., McDougall, A., Dampney, R., 1986. Behavioural and cardiovascular components of the defense reaction: role of the midbrain periaqueductal gray. In: Oomura, Y. (Ed.), Emotions, Neuronal and Chemical Controls. Karger, Basel, pp. 405–416.

Bard, P., Match, M.B., 1951. In: Wolstenholme, G.E.W., O'Connor, C.M. (Eds.), The Neurobiological Basis of Behavior. The Ciba Foundation, J. & A. Churchill, London, UK, pp. 55–75.

Beckett, S.R.G., Lawrence, A.J., Marsden, C.A., Marshall, P.W., 1992. Attenuation of chemically induced defense response by 5-HT1 receptor agonists administered into the periaqueductal gray. Psychopharmacology 108, 110–114.

Behbehani, M.M., 1995. Functional characteristics of the midbrain periaqueductal gray. Prog. Neurobiol. 46, 575–605.

Blanchard, D.C., Williams, G., Lee, E.M.C., Blanchard, R.J., 1981. Taming of wild *Rattus norvegicus* by lesions of the mesencephalic central gray. Physiol. Psychol. 9, 157–163.

Blanchard, R.J., Blanchard, D.C., Hori, K., 1989. An ethoexperimental approach to the study of defense. In: Blanchard, R.J., Brain, P.F., Blanchard, D.C., Parmigiani, S. (Eds.), Ethoexperimental Approaches to the Study of Behavior. Kluwer Academic Publishing, Dordrecht, pp. 114–136.

Bovier, P., Broekkamp, C.L.E., Lloyd, K.G., 1982. Enhancing GABAergic transmission reverses the aversive state in rats induced by electrical stimulation of the periaqueductal grey region. Brain Res. 248, 397–402.

Brandão, M.L., Aguiar, J.C., Graeff, F.G., 1982. GABA mediation of the antiaversive action of the minor tranquilizers. Pharmacol. Biochem. Behav. 16, 397–402.

Brandão, M.L., Cardoso, S.H., Melo, L.L., 1983. Defense mechanisms in the inferior colliculus. Behav. Brain Res. 58, 49–55.

Brandão, M.L., DiScala, G., Bouchet, M.J., Schmitt, P., 1986. Escape behavior induced by blockade of glutamic acid decarboxilase (GAD) in mesencephalic central gray or medial hypothalamus. Pharmacol. Biochem. Behav. 24, 497–501.

Brandão, M.L., Tomaz, C., Leão Borges, P.C., Coimbra, N.C., Bagri, A., 1988. Defence reaction induced by microinjection of bicuculline into the inferior colliculus. Physiol. Behav. 44, 361–365.

Brandão, M.L., Coimbra, N.C., Leão Borges, P.C., 1990. Effects of morphine and midazolam on reactivity to peripheral noxious and central aversive stimuli. Neurosci. Biobehav. Rev. 14 (4), 495–499.

Brandão, M.L., Cardoso, S.H., Melo, L.L., Motta, V., Coimbra, N.C., 1994. The neural substrate of defensive behavior in the midbrain tectum. Neurosci. Biobehav. Rev. 18 (3), 339–346.

Brandão, M.L., Troncoso, A.C., Melo, L.L., Sandner, G., 1997. Active avoidance learning using brain stimulation applied to the inferior colliculus as negative reinforcement in rats: evidence for latent inhibition. Neuropsychobiology 35, 30–35.

Brandão, M.L., Anseloni, V.Z., Pandóssio, J.E., De Araújo, J.E., Castilho, V.M., 1999. Neurochemical mechanisms of the defensive behavior in the dorsal midbrain. Neurosci. Biobehav. Rev. 23, 863–875.

Brandão, M.L., Coimbra, N.C., Osaki, M.Y., 2001. Changes in the auditory evoked potentials induced by fear-evoking stimulations. Physiol. Behav. 72, 365–372.

Cardoso, S.H., Melo, L.L., Coimbra, N.C., Brandão, M.L., 1992. Opposite effects of low and high doses of morphine on neural substrates of aversion in the inferior colliculus. Behav. Pharmacol. 3, 489–495.

Cardoso, S.H., Brandão, M.L., Coimbra, N.C., 1994. Defensive reactions evoked by activation of NMDA receptors in distinct sites of the inferior colliculus. Behav. Brain Res. 63, 17–24.

Carobrez, A.P., Teixeira, K.V., Graeff, F.G., 2001. Modulation of defensive behavior by periaqueductal gray NMDA/glycine-B receptor. Neurosci. Biobehav. Rev. 25, 697–709.

Carrive, P., 1991. Functional organization of PAG neurons controlling regional vascular beds. In: DePaulis, A., Bandler, R. (Eds.), The Midbrain Periaqueductal Grey Matter: Functional, Anatomical and Immunohistochemical Organization. Plenum, New York, pp. 67–100.

Carrive, P., Dampney, R.A.L., Bandler, R., 1989. Excitation of neurons in a restricted portion of the midbrain periaqueductal gray elicits both behavioural and cardiovascular components of the defense reaction I the unanaesthetised decerebrate cat. Neurosci. Lett. 81, 273–278.

Casseday, J.H., Covey, E., 1996. A neuroethological theory of the operation of the inferior colliculus. Brain Behav. Evol. 47, 311–336.

Castilho, V.M., Brandão, M.L., 2001. Conditioned antinociception and freezing using electrical stimulation of the dorsal periaqueductal gray or inferior colliculus as unconditioned stimulus are differentially regulated by $5\text{-}HT_{2A}$ in rats. Psychopharmacology 155, 154–162.

Coimbra, N.C., Brandão, M.L., 1997. Effects of $5\text{-}HT_2$ receptors blockade on fear-induced analgesia elicited by electrical stimulation of the deep layers of the superior colliculus and dorsal periaqueductal gray. Behav. Brain Res. 87, 97–103.

Coimbra, N.C., Tomaz, C., Brandão, M.L., 1992. Evidence for the involvement of serotonin in the antinociception induced by electrical and chemical stimulation of the mesencephalic tectum. Behav. Brain Res. 50, 77–83.

Coimbra, N.C., Eichenberger, G.C.D., Gorchinski, R.T., Maisonnette, S., 1996. Effects of the blockade of opioid receptor on defensive reactions elicited by electrical stimulation within the deep layers of the superior colliculus and dorsal periaqueductal gray. Brain Res. 736, 348–352.

Coimbra, N.C., Osaki, M.Y., Eichenberger, G.C.D., Ciscato, J.G., Jucá, C.E.B., Biojone, C.R., 2000. Effects of opioid receptor blockade on defensive behavior elicited by electrical stimulation of the aversive

substrates of the inferior colliculus in *Rattus norvegicus*. Psychopharmacology 152, 422–430.

Condorelli, D.F., Albani, P.D., Amico, C., Lukasiuk, K., Kaczmarek, L., Guffrida-Stella, A.M., 1994. Glutamate-receptor driven activation of transcription factors in primary neuronal cultures. Neurochem. Res. 19, 489–499.

Cuadra, G., Zurita, A., Macedo, C.E., Molina, V.A., Brandão, M.L., 2000. Electrical stimulation of the midbrain tectum enhances dopamine release in the frontal cortex. Brain Res. Bull. 52, 413–418.

Da-Silva, S.L., Brandão, M.L., Tomaz, C., 1989. Behavioral effects of neurotensin applied to periventricular structures of rats. Braz. J. Med. Biol. Res. 22, 711–715.

Dean, P., Mitchell, I.J., Redgrave, P., 1988. Responses resembling defensive behavior produced by microinjections of glutamate into superior colliculus of rats. Neuroscience 24, 501–510.

Dean, P., Redgrave, P., Westby, M., 1989. Event or emergency? Two response systems in the mammalian superior colliculus. Trends Neurosci. 12, 137–147.

De Araújo, J.E., Huston, J.P., Brandão, M.L., 2001a. Opposite effects of the C-terminal (anxiogenic) and N-terminal (anxiolytic) of SP in the dorsal periaqueductal gray: only the C-terminal effects are mediated by the tachychinin NK1-receptors. Eur. J. Pharmacol. 432, 43–51.

De Araújo, J.E., Hasenöhrl, R.U., Huston, J.P., Brandão, M.L., 2001b. Hyperanxiety produced by periaqueductal gray injection of chondroitin sulphate glycosaminoglycan. NeuroReport 12 (4), 3081–3084.

De Molina, A.F., Hunsperger, R.W., 1962. Organization of the subcortical system governing defense and flight reactions in the cat. J. Physiol. 160, 200–213.

De Oliveira, R.M.W., Del Bel, E.A., Mamede-Rosa, M.L.N., Padovan, C.M., Deakin, J.F.W., Guimarães, F.S., 2000. Expression of neuronal nitric oxide synthase mRNA in stress-related brain areas after restraint in rats. Neurosci. Lett. 289, 123–126.

DePaulis, A., Vergnes, M., 1986. Elicitation of intraspecific defensive behavior in the rat by microinjections of picrotoxin, a γ-aminobutyric antagonist, into the midbrain periaqueductal gray matter. Brain Res. 367, 87–95.

DiScala, G., Sandner, G., 1989. Conditioned place aversion produced by microinjections of semicarbazide into the periaqueductal gray of the rat. Brain Res. 483, 91–97.

DiScala, G., Schmitt, P., Karli, P., 1983. Unilateral injection of GABA agonists in the superior colliculus: asymmetry to tactile stimulation. Pharmacol. Biochem. Behav. 19, 281–285.

Eichenberger, G.C.D., Ribeiro, S.J., Osaki, M.Y., Maruoka, R.Y., Resende, G.C.C., Castellan-Baldan, L., Correa, S.A.L., Da Silva, L.A., Coimbra, N.C., 2002. Neuroanatomical and psychopharmacological evidence for interaction between opioid and GABAergic neural pathways in the modulation of fear and defense elicited by electrical and chemical stimulation of the deep layers of the superior colliculus and dorsal periaqueductal gray matter. Neuropharmacology 42, 48–59.

Fanselow, M.S., 1991. The midbrain periaqueductal gray as a coordinator of action in response to fear and anxiety. In: DePaulis, A., Bandler, R. (Eds.), The Midbrain Periaqueductal Grey Matter: Functional, Anatomical and Immunohistochemical Organization. Plenum, New York, pp. 151–173.

Graeff, F.G., 1990. Brain defense systems and anxiety. In: Roth, M., Burrow, G.D., Noyes, R. (Eds.), Handbook of Anxiety, vol. 3. Elsevier, New York, pp. 307–354.

Graeff, F.G., 1997. Serotonergic systems. Psychiatr. Clin. North Am. 20, 723–739.

Graeff, F.G., Brandão, M.L., Audi, E.A., Schultz, M.T.B., 1986. Modulation of the brain aversive system by GABAergic and serotonergic mechanisms. Behav. Brain Res. 21 (1), 65–72.

Guimarães, F.S., Carobrez, A.P., DeAguiar, J.C., Graeff, F.G., 1991. Anxiolytic effect in the elevated plus-maze of the NMDA receptor antagonist AP7 microinjected into the dorsal periaqueductal gray. Psychopharmacology 103, 91–94.

Guimarães, F.S., Russo, A.S., De Aguiar, J.C., Ballejo, G., Graeff, F.G., 1992. Anxiogenic-like effect of CCK-8 microinjected into the dorsal periaqueductal grey of rats in the elevated plus maze. In: Cooper, S.J., Iversen, S.D., Iversen, L.L. (Eds.), Multiple Cholecystokinin Receptors in the CNS. Oxford Univ. Press, Oxford, pp. 149–154.

Hess, W.R., Brügger, M., 1943. Das subkortikale zentrum der affectiven abwerreaktion. Helv. Physiol. Pharmacol. Acta 1, 33–52.

Hilton, S.M., Redfern, W.S., 1986. A search for brain stem cell groups integrating the defense reaction in the rat. J. Physiol. 378, 213–228.

Hou, Y.-N., Cebers, G., Terenius, L., Liljequist, S., 1997. Characterization of NMDA- and AMPA-induced enhancement of AP-1 DNA binding activity in rat cerebellar granule cells. Brain Res. 754, 79–87.

Hunsperger, R.W., 1956. Affektreaktionen auf elektrische Reizung in Himstamm der Katze. Helv. Physiol. Pharmacol. Acta 14, 70–92.

Huston, J.P., Nef, B., Papadoulos, G., Welz, H., 1980. Activation and lateralization of sensorimotor field for peripheral biting reflex by intranigral GABA agonist and by systemic apomorphine in the rat. Brain Res. Bull. 5 (6), 745–749.

Huston, J.P., Steiner, H., Weiler, H.-T., Morgan, S., Schwarting, R.K.W., 1990. The basal ganglia–orofacial system: studies on neurobehavioral plasticity and sensory-motor turning. Neurosci. Biobehav. Rev. 14, 433–446.

Imperato, A., DiChiara, G., 1981. Behavioural effects of GABA agonists and antagonists infused in the mesencephalic reticular formation-deep layers of superior colliculus. Brain Res. 224, 185–194.

Iwata, J., Chida, K., LeDoux, J.E., 1987. Cardiovascular responses elicited by stimulation of neurons in the central amygdaloid nucleus in awake but not anaesthetized rats resemble conditioned emotional responses. Brain Res. 418, 183–188.

Jacquet, Y.F., Lajtha, A., 1974. Paradoxical effects after microinjections of morphine in the periaqueductal gray matter in the rat. Science 185, 1055–1057.

Jenck, F., Schmitt, P., Karli, P., 1983. Morphine applied to the mesencephalic central gray suppresses brain stimulation escape. Pharmacol. Biochem. Behav. 19, 189–195.

Jenck, F., Moreau, J.L., Martin, J.R., 1995. Dorsal periaqueductal gray-induced aversion as a simulation of panic anxiety: elements of face and predictive validity. Psychiatry Res. 57, 181–191.

Jenck, F., Martin, J.R., Moreaus, J.L., 1996. Behavioural effects of CCKB receptor ligands in a validated simulation of panic anxiety in rats. Eur. Neuropsychopharmacol. 8, 291–298.

Jurgens, U., Richter, K., 1986. Glutamate-induced vocalization in the squirrel monkey. Brain Res. 373, 349–358.

Krieger, J.E., Graeff, F.G., 1985. Defensive behavior and hypertension induced by glutamate in the midbrain central gray of the rat. Braz. J. Med. Biol. Res. 18, 61–67.

Lamprea, M.R., Cardenas, F., Vianna, D.M., Castilho, V.M., Cruz-Morales, S., Brandão, M.L., 2002. The distribution of *fos* immunoreactivity in rat brain following freezing and escape responses elicited by electrical stimulation of the inferior colliculus. Brain Res. 950, 186–194.

LeDoux, J.E., Iwata, J., Pearl, D., Reis, D.J., 1986. Disruption of auditory but not visual learning by destruction of intrinsic neurons in the medial geniculate body of the rat. Brain Res. 371, 395–399.

Lovick, T., 1993. Serotonergic influence from nucleus raphe obscurus on neurones in the periaqueductal grey matter in the rat. Brain Res. 606, 92–98.

Lovick, T., 2000. Panic disorder—a malfunction of multiple transmitter control systems within the midbrain periaqueductal gray matter? Neuroscientist 6, 48–59.

Luo, B., Cheu, J.W., Siegel, A., 1998. Cholecystokinin B receptors in the periaqueductal gray potentiate defensive rage behavior elicited from the medial hypothalamus of the cat. Brain Res. 796, 27–37.

MacDonne, M.F., Flynn, J.P., 1966. Control of sensory fields by stimulation of hypothalamus. Science 152, 1406–1414.

Maisonnette, S.S., Kawasaki, M.C., Coimbra, N.C., Brandão, M.L., 1996. Effects of lesions of amygdaloid nuclei and substantia nigra on aversive responses induced by electrical stimulation of the inferior colliculus. Brain Res. Bull. 40 (2), 93–98.

Maisonnette, S., Villela, C., Carotti, A.P., Landeira-Fernandez, J., 2000. Microinfusion of nefazodone into the basolateral nucleus of the amygdala enhances defensive behavior induced by NMDA stimulation of the inferior colliculus. Physiol. Behav. 70, 243–247.

Melo, L.L., Brandão, M.L., 1995. Role of 5-HT_{1A} and 5-HT_2 receptors in the aversion induced by electrical stimulation of inferior colliculus. Pharmacol. Biochem. Behav. 51, 317–321.

Melo, L.L., Cardoso, S.H., Brandão, M.L., 1992. Antiaversive action of benzodiazepines on escape behavior induced by electrical stimulation of the inferior colliculus. Physiol. Behav. 51 (3), 557–562.

Merzenick, M.M., Reid, M.D., 1974. Representation of the cochlea within the inferior colliculus of the cat. Brain Res. 77, 397–415.

Mongeau, R., DeOca, B.M., Fanselow, M.S., Marden, C.A., 1998. Differential effects of neurokinin-1 receptor activation in subregions of the periaqueductal gray matter on conditioned and unconditioned fear behaviors in rats. Behav. Neurosci. 112, 1125–1135.

Motta, V., Penha, K., Brandão, M.L., 1995. Effects of microinjections of μ and κ receptors agonists in the dorsal periaqueductal gray of rats submitted to the plus maze. Psychopharmacology 120, 470–474.

Nashold, B.S., Wilson, W.P., Slaughter, D.G., 1969. Sensation evoked by stimulation in the midbrain of man. J. Neurosurg. 30, 14–24.

Nogueira, R.L., Graeff, F.G., 1995. Role of 5-HT receptor subtypes in the modulation of dorsal periaqueductal gray generated aversion. Pharmacol. Biochem. Behav. 52, 1–6.

Olds, M.E., Olds, J., 1963. Approach-avoidance analysis of rat diencephalon. J. Comp. Neurol. 388, 1–40.

Pandossio, J.E., Brandão, M.L., 1999. Defensive reactions are counteracted by microinjections of midazolam and muscimol and elicited by activation of glutamate receptors into the inferior colliculus. Psychopharmacology 142, 360–368.

Redgrave, P., Dean, P., Souki, W., Lewis, G., 1981. Gnawing and changes in reactivity produced by microinjections of picrotoxin into the superior colliculus of rats. Psychopharmacology 75, 198–203.

Rose, J.E., Greenwood, D.D., Goldberg, J.M., Hind, J.E., 1963. Some discharge characteristics of single neurons in the inferior colliculus of the cat: I. Tonotopical organization, relation of spike counts to tonic intensity and firing patterns of single elements. J. Neurophysiol. 26, 294–320.

Sandner, G., Oberling, P., Silveira, M.C.L., Di Scala, G., Rocha, B., Bagri, A., Depoortere, R., 1993. What brain structures are active during emotions? Effects of brain stimulation elicited aversion on c-*fos* immunoreactivity and behaviour. Behav. Brain Res. 58, 9–18.

Santos, N.R., Huston, J.P., Brandão, M.L., 2001. Escape behavior under tonic inhibitory control of histamine H_2-receptor mediated mechanisms in the midbrain tectum. Behav. Brain Res. 124, 167–175.

Santos, N.R., Huston, J.P., Brandão, M.L., 2002. Further evidence for the involvement of histamine H_2-receptor in the control of defensive behavior generated in the midbrain tectum. Behav. Pharmacol. 13, 73–81.

Schenberg, L.C., DeAguiar, J.C., Graeff, F.G., 1983. GABA modulation of the defense reaction induced by brain electrical stimulation. Physiol. Behav. 31, 429–438.

Schmitt, P., Di Scala, G., Jenck, J., Sandner, G., 1984. Periventricular structures, elaboration of aversive effects and processing of sensory information. In: Bandler, R. (Ed.), Modulation of Sensorimotor Activity During Alterations in Behavioral States. Alan Liss, New York, pp. 393–414.

Schmitt, P., Carrive, P., DiScala, G., Jenck, F., Brandão, M.L., Bagri, A., Moreau, J.L., Sandner, G., 1986. A neuropharmacological study of the periventricular neural substrate involved in flight. Behav. Brain Res. 22, 181–190.

Schulz, D., Canbeyli, R.S., 1999. Freezing behavior in BNST-lesioned Wistar rats. Ann. N.Y. Acad. Sci. 877, 728–731.

Schulz, D., Canbeyli, R.S., 2000. Lesion of the bed nucleus of the stria terminalis enhances learned despair. Brain Res. Bull. 52, 83–87.

Sharpe, L.G., Garnett, J.E., Cicero, T.J., 1974. Analgesia and hyperactivity produced by intracranial microinjections into the dorsal periaqueductal gray of the rat. Behav. Biol. 11, 303–313.

Silveira, M.C.L., Sandner, G., Graeff, F.G., 1993. Induction of Fos immunoreactivity in the brain by exposure to the elevated plus-maze. Behav. Brain Res. 56, 115–118.

Silveira, M.C.L., Sandner, G., Di Scala, G., Graeff, F.G., 1995. c-*fos* immunoreactivity in the brain following electrical or chemical stimulation of the medial hypothalamus of freely moving rats. Brain Res. 674, 265–274.

Silveira, M.C.L., Zangrossi, H., Viana, M.B., Silveira, R., Graeff, F.G., 2001. Differential expression of Fos protein in the rat brain induced by performance of avoidance or escape in the elevated T-maze. Behav. Brain Res. 126, 13–21.

Troncoso, A.C., Osaki, M.Y., Mason, S., Borelli, K.G., Brandão, M.L., 2003. Apomorphine enhances conditioned responses induced by aversive stimulation of the inferior colliculus. Neuropsychopharmacology (in press).

Vaccarino, F.M., Hayward, M.D., Nestler, E.J., Duman, R.S., Tallman, F., 1992. Differential induction of immediate early genes by excitatory amino acid receptor types in primary cultures of cortical and striatal neurones. Mol. Brain Res. 12, 233–241.

Vianna, D.M.L., Graeff, F.G., Brandão, M.L., Landeira-Fernandez, J.M., 2001. Dorsolateral and ventral regions of the periaqueductal gray matter are involved in distinct types of fear. Neurosci. Biobehav. Rev. 25, 711–719.

Watkins, J.C., Krogsgaard-Larsen, P., Honoré, T., 1990. Structure–activity relationships in the development of excitatory amino acid receptor agonists and competitive antagonists. Trends Pharmacol. Sci. 11, 25–33.

Welzl, H., Schwarting, R., Huston, J.P., 1984. Perioral biting reflex and turning after intranigral injection of a GABA- or metenkephalin-agonist: role of the thalamus and superior colliculus. Exp. Brain Res. 55, 438–444.

Zhang, S.P., Bandler, R., Carrive, P., 1990. Flight and immobility evoked by excitatory amino acid microinjection within distinct parts of the subtentorial midbrain periaqueductal gray of the cat. Brain Res. 520, 73–82.