BIOCHEMICAL FACTORS IN ANXIETY AND RELATED DISORDERS

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In coping with certain stressful stimuli, a biologic "fight or flight" response becomes a necessity and would be considered a "normal" anxiety response. However, if such a reaction was excessive in intensity and duration, or occurred without sufficient objective reasons, then it would be considered an "abnormal" anxiety response (Hoehn-Saric, 1979). Differentiating "pathological" from "normal" anxiety requires careful clinical assessment, and use of specifically defined diagnostic criteria for anxiety disorders such as the Diagnostic and Statistical Manual of Mental Disorders, third edition revised (DSM-III-R) published by the American Psychiatric Association (1987) are essential. According to the most recent Epidemiologic Catchment Area (ECA) study, anxiety disorders for persons 18 years and older were the most prevalent of all the major psychiatric disorders with a one-month prevalence rate of 7.3% (Regier et al., 1988). For the above reasons, understanding the pathology of anxiety and its treatment is necessary.

Although, psychoanalytic, behavior, and biologic models exist in explaining "pathological" anxiety, this chapter will focus on the recent developments related to the biochemical factors implicated in the genesis of

anxiety.

The correlation between central nervous system (CNS) anatomy and anxiety has not been fully elucidated. However, during the last few decades, significant advancements in the neurosciences has led to investigations into the biochemistry of anxiety, the mechanisms of biological treatments for anxiety, and how these are related to each other. Neurotransmitters mediating anxiety have been discovered and include norepinephrine (NE), serotonin (5-HT), and gamma-aminobutyric acid (GABA).

NOREPINEPHRINE

Catecholamines such as norepinephrine mediate a variety of physiologic responses initiated by interactions at specific cellular membrane sites known as adrenergic receptors. The binding of catecholamines or related drugs induces changes in these receptors that lead to cellular events resulting in the characteristic physiologic effect of the neurotransmitter or drug. Agonist drugs are capable of inducing a response, with full agonists producing a maximal response and partial agonists producing a qualitatively similar response, but of lesser magnitude. An antagonist interacts with the receptor and elicits no response on its own but may reduce the effect of an agonist by occupying the receptor (Hoffman & Lefkowitz, 1980).

Norepinephrine is the principal postganglionic sympathetic

neurotransmitter. Sympathetic nervous system pathways originate from preganglionic neurons with cell bodies in the thoracolumbar segments of the spinal cord that synapse in the sympathetic ganglia with postganglionic neurons which innervate end-organs, including the heart and vascular, gastrointestinal, and genitourinary smooth muscle. Stimulation of the sympathetic nervous system leads to the classic flight or fight response (Lefkowitz, 1988). Based on the relative potencies of several adrenergic agonists, Ahlquist (1948) classified adrenergic receptors into two distinct types, termed alpha and beta. Alpha-adrenergic receptors recognize epinephrine and norepinephrine with high affinity and isoproterenol with much lower affinity while beta-adrenergic receptors have a higher affinity for the agonist isoproterenol than either epinephine or norepinephrine. The development of highly specific antagonist drugs has led to the identification of two subtypes of both alpha- and beta-adrenergic receptors. Beta receptors are subtyped based on the relative potencies of epinephrine and nonepinephrine. At beta 1-receptors, which mediate positive inotropic effects in the heart and lipolysis in adipose tissue, epinephrine and norepinephrine are of similar potency. In contrast, at beta2-receptors, epinephrine is much more potent than norepinephrine; these receptors mediate vascular and bronchial smooth muscle relaxation. Both beta1- and beta2-adrenergic receptors are believed to act by stimulating the plasma membrane-bound enzyme adenylate cyclase, generating cyclic adenosine monophosphate

(cAMP) (Lefkowitz, 1988).

Alpha-adrenergic receptor subtypes differ in pharmacologic specificity and in mechanism of action. Alpha 1-receptors are found in vascular smooth muscle and mediate the vasoconstrictor effects of sympathetic stimulation. These receptors interact with specific agonists and antagonists and, while their exact mechanism of action is not clearly understood, alpha 1-adrenergic effects do not appear to involve changes in activity of adenylate cyclase. On the other hand, stimulation of alpha2-adrenergic receptors mediates platelet aggregation via inhibition of adenylate cyclase activity and reduction in cellular cAMP levels. It is not known whether a similar mechanism accounts for the other known alpha2-adrenergic effects of presynaptic nerve terminal inhibition of norepinephrine release and postsynaptic smooth muscle contraction in some vascular beds (Lefkowitz, 1988).

In addition to the spinal sympathetic pathways, there are central nervous system noradrenergic pathways that originate in the pons and the medulla oblongata. The brain's principal norepinephrine-containing nucleus, the pontine nucleus locus ceruleus, provides noradrenergic innervation to the cerebral and cerebellar cortices, the limbic system, the brain stem, and the spinal cord; it also receives extensive afferent projections. Electrical stimulation of the locus ceruleus in monkeys and drugs, such as the alpha2-adrenergic antagonist yohimbine, which stimulate locus ceruleus activity and

norepinephrine release, have been shown to induce anxiety in monkeys and man; drugs that decrease locus ceruleus function, such as the alpha2-agonist clonidine, have anxiety-reducing properties (Hoehn-Saric, 1982; Redmond & Huang, 1979). An exception to this is buspirone, a nonbenzodiazepine anxiolytic that increases locus ceruleus adrenergic activity (Sanghera, McMillen, & German, 1983). Therefore, with its unique anatomic and functional connections, the locus ceruleus is thought to be involved in but not totally responsible for the manifestation of anxiety, which requires numerous CNS systems for full expression. It has been suggested that the locus ceruleus may function as an "alarm system" that modulates emotional and autonomic responses, with clinical anxiety resulting from alterations in the operation of this system (Redmond & Huang, 1979).

Both types of alpha- and beta-adrenergic receptors are present in the brain. Alpha2-autoreceptors present in the locus ceruleus respond to norepinephrine and noradrenergic agonists by inhibiting norepinephrine release in the brain, probably as part of a negative feedback loop that regulates locus ceruleus activity (Hoehn-Saric, 1982). In addition to norepinephrine, other neurotransmitters such as epinephrine, serotonin, Metenkephalin, GABA, and glycine are reported to influence activity in noradrenergic locus ceruleus neurons. Various psychotherapeutic agents, including amphetamine, morphine, benzodiazepines, and tricyclic antidepressants, also inhibit these cells (Foote, Bloom, & Aston-Jones, 1983).

It has been postulated that agents which decrease net noradrenergic function should have anxiolytic properties (Redmond & Huang, 1979). This may account for the antianxiety effects of certain tricyclic antidepressants by increasing norepinephrine levels at alpha2-adrenergic autoreceptors in the locus ceruleus. However, both nonsedating tricyclic and monoamine oxidase inhibitor antidepressants have no immediate anxiolytic effects and usually require several days to several weeks of continuous treatment to be effective. Therefore, the actions of these antidepressants must be ascribed to long-term adjustments rather than short-term changes in neurotransmission (Hoehn-Saric, 1982).

Norepinephrine has long been implicated in physiological and psychological responses to stress, and it has recently been suggested that, in addition to norepinephrine-mediated sympathetic effects, some somatic and psychological symptoms of stress may be attributable to circulating adrenaline (Greenwood, 1990). Patients with flight phobia demonstrated increased heart rate, plasma adrenaline, blood pressure, and perceived anxiety during flight while plasma noradrenaline did not change (Ekeberg, Kjeldsen, Greenwood, & Enger, 1990). Examination stress in medical students has been shown to be associated with reduced platelet alpha2-adrenergic receptor binding affinity and increased levels of plasma norepinephrine and reported anxiety (Freedman et al,, 1990).

Noradrenergic Status in Specific Anxiety Disorders

As described above, preclinical and clinical studies have generally supported a role for norepinephrine in stress and anxiety. Recent clinical investigations using the alpha2-adrenergic antagonist yohimbine and alpha2-receptor agonist clonidine have attempted to clarify noradrenergic function in specific anxiety disorders.

Panic Disorder

The effects of yohimbine and clonidine indicate dysregulation of noradrenergic activity in some patients with panic disorder (Charney & Heninger, 1986). More specifically, the ability of yohimbine to preferentially induce panic attacks in patients with panic disorder compared to controls supports the hypothesis that increased noradrenergic neuronal activity is associated with the pathophysiology of panic attacks in a subgroup of panic disorder patients (Charney et al,, 1990). Consistent with this theory is clonidine's ability to decrease noradrenergic function and reduce anxiety in panic disorder patients compared to controls. While intravenous clonidine has been demonstrated to have short-term anxiolytic effects in panic disorder patients, the effects reportedly did not persist in most patients with long-term oral administration (Uhde et al,, 1989). Regulation of beta-adrenergic receptors may also be abnormal in panic disorder patients. The lack of

consistent noradrenergic responses, as well as the ability of drugs such as lactate and caffeine to induce similar panic states, suggests that it is unlikely that panic anxiety is associated with disturbances in a single neurotransmitter system (Charney et al., 1990).

Obsessive-Compulsive Disorder (OCD)

Recent data suggest that abnormal noradrenergic function probably does not play a primary role in the pathogenesis of obsessive-compulsive disorder. Following administration of yohimbine, behavioral responses and plasma levels of the norepinephrine metabolite 3-methoxy-4-hydroxyphenylglycol (MHPG) failed to differentiate OCD patients from healthy controls (Rasmussen, Goodman, Woods, Heninger, & Charney, (1987). Other studies have failed to show differences between OCD patients and controls in pulse, blood pressure, growth hormone, and MHPG responses to clonidine (Hollander et al., 1991; Lee et al., 1990). One report did indicate a transient but significant reduction in obsessions and compulsions correlated with growth hormone response to clonidine, suggesting noradrenergic mediation (Hollander et al., 1991).

Generalized Anxiety Disorder (GAD)

Recent studies have reported conflicting results about noradrenergic

activity in generalized anxiety disorder. One report indicated that noradrenergic activity appeared to be increased in patients with GAD compared to normal controls and patients with major depression (Sevy, Papadimitriou, Surmont, Goldman, & Mendlewicz, 1989). Another study found that behavioral, cortisol, and cardiovascular responses to yohimbine did not differ between GAD patients and healthy controls, suggesting that GAD patients do not exhibit noradrenergic hyperactivity (Charney, Woods, & Heninger, 1989).

Other Anxiety Disorders

A recent review of clinical and preclinical studies describes evidence for the involvement of central noradrenergic systems in the pathophysiology of post-traumatic stress disorder (PTSD) (Krystal et al., 1989). Clinical improvement reported in social phobia patients following treatment with beta-adrenergic blocking drugs suggests possible peripheral catecholamine mediation of symptoms, but the pathophysiology of social phobia remains uncertain (Gorman & Gorman, 1987; Liebowitz, Gorman, Fyer, & Klein, 1985).

Implications for Treatment

The effectiveness of some tricyclic and monoamine oxidase inhibitor (MAOI) antidepressants in panic disorder may be related to their inhibitory

noradrenergic effects, including reductions in tyrosine hydroxylase activity, locus ceruleus neuronal firing rates, norepinephrine turnover, and postsynaptic beta-adrenergic receptor sensitivity. Interestingly, bupropion and trazodone, two antidepressants without antipanic efficacy, have different effects on noradrenergic function (Charney et al., 1990; Sheehan, Davidson, & Manschrek, 1983).

Clonidine has been shown to have some antianxiety effects independent of sedation in patients with panic disorder and generalized anxiety disorder, although the degree of response was highly variable and less impressive than with other drugs (Hoehn-Saric, 1982). As described, clonidine has been shown in research settings to produce transient improvement in OCD symptoms (Hollander et al., 1991), limiting its clinical usefulness.

Beta-adrenoreceptor blocking drugs have been used in the treatment of a variety of anxiety and stress-related disorders, such as performance anxiety. In general, they are most effective in treating somatic or autonomic symptoms of anxiety, such as tremor and palpitations, and are thought to do so by blocking peripheral beta-adrenergic receptors (Tyrer, 1987). Some improvement in psychic anxiety has also been reported with propranolol treatment at higher doses for longer periods of time. It is postulated that the antianxiety effects of lipophilic beta-adrenergic blockers such as propranolol may be partly due to central mechanisms involving other neurotransmitter

systems (Greenwood, 1990).

Summary

There is extensive preclinical and clinical information implicating excessive noradrenergic activity in the etiology of stress and anxiety, and abnormalities in norepinephrine function have been associated with specific anxiety disorders. Antidepressants may exert their antianxiety effects by reduction of overall noradrenergic activity via stimulation of presynaptic alpha2-adrenergic autoreceptors in locus ceruleus neurons as part of a negative feedback (inhibitory) control mechanism. Beta-adrenergic blocking drugs are thought to reduce somatic manifestations of anxiety primarily by blocking peripheral beta-adrenergic receptors, although some central nervous system actions have also been postulated. While norepinephrine apparently plays a role in the pathophysiology of certain types of anxiety, its role certainly does not seem to be an exclusive one as other neurotransmitters are also implicated.

SEROTONIN

When serotonin was initially studied and isolated in the 1930s and 1940s, the primary interest was its vasoconstrictive properties. It is unlikely that anyone at that time could have anticipated the emerging role serotonin

would assume as an important neurotransmitter involved in anxiety disorders.

Researchers at the Cleveland Clinic first crystallized this substance and named it serotonin (Rapport, Green, & Page, 1948) because it was found in blood (*sero*) and induced contraction (*tonin*) of the gastrointestinal smooth muscle. Later Rapport determined that the chemical structure for serotonin was 5-Hydroxytryptamine (5-HT). In the mid-1950s, serotonin was demonstrated to be in brain tissue (Amin, Crawford, & Gaddum, 1954).

Serotonin is present throughout the body with 90% in the enterochromaffin cells of the gastrointestinal tract. Most of the rest is found in the central nervous system and in platelets. The function of serotonin in these non-nervous system areas includes regulating gastrointestinal motility, hemostasis involving platelets, and vasospasms associated with certain vascular diseases (Garrison, 1990). In the central nervous system, serotonin's role includes being the precursor for the pineal hormone melatonin as well as a neurotransmitter. As a neurotransmitter, serotonin is noted to mediate pain sensation, itch sensation, appetite regulation, migraines, depression, and anxiety. This extensive and diverse list of functions for serotonin reveals how prophetic Brodie and Shore's words were in 1957 when proposing serotonin as a chemical mediator in the brain, "It is fascinating to learn how the discovery of serotonin, a substance that appears to have no part in the

general metabolism of cells, has proved to be of such significance to the pharmacologist, the biochemist, the neurologist, and possibly, to the psychiatrist'' (1957).

The investigation of serotonin's role in anxiety has centered on the heterogeneous and complex nature of the various serotonin receptor sites and serotonin re-uptake sites in both normal and abnormal brains as well as the effects of various serotonin agonists and antagonists. There are currently seven different serotonin receptor sites that have been identified and located in a variety of pre- and postsynaptic sites throughout the brain (Gonzalez-Heydrich & Peroutka, 1990). These seven receptors are broken into three classes: 5-HT_1 class (5-HT_{1A}, 5-HT_{1B}, and 5-HT_{1D}), 5-HT class (5-HT_{1C} 5-HT_{2A}, and 5-HT_{2B}), and the 5-HT, class (5-HT_1). 5-HT_{lc} is grouped in the 5-HT_2 family because of its similarities to the 5-HT_{2A} and 5-HT_{2B} subtypes. It is expected that this list will expand in the future.

To complicate things further, different second messenger systems have been associated with different serotonin receptors (Gonzalez-Heydrich & Peroutka, 1990). All of this helps explain the seemingly conflicting data obtained from various animal and human studies involving serotonin.

Recent studies of the anxiety disorders (obsessive-compulsive disorder, panic disorder, and generalized anxiety disorder), have implicated the

involvement of serotonin. This common factor is remarkable when compared to the distinct signs and symptoms of these disorders as categorized in the DSM-III-R. 5-HT selective drugs such as 5-HT releasing agents (e.g., fenfluramine) and 5-HT receptor agonists (e.g., metachlorophenylpiperazine [*m*-CPP], buspirone) have distinguished anxiety disorders in terms of behavioral and neuroendocrine responses. Fenfluramine by itself had minimal behavioral effects in obsessive-compulsive disorder (OCD) patients but produced panic episodes in panic disorder (PD) patients (Murphy & Pigott, 1990).

m-CPP produced changes in neuroendocrine responses as well as behavioral differences among various psychiatric patient groups as well as when compared to control groups. Some of the anxiety related responses could be differentiated depending on the dosage and route of administration of *m*-CPP.

When obsessive-compulsive disorder patients were given oral *m*-CPP, they showed transient exacerbations of their obsessive-compulsive symptoms while normal controls had no response (Zohar, Mueller, Insel, Zohar-Kadouch, & Murphy, 1987). This exacerbation of symptoms occurred in 11 of 12 OCD patients. Also notable is that a number of these affected patients reported the emergence of new symptoms or symptoms that had not been present for many months. A review of a number of studies using *m*-CPP

showed that panic symptoms could be significantly increased in PD patients at oral doses of 0.25 mg/kg and in OCD patients at oral doses of 0.5 mg/kg while in healthy volunteers minimal anxiety symptoms occurred with 0.5 mg/kg (Zuardi, 1990).

Cortisol level responses to intravenous and oral doses of *m*-CPP are different between PD patients and OCD patients. These in turn are distinct from control group responses (Murphy & Pigott, 1990).

Other evidence for the involvement of serotonin includes the actions of psychopharmacological agents used to treat anxiety disorders. Of the tricyclic antidepressants, clomipramine has shown a significantly greater ability to stop the re-uptake of 5-HT into brain synaptic terminals. In the treatment of OCD. clomipramine has proven superior to other tricyclics and monoamine oxidase inhibitors in numerous studies (Murphy & Pigott, 1990). Another 5-HT re-uptake blocker, fluoxetine, is also very effective in treating OCD.

The mechanism of action of agents like clomipramine and fluoxetine in blocking 5-HT re-uptake is not well understood. It is thought not to be related to the specificity of action of the agent on serotonin transport after acute administration of the agent but rather to adaptive changes in these sites following chronic administration (Leonard, 1988). This stands in contrast to GABA-mediated agents such as benzodiazepines that show reduction in

anxiety within hours after intake. 5-HT_{IA} receptor agonists are currently being established as effective tools to treat GAD. Again, the mechanism of action is not clearly identified. Agents such as buspirone act as agonists on presynaptic 5-HT_{1A} receptors but are also partial agonists at the postsynaptic 5-HT_{1A} receptor (Eison, 1990). An important distinction between these 5-HT_{1A} agonists and the previously discussed serotonin re-uptake blockers is that the 5-HT_{1A} agonists act specifically at 5-HT_{1A} while the re-uptake blockers make serotonin available to interact with all subtypes of 5-HT receptors (Eison, 1990).

Buspirone has been shown to be as equally effective in treating generalized anxiety disorder as benzodiazepines (e.g., diazepam). Two significant differences should be noted. First, benzodiazepines work after the first dose while buspirone takes weeks of administration to get a similar result. Two, the side effect profile of buspirone does not include sedation, ataxia, amnesia, and withdrawal problems that can be associated with benzodiazepines (Charney, Krystal, Delgado, & Heninger, 1990). Clinical indications for using buspirone in GAD include patients who are elderly, have concurrent medical problems, show mixed symptoms of depression and anxiety, and those who do not demand immediate gratification or the immediate relief they associate with a benzodiazepine response (Rickels, 1990).

The 5-HT_2 receptor is also being studied because various 5-HT_1 receptor antagonists, such as ritanserin, have been effective in GAD (Charney, Krystal, Delgado, & Heninger, 1990). Early animal studies looking at 5-HT_3 receptor antagonists indicate it may have anxiolytic properties (Jones et al., 1988).

GABA/BENZODIAZEPINE RECEPTOR COMPLEX

Gamma-aminobutyric acid (GABA) is an amino acid neurotransmitter in the CNS which, as the most important inhibitory neurotransmitter, is potent in its ability to affect neuronal discharge. There are two subtypes of GABA receptors: GABA-A receptors where benzodiazepines enhance the binding of GABA, and GABA-B receptors where benzodiazepines do not enhance the binding of GABA (Wojcik & Neff, 1984). Currently, the most predictable anxiolytic effects are associated with the benzodiazepines, which facilitate the activity of GABA. The benzodiazepines diazepam and alprazolam have been widely prescribed for generalized and anticipatory anxiety, and panic disorder (Rickels, Schweizer, Csanalosi, Case, & Chung, 1988; Sheehan, 1987), respectively. Such linkage between the GABA-ergic subsystem and specific benzodiazepine receptors has provided a molecular basis of anxiety and understanding the neurobiology of anxiety. Presently, there are two receptor hypotheses for the genesis of anxiety. The first one is that changes in activity of endogenous ligands for the benzodiazepine receptor (e.g., an excess of

anxiogenic) or a deficiency of anxiolytic substances regulates anxiety. The second one is that shifts in the benzodiazepine receptor sensitivity (e.g., increased or decreased receptor sensitivity to agonist drugs) may regulate anxiety (Nutt, Glue, & Lawson, 1990).

In 1977, two independent groups of researchers in Denmark and Switzerland reported the existence of saturable, high affinity, and stereospecific binding sites for benzodiazepines in the CNS of reptiles and mammals (Mohler & Okada, 1977; Squires & Braestrup, 1977). The highest concentrations of benzodiazepine receptors are found in the cerebral cortex, cerebellum, and amygdala, and lesser concentrations in the hippocampus, striatum, and spinal cord (Braestrup, Albrechtsen, & Squires, 1977). Studies exploring the relationship between the GABA-ergic system and benzodiazepines have substantiated the presence of a pharmacologic receptor for benzodiazepines in brain.

Benzodiazepines potentiate the effects of GABA, but do not produce anxiolysis when GABA is absent (Guidotti, 1981). Activation of benzodiazepine binding sites causes an allosteric change in the GABA receptor recognition site, consequently increasing receptor sensitivity to GABA (Enna, 1984; Paul & Skolnick, 1983; Tallman, Thomas, & Gallager, 1978; Tallman, Paul, Skolnick, & Gallager, 1980). Small permeable anions such as chloride also increase the binding of GABA to their receptors (Costa,

Rodbard, & Pert, 1979). These two effects combined suggest that GABA inhibits neuronal excitability by opening the chloride channel ionophore directly linked to GABA receptors. Consequently, chloride conductance increases and allows chloride ions to move more readily from the extracellular space to the inside of the neuron (McDonald & Barker, 1979). Therefore, a structural and functional model of the GABA/Benzodiazepine receptor "supramolecular receptor complex" consisting of a chloride ion channel and two binding sites has been formulated. One receptor site binds GABA and the other one binds benzodiazepine (Breier & Paul, 1990). Barbiturates also enhance the GABA receptors by interacting directly with the chloride channel (Enna, 1984).

It has been proposed that hyperexcitability of certain neuropathways is associated with anxiety. In an overactive state, a feedback signal to a GABA neuron is sent and then GABA is released into the synaptic cleft. GABA binds to its receptor to open chloride channels and increase the influx of chloride ions into the neuron. The net effect of enhanced chloride permeability causes hyperpolarization of the nerve membrane. Hyperpolarization makes the neuron less likely to be excitable and this is associated with the alleviation of anxiety. With the administration of a benzodiazepine, GABA-mediated chloride conductance is facilitated and excitability of the neuron is further inhibited (Goldberg, 1984).

Isotope-labeled ligands of benzodiazepine receptors have been used to explore the neurochemical basis of epileptic patients: indirect evidence for the role of the GABA/benzodiazepine receptor in the pathophysiology of anxiety has been found (Savic et al., 1988).

Reduced benzodiazepine sensitivity was reported through a study in response to intravenous administration of diazepam. Saccadic eye movement velocity decreased less in patients with panic disorder than in nonanxious control subjects and suggests that panic disorder is associated with a functional subsensitivity of the GABA/benzodiazepine supramolecular complex in brainstem areas controlling saccadic eye movements (Roy-Byrne, Cowley, Greenblatt, Shader, & Hommer, 1990). The reason for the reduced sensitivity is not clear but may be related to the anxiety disorder or may be related to the effect of benzodiazepine (Hoehn-Saric, 1991).

Under stressful conditions, the secretion of steroid hormones such as progesterone and deoxycorticosterone will increase and significantly affect the CNS function. Their metabolites have potent benzodiazepine like effects that mediate through recognition sites on the GABA/ benzodiazepines receptor (Breier & Paul, 1990).

Data from the use of β-carboline-3-carboxylate ethyl ester ((β-CCE) supports the role of the GABA receptor in mediating anxiety. β-CCE, a

benzodiazepine receptor antagonist, has been used to probe the neurobiological base of anxiety (Braestrup & Nielsen, 1981). Administration of β-CCE to rhesus monkeys and a β-carboline derivative, FG-7142, to humans induced behaviors similar to the stress-related responses of behavioral "agitation" accompanied by marked physiologic and endocrine changes. Administration of a benzodiazepine then blocks such responses (Dorow, Horowski, Paschelke, Amin, & Braestrup, 1983; Insel et al., 1984).

At present, the activation of a GABA-ergic subsystem has the most predictable anxiolytic effects (Hoehn-Saric, 1982). More sophisticated research in this area has been carried out and will eventually lead to a better biochemically based explanation into the genesis of anxiety disorders in the future.

OTHER BIOCHEMICAL MEASUREMENTS IN ANXIETY

Lactic Acid (Lactate)

Higher blood concentrations of lactate were found in anxiety-prone individuals than in normal individuals after exercise (Linko, 1950). Pitts and McClure (1967) and other researchers (Gorman et al., 1988, 1989) intravenously infused lactate solution, anxiety symptoms were provoked only in susceptible individuals. This precipitation can be successfully inhibited or

attenuated by the administration of tricyclic antidepressants (TCAs) or monoamine oxidase inhibitors (MAOIs), but not by benzodiazepine or β-adrenergic antagonists (e.g., propranolol) (Liebowitz et al., 1984). Although the mechanism for lactate's effect is not clearly understood, it may be related to the rise in the lactate-pyruvate ratio, lowering the level of ionized calcium, and the concomitant fall of intraneuronal pH in the chemoreceptor (Carr & Sheehan, 1984). The biochemistry of these chemically induced anxiety symptoms still needs to be studied further (Gaffney, Fenton, Lane, & Lake, 1988; Gorman et al., 1989; Reiman et al., 1989).

Carbon Dioxide

The investigation of carbon dioxide's role in causing anxiety remains confusing and perplexing to researchers. Carbon dioxide levels are adjusted acutely through a person's respiratory rate. Increasing an individual's breathing rate (hyperventilation) leads to hypocapnia (reduced carbon dioxide in the circulating blood) while decreasing the breathing rate (hypoventilation) leads to hypercapnia (elevated carbon dioxide in the circulating blood). Investigators have used this phenomenon as well as exposing patients to predetermined concentrations of carbon dioxide to accurately measure its effect on anxiety, especially panic attacks.

One initial proposal was that alterations in blood carbon dioxide, which

can have profound effects on cerebral blood flow, caused anxiety. However, repeated studies have shown that hypercapnia and hypocapnia have opposite actions on blood flow but both are anxiogenic (Nutt, 1990). The possibility that marked hyperventilation seen in compensation to hypercapnia as the cause of panic has also been discredited.

Another hypothesis involves carbon dioxide induced stimulation of noradrenergic neuronal function leading to panic attacks. Although animal studies have shown evidence for this, it has not been replicated in human studies (Woods, Charney, Goodman, & Heninger, 1988). Along these lines, one recent study has implicated hyperventilation to increased vagal tone and subsequent reduced parasympathetic nervous system activity. This would result in a relative increase in sympathetic activity without direct sympathetic excitation (George et al., 1989).

A combination cognitive-physiological model has also been suggested. Hyperventilation or hypoventilation induces body sensations that are perceived as unpleasant and are interpreted in a catastrophic manner. This would account for the paradoxical anxiogenic effect of both hypo- and hyperventilation. Also, this is consistent with studies showing evidence for behavioral hypersensitivity to carbon dioxide and weak or no evidence for physiologic hypersensitivity to carbon dioxide in patients with panic disorders (Woods, Charney, Goodman, & Heninger, 1988).

Caffeine

Because it is thought to have stimulant actions that could elevate mood, decrease fatigue, and increase capacity to work, caffeine is a popular ingredient in a variety of drinks, foods, and medications (Rail, 1990).

Caffeine is found mostly in drinks such as coffee (90 mg to 125 mg/ 250 ml), tea (40-60 mg/ml), cola drinks (40 mg/330 ml), and hot chocolate (5 mg/225 ml). Foods with coffee or chocolate also contain caffeine. Chocolate bars have about 20 mg per small bar. A significant number of over-the-counter preparations for analgesia, coughs, colds, and asthma also contain caffeine (Bruce, 1990). It is estimated that 80% of the world's population consumes caffeine. The per capita consumption among American adults is about 200 mg per day (Jaffe, 1990).

Caffeine is a methylxanthine and is similar in structure to xanthine, theophylline, and theobromine (Rail, 1990). As a central nervous system stimulant, caffeine's biochemical action is thought to be mediated by blocking receptors for adenosine (Boulenger, Patel, & Marangos, 1982). Caffeine does cause increased cerebrovascular resistance with resultant reduction in cerebral blood flow (Rail, 1990). However, attempts to correlate this specific action to caffeine's anxiogenic properties has been unsuccessful (Mathew & Wilson, 1990).

The clinical presentation of caffeine-induced anxiety can mimic other anxiety disorders such as PD and GAD (Charney, Heninger, & Breier, 1984). The DSM-III-R lists three criteria for the diagnosis of caffeine intoxication. One, the recent consumption of caffeine, usually more than 250 mg. Two, at least five of the following signs: restlessness, nervousness, excitement, insomnia, flushed face, diuresis, gastrointestinal disturbance, muscle twitching, rambling flow of thought and speech, tachycardia or cardiac arrhythmia, periods of inexhaustibility, or psychomotor agitation. Three, that the anxiety is not due to any physical or other mental disorder. Thus caffeine toxicity can be difficult to distinguish from other anxiety disorders unless an accurate history is obtained or toxicology tests are ordered.

In addition to appearing similar to other states, caffeine is known to complicate and worsen the conditions of persons with a pre-existing anxiety disorder. This has been noted in the clinical observation that many panic patients will put themselves on a caffeine-free diet because the subjective effects of caffeine (arousal, insomnia, upset stomach, and tremor) are unpleasant (Nutt, 1990).

In one study, patients with either generalized anxiety or panic disorders who underwent caffeine abstinence showed a significant reduction in long-standing anxiety symptoms as well as reductions in their anxiolytic medications. Some of these patients consumed less than 200 mg of caffeine a

day prior to abstaining, thus illustrating the powerful effect of even low doses of caffeine (Bruce & Lader, 1989).

Caffeine is also associated with a withdrawal syndrome. Headache and fatigue are the most frequently listed symptoms along with anxiety, impaired performance, nausea, and vomiting. The onset is 12 to 24 hours with a peak at 20 to 48 hours and lasting up to one week. Thus patients wishing to abstain from caffeine should be advised of the short-term withdrawal symptoms and high users of caffeine should taper their intake over one to two weeks to minimize these symptoms (Bruce, 1990).

Miscellaneous Biochemical Measurements

Other studies suggest certain physiological differences between individuals with and without anxiety. Individuals with anxiety disorders show an increased resting forearm blood flow (Kelly & Walter, 1968), brisker deep tendon reflexes, and an elevated resting pulse-rate (Claycomb, 1983). They can also be more sensitive to various types of painful stimuli, have a low exercise tolerance, and experience spontaneous fluctuations of galvanic skin response (Lader, Gelden, & Marks, 1967).

Brain imaging methods, especially positron emission tomography (PET) and single photon emission computerized tomography (SPECT) have been used to study blood flow, oxygen consumption, and receptors (Sadzot, Frost,

& Wgner, 1989; Innis et al., 1989). During an anticipatory anxiety state, activity in the bilateral temporal poles increased. They also occurred in lactate-induced anxiety states. These findings suggest that anxiety state is related to the function of the temporal cortex (Reiman, Fusselman, Fox, & Raichle, 1989; Reiman et al., 1989).

CONCLUSION

The pathophysiology of anxiety most likely involves the interactions between different brain neuronal systems. The GABA-ergic system associated with benzodiazepine receptors, the noradrenergic (NA) systems and the serotonergic systems are definitely involved in the biochemistry of anxiety. An interaction between the NA system and GABA/Benzodiazepine receptor system has been proposed (Redmond & Huang, 1979), and serotonin has been proposed to be involved in the anxiolytic properties of benzodiazepine (Paul, Marangos, & Skolnick, 1981). Insel, et al., (1984) suggested that there are two different neuropharmacological models of anxiety: NA and GABA-ergic pathways. The GABA-ergic system corresponds to “fear or conflict” or “psychic” manifestations of anxiety that often require benzodiazepine treatment. The NA activation corresponds to “alarm” or “autonomic” manifestations of anxiety such as panic attacks and these respond favorably to tricyclic antidepressant therapy. Anxiety induced by yohimbine, a noradrenergic a-2 receptor antagonist, has been successfully attenuated by

alprazolam, a triazolobenzodiazepine. This suggests that a yohimbine-induced anxiety state may be related to both NA and GABA activities (Charney, Breier, Jatlow, & Heninger, 1986). Also, other data suggest that the serotonergic pathway is involved in anxiety. At present, the role of other neurotransmitters such as acetylcholine, dopamine, histamine, adenosine, and neuropeptides, is not well understood; it appears to be minimal (Hoehn-Saric, 1982).

Of all the discussed hypotheses and methods for exploring the biochemistry of anxiety disorders, none is without flaws. As research activities continue, certain subtypes of pathological anxiety will be better understood by applying certain biochemical paradigms. These models can also help clinicians to identify specific biological etiologies for their patients which will promote an accurate diagnosis and effective treatment.

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