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β-Adrenoreceptor Coupling to G_s Protein in Alcohol Dependence, Panic Disorder, and Patients with Both Conditions

George N. M. Gurguis, M.D., Jukka Turkka, M.D., Ph.D., David T. George, M.D., and Markku Linnoila, M.D., Ph.D.

Ethanol may downregulate G-protein–coupled beta-adrenoreceptors (βAR). βAR may also be dysregulated in panic disorder (PD). In clinical samples, many patients have comorbid alcohol dependence (AD) and PD. Therefore, we investigated βAR coupling in patients with these disorders. We harvested polymorphonuclear leukocytes from 24 healthy volunteers (Vs), and from 22 abstinent AD patients, 7 PD patients, and 9 patients with comorbid AD/PD. βAR were assayed using saturation and agonist-displacement experiments. Group differences were tested using one-way analysis of variance (ANOVA). All βAR binding parameters were similar in AD patients and Vs. The ratio of the agonists' dissociation constant from the receptor in the low affinity state (K_L) to that in the high affinity state (K_H) was significantly higher in PD patients than in AD patients and Vs (930.97 ± 440.80 vs. 226.2 ± 94.47 vs. 197.05 ± 61.03, respectively, p < .01). This finding suggests that βAR are supercoupled to G_S in patients with PD. There was a trend for higher total receptor density (R_T) in AD/PD and PD patients (Vs = 39.06 ± 42.57 vs. AD = 27.93 ± 23.07 vs. AD/PD = 66.85 ± 79.02 vs. PD = 68.36 ± 49.20, p < .08). There were no differences between AD/PD and PD patients, who combined had a significantly higher R_T than Vs and AD patients (Vs = 38.95 ± 8.81 vs. AD = 29.63 ± 5.07 vs. AD/PD = 67.51 ± 17.00, fmol/mg protein, p < .04). Finally, AD/PD patients had a significantly higher receptor density in the low-affinity conformational state than Vs and AD patients, but not PD patients (25.96 ± 11.59 vs. 10.69 ± 1.53 vs. 7.62 ± 1.08 vs. 17.07 ± 5.26 fmol/mg protein, respectively, p < .005). βAR function in polymorphonuclear leukocytes is normal in abstinent alcoholics. The previously reported abnormal βAR regulation in alcoholism may be state dependent. The higher R_T and K_L/K_H ratio in AD/PD and PD, but not in AD patients, suggest that increased βAR function may be important in the pathophysiology of PD. ***[Neuropsychopharmacology 16:69–76, 1997]***

KEY WORDS: *Alcoholism; Panic disorder; β-Adrenoreceptors*

Clinical and epidemiological studies have found more than the expected overlap between anxiety disorders and alcoholism (Kushner et al. 1990; Klerman et al. 1991; Schuckit and Hesselbrock 1994). The lifetime prevalence of agoraphobia and panic disorder (PD) in alcoholic patients has been reported to be as high as 43% and 10%, respectively. Moreover 16% to 20% of PD patients have a history of alcohol abuse or alcohol-related problems (Cox et al. 1990; Klerman et al. 1991). Results of the recent national comorbidity survey have shown that anxiety disorders are chronic by nature. Furthermore, anxiety disorders and alcoholism are the most prevalent psychiatric disorders among men (Kessler et al. 1994). It is unclear whether anxiety disorders in alcoholics

From the Psychiatry Service (GNMG), Laboratory of Clinical Neuroscience and Autonomic Pharmacology, Department of Veterans Affairs Medical Center, Dallas, TX, USA; and the Department of Neurology (JT), University of Oulu, Oulu, Finland.

Address correspondence to: Markku Linnoila, M.D., Ph.D., Laboratory of Clinical Studies, DICBR National Institute on Alcohol Abuse and Alcoholism, NIH 10, Room 3C103, 10 Center Drive MSC 1256, Bethesda, MD 20892-1256.

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represent a true comorbidity or have evolved secondary to the abuse of alcohol. Recent work from our laboratory suggests, however, that a subgroup of patients with comorbid alcohol dependence (AD) and PD show a lower incidence of panic attacks in response to lactate infusion than patients with PD (George et al. 1989b). This preliminary finding suggests that patients with comorbid AD/PD may have distinct pathophysiological features that await further characterization.

Dysregulation of beta-adrenergic receptors (βAR) has been postulated to play a role in the pathophysiology of both anxiety disorders and alcoholism. Clinical studies have reported low plasma levels of cyclic adenosine monophosphate (cAMP) (Lykouras et al. 1988) and reduced lymphocyte cAMP responses to ethanol and isoproterenol in alcoholics (Diamond et al. 1987; Maki et al. 1990). Lymphocyte βAR density was found to be lower in 10 abstinent male alcoholics than controls, but increases in βAR density and basal and isoproterenol-stimulated cAMP responses were observed during ethanol withdrawal (Maki et al. 1990). The finding of reduced βAR density in abstinent alcoholics has not, however, been observed by all investigators (Bylund et al. 1984). In postmortem brain samples, total βAR density (R_T) in both abstinent and intoxicated alcoholics was not different from controls, but receptor density in the high-affinity state (R_H) in the cerebral cortex but not cerebellum from intoxicated alcoholics was low and compatible with βAR uncoupling from adenylate cyclase (AC) (Valverius et al. 1989a).

Animal studies on the effects of ethanol on βAR have found decreased R_T in brain and cardiac tissues from rats chronically treated with ethanol and a rebound increase in receptor density 48 to 72 hours after ethanol withdrawal (Banerjee et al. 1978). Valverius and colleagues (1987; 1988; 1989b) reported reduced density of βAR in the high-affinity conformation (R_H) and a decrease in the agonist's dissociation constant in cerebral cortical and hippocampal membranes of ethanol-fed mice in the absence of changes in R_T. The authors suggested that ethanol may promote βAR uncoupling from AC. These observations could not, however, be replicated in S49 lymphoma cell cultures (Bode and Molinoff, 1988a, 1988b) nor were they found in cerebral cortical, hippocampal, or cardiac cell membranes from rats chronically exposed to ethanol (Segel and Mason 1982; Rommelspacher and Strauss 1987; Turkka et al. 1990). Other lines of evidence suggest that chronic ethanol exposure induces postreceptor (heterologous) desensitization of AC-coupled systems that may partly contribute to decreased βAR function. Guanylyl-imido-diphosphate[Gpp(NH)p]-stimulated AC activity was bound to be lower in the cerebral cortical and hippocampal membranes from mice chronically fed ethanol and a fourfold dose-dependent increase in isoproterenol's EC_{50} for stimulating AC was reported in S49 lymphoma cell cultures chronically exposed to ethanol (Rommelspacher and Strauss 1987; Saito et al. 1987; Bode and Molinoff 1988a; Valverius et al. 1989b). Furthermore, a 38.5% decrease in the α subunit of the G_S protein and 30% decrease in the α_S-mRNA was observed in the NG 108-15 glioma × neuroblastoma hybrid cell cultures chronically treated with ethanol (Mochly-Rosen et al. 1988). Therefore, it is likely that ethanol affects AC-coupled βAR function through mechanisms that involve both the membrane receptor and intracellular signal transduction.

As in alcoholism, low lymphocyte βAR density has been reported in PD (Brown et al. 1988; Maddock et al. 1993), and heart rate responses to isoproterenol infusion were observed to be low in PD patients (Nesse et al. 1984), as were cardiovascular responses in rats chronically exposed to alcohol (Segel et al. 1979). Contradicting these studies, high lymphocyte βAR density has also been reported in PD (Albus et al. 1986). The reports of decreased βAR function in PD and AD patients are inconsistent with the symptomatology of either panic attacks or somatic anxiety symptoms in alcoholism (George et al. 1988), which are more compatible with βAR supersensitivity (Frohlich et al. 1970).

So far, evidence of altered βAR function in patients with either AD or PD remains inconclusive. Furthermore, although decreased βAR function may be common in both AD and PD, mechanisms underlying the postulated decrease of βAR function have not been clarified and may differ among the disorders. Previous clinical studies of βAR regulation in both AD and PD have employed saturation experiments with antagonists as ligands. Hence, the only meaningful variable that has been quantified is R_t, as the physiological relevance of the antagonists' dissociation constants from the receptor remains unknown. The rationale for the present study was the hypothesis that shifts in the distribution of the receptor fractions in the high- and low-affinity states may occur even in the absence of observable changes in R_T. Because antagonist binding does not differentiate between these physiologically important affinity states (Weiland et al 1979; Kent et al. 1980), agonist displacement studies are necessary to investigate the physiological state of βAR coupling. In this study we investigated βAR coupling in patients with AD, PD, and comorbid AD and PD using both antagonist-saturation and agonist-displacement experiments. Consequently, we were able to quantify receptor density in the high- and low-affinity states (R_H and R_L, respectively) and the agonist's dissociation constant from both affinity states (K_H and K_L, respectively). Measures of receptor coupling to G_S-protein and AC were defined as the percentage of receptors in the high-affinity state (% R_H) and the K_L/K_H ratio. Both the % R_H and the K_L/K_H ratio have been shown to correlate with the agonist's intrinsic activity and with receptor coupling to G_S-protein and AC (Davies and Lefkowitz 1980, 1981; Kent et al. 1980). Hence, these variables can

There was no statistically significant difference in K_d between the four groups (Vs = 30.49 ± 23.21; AD = 29.19 ± 19.28; AD/PD = 22.92 ± 13.74; PD = 40.75 ± 16.59 pM, $F = 1.06$, p = NS). There was a trend for a difference in R_T (Vs = 38.95 ± 8.81; AD = 29.63 ± 5.07; AD/PD = 76.12 ± 35.66; PD = 66.68 ± 16.84 fmol/mg protein; $F = 2.59$, $p < .06$). Post hoc comparisons showed no statistically significant differences between any of the three patient groups in comparison to volunteers. However, AD/PD patients had significantly higher R_T than patients with AD. Analyses performed on log-transformed data yielded similar results.

Results of agonist, R_H, R_L, R_T, and % R_H are summarized in Table 1. Significant group differences were observed in R_L only. Post hoc analysis showed that patients with AD/PD had higher R_L than AD patients and Vs. After log transformation, only the difference between AD/PD patients and healthy volunteers remained significant ($t = 2.145$, $p = 0.04$).

Results of the agonist dissociation constants K_L, K_H, and the K_L/K_H ratio are summarized in Table 2. No group differences were observed in either K_L or K_H. Significant group differences were observed in the K_L/K_H ratio, which was due to a higher K_L/K_H ratio in PD patients than in Vs and AD patients, but not AD/PD patients. Analyses performed on log-transformed data yielded similar results.

Since there were no significant differences in any binding parameters between AD/PD and PD patients, and because of the small number of subjects in these two groups, we repeated the ANOVA comparing Vs to AD and PD (with or without AD) patients. This analysis showed that PD patients had a significantly higher R_T (overall test: $F = 3.37$, $p < .04$) than AD patients ($p < .05$, Bonferroni-corrected) and a trend towards higher R_T than in Vs ($p < .058$ not Bonferroni-corrected). The K_L/K_H ratio also was significantly higher (overall test: $F = 5.26$, $p < .008$) in the PD than AD patients ($p < .05$, Bonferroni-corrected) and Vs ($p < .05$, Bonferroni-corrected). Log transformation did not change the levels of statistical significance observed in the analyses of the non-transformed data on R_T and the K_L/K_H ratio. Results of the analyses of the other binding parameters were similar to the results observed when the four groups were compared.

In summary, abstinent AD patients did not show abnormalities in any of the parameters gauging βAR regulation, whereas PD patients showed evidence of βAR upregulation and supercoupling to the G_S-protein.

DISCUSSION

The results show no evidence of abnormal βAR regulation in abstinent AD patients, particularly in terms of coupling to G_S. On the other hand, PD patients showed evidence of βAR upregulation and supercoupling to the G_S protein that might contribute to symptoms of apparent sympathetic overactivity during panic attacks. Patients with comorbid disorders had higher R_L and R_T than AD patients, and their K_L/K_H ratio fell between those of AD and PD patients, suggesting that this subgroup of patients may have a distinct pathophysiology. To our knowledge, this is the first report to examine parameters of βAR coupling to G_S protein, βAR density quantified with an agonist, and agonist dissociation constant from the receptor in the high- and low-affinity conformational states in alcoholism and in PD.

Our observations on βAR binding parameters in AD suggest that a history of heavy ethanol exposure has no effect on neutrophil βAR function in vivo. Thus, the previously reported effects of ethanol on βAR may be state dependent, and abnormal βAR regulation is likely to be observed in alcoholics only during the active phase of drinking or during withdrawal but not during prolonged abstinence. It is noteworthy that our AD patients underwent supervised abstinence on a closed research ward for 3 weeks prior to participating in this study. Consequently, it is likely that the postulated pathophysiological processes associated with alcoholism [such as downregulation of R_T, receptor uncoupling, decrease in α_S-mRNA, or abnormal G_S-protein/AC interaction in alcoholics (Saito et al. 1987; Bode and Molinoff 1988a; Mochly-Rosen et al. 1988; Valverius et al. 1989b; Maki et al. 1990) represent reversible or short-lived effects of alcohol]. In this regard, the present findings parallel

Table 1. β–Receptor Density in the High-(R_H) and Low-Affinity (R_L) States, Total Receptor Density (R_T) and the Percentage of Receptors in the High-Affinity State (% R_H)

	Vs (n = 24)	AD (n = 22)	AD/PD (n = 9)	PD (n = 7)	F Statistic	p Value
R_H	28.25 ± 8.06	22.01 ± 4.32	50.17 ± 24.84	49.60 ± 12.11	1.744	NS
R_L	10.69 ± 1.53	7.62 ± 1.08[a]	25.96 ± 11.59[b]	17.07 ± 5.26	4.730	.005
R_T	38.95 ± 8.81	29.63 ± 5.07	76.12 ± 35.66[c]	66.68 ± 16.84	2.59	.06
% R_H	65.16 ± 3.38	70.11 ± 2.55	61.51 ± 8.19	71.65 ± 6.00	0.881	NS

Values represent means ± SEM. Receptor density is expressed as fmol/mg protein.
[a] $p < .005$ compared to AD/PD patients (Bonferroni–corrected).
[b] $p < .02$ compared to Vs (Bonferroni-corrected).
[c] $p < .04$ compared to AD patients (not Bonferroni–corrected).

elucidate sites of regulation at the interface between βAR and intracellular signal transduction mechanisms in a physiologically meaningful way.

METHODS

Subjects

Twenty-four healthy volunteers (Vs; 20 males, 4 females) and 22 patients (20 males, 2 females) who met both DSM-III-R (American Psychiatric Association 1987) and research diagnostic criteria (Spitzer et al. 1978) for AD, 9 patients (8 males, 1 female) with comorbid AD/PD, and 7 patients with PD (5 males, 2 females) participated in this study. To determine the diagnosis, all subjects received a comprehensive psychiatric interview (DTG). Physical examination, laboratory workup (CBC, Chem 24, TFTs, UA), and electrocardiograms were all normal. All patients and controls were medication free for a minimum of 3 weeks. No patient had received monoamine oxidase or serotonin uptake inhibitors prior to participating in the study. Patients with AD and AD/PD were abstinent from alcohol for 3 weeks (ascertained by random breath testing on a locked research ward). All subjects had a negative urine drug screen at the time of admission. They adhered to a low-monoamine, low-caffeine, alcohol-free diet for 3 days prior to the blood sampling. All subjects signed a written informed consent approved by the NIAAA DICBR Institutional Review Board before participating in the study.

Procedures

Blood was sampled on the inpatient unit of the NIAAA in the morning after a night's sleep. Subjects laid supine in a hospital bed. An intravenous line was started (between 7:00 and 8:00 A.M.) and kept open using a slow drip of physiological saline solution. After a 60-minute rest period, when the heart rate had returned to stable resting levels, a 55-ml blood sample was drawn on heparin for the receptor assays.

β-Adrenoreceptor Membrane Preparation and Binding Assay. Fresh membranes were prepared, and radioligand binding assays were conducted according to Davies and Lefkowitz (1980, 1981). In summary, blood was mixed with 20 mL of 3% dextran and 9% saline solution and allowed to stand at room temperature for 30 minutes. The plasma-dextran supernatant was then subfused with Ficoll-Paque (Pharmacia, Biotechnology) and centrifuged at 500 × g. The erythrocyte-neutrophil pellet was washed in 0.9% saline solution and erythrocytes were lysed using cold deionized water. Neutrophils were recentrifuged at 300 × g at 4°C, resuspended, and homogenized by a polytron for 12 s. A pure membrane preparation was obtained by spinning the supernatant resulting from a 1,900 × g initial centrifugation at 41,000 × g at 4°C. Membranes were resuspended in the incubation buffer for a final protein concentration of 10 to 20 mg/mL.

^{125}[I]Iodocyanopindol (ICYP; Sp act 2,200 Ci/mmol) was used as the radioligand. Saturation experiments used seven different concentrations (3–30 pmol) to quantify the antagonist, ICYP, affinity (K_d) and R_T. Nonspecific binding was detected in the presence of 0.1 mmol isoproterenol. Specific binding was 75% to 80% of the total binding. Displacement experiments were performed with 15 different concentrations of isoproterenol (1 nM to 1 mM) to displace ICYP binding (12 pM) and characterize the binding properties of the β-agonist. The tubes were incubated at 37°C for 40 minutes. The binding reaction was terminated by adding 20 mL of wash buffer (75 mm Tris, 25 mM $MgCl_2$, pH 7.65), and membranes were harvested over Whatman GF/C glassfiber filters. Dried filters were placed in polystyrene tubes and loaded on a gamma counter (Beckman 5500) with 75% counting efficiency.

Analysis of Binding Data

Binding data from saturation and displacement experiments were analyzed using LIGAND (Munson and Rodbard 1983); an iterative nonlinear curve–fitting program. The K_d and R_T were measured from saturation experiments. Data from the displacement experiment were tested for the presence of a two-site model, which was accepted only if its goodness of fit was better than that of a one-site model (F test, $p < .05$). The R_H, R_L, K_H and K_L were measured from displacement experiments, and the % R_H and the K_L/K_H ratio were computed.

Statistical Analysis

We used a one-way analysis of variance (ANOVA) to compare the four groups of subjects and computed post hoc analyses when the p value for the overall ANOVA was statistically significant or showed a trend (< .10) toward significance. To reduce the risk of statistical artifacts due to unequal variances we computed a second set of confirmatory analyses using log-transformed data.

RESULTS

There was no difference in age between the four groups (Vs, AD, AD/PD, and PD patients; 36.54 ± 9.08 vs. 36.45 ± 9.95 vs. 40.90 ± 10.19 vs. 38.57 ± 6.29 years, respectively, F = 0.647, df = 3,59; p = NS). There was no significant difference in gender distribution across the four groups ($\chi^2 = 1.742$, df = 1,3, p = NS). Both saturation and displacement experiments were available from all subjects except for three displacement experiments that had technical problems.

Table 2. Agonist's (ISO) Dissociation Constants from βAR in the Low (K_L) and High-Affinity (K_H) States and the K_L/K_H Ratio

	Vs (n = 24)	AD (n = 22)	AD/PD (n = 9)	PD (n = 7)	F Statistic	p Value
K_L	32.95 ± 14.49	36.73 ± 19.07	20.37 ± 10.64	65.97 ± 28.21	0.453	NS
K_H	66.40 ± 20.45	79.08 ± 28.54	24.14 ± 6.24	70.51 ± 28.79	0.408	NS
K_L/K_H	197.05 ± 61.03	226.21 ± 94.47[a]	591.86 ± 273.90[b]	930.97 ± 440.80[c]	3.928	.01

Values are means ± SEM. K_L is expressed as μM, K_H is expressed as nM.
[a] $p < .05$ compared to PD (Bonferroni-corrected).
[b] $p < .03$ compared to Vs (not Bonferroni-corrected).
[c] $p < .05$ compared to Vs (Bonferroni-corrected).

results of Valverius et al. (1989a), which were obtained by investigating cerebral cortical membranes harvested postmortem from alcoholics. Analogous findings have been reported regarding blunted α_2-adrenoreceptor–mediated physiological responses during alcohol withdrawal that also are normalized during abstinence (Glue et al. 1988). The normal βAR function in the polymorphonuclear leukocytes of abstinent alcoholics does not preclude the possibility that trait-related differences in intracellular AC-coupled signal transduction systems may exist in alcoholics compared to healthy volunteers. For example, it would be of interest to study further the adaptational mechanisms of these systems in AD patients in response to an acute ethanol challenge. The results of this study, therefore, may not be at odds with the findings of others (Diamond et al. 1987; Maki et al. 1990) that reflect βAR function in inebriated alcoholics or during withdrawal. Indeed, one group (Maki et al. 1990) found no difference in receptor density beyond the first day of withdrawal. In that study, the use of chlordiazepoxide to ameliorate withdrawal symptoms could, however, have accelerated the return of βAR function to normal, possibly because of reduced peripheral epinephrine and norepinephrine levels produced by the benzodiazepine. Another group (Bylund et al. 1984) failed to demonstrate differences in βAR density either in neutrophils or lymphocytes in 10 alcoholic patients (only four of whom had measurable blood ethanol levels), further supporting our notion that ethanol-induced βAR dysregulation may be observed only during intoxication and early abstinence.

In PD patients R_T was 71% higher than in Vs. Similarly in AD/PD patients, R_T was 95% higher than in Vs. When the two groups were combined R_T was significantly higher than in both AD patients and Vs. Furthermore, PD patients had a significantly higher K_L/K_H ratio than did AD patients and Vs, suggesting βAR supercoupling to G_S-protein and AC. The K_L/K_H ratio also was 200% higher in the AD/PD patients than in Vs. When the PD and AD/PD groups were combined, their K_L/K_H ratio was significantly higher than in both AD patients and Vs. These observations suggest that high R_T and K_L/K_H ratio may play a role in the pathophysiology of PD and provide evidence of increased βAR function both at the level of receptor density and receptor coupling to G_S-protein (i.e., at the level of intracellular signal transduction). The higher R_T and K_L/K_H ratio at baseline also are consistent with the therapeutic efficacy of tricyclic antidepressants in PD. These medicines downregulate βAR density and uncouple the βAR from G-proteins (Sulser et al. 1984; Hancock and Marsh 1985; Okada et al. 1986; Fishman and Finberg 1987; Manier et al. 1987).

At the molecular level, βAR supercoupling may explain the hyperdynamic β-adrenergic state during panic attacks (Frohlich et al. 1970). βAR from polymorphonuclear leukocytes (PMN) or lymphocytes are of the β_2AR subtype and have similar pharmacological characteristics to those on peripheral organs (i.e., heart, lungs, vasculature) whose activity is involved in mediating many somatic symptoms associated with anxiety (French et al. 1976; Brown et al. 1985; Brodde et al. 1986). The majority of studies on PD have reported normal plasma catecholamine levels at baseline and during panic attacks (Ballenger et al. 1984; Cameron et al. 1984, 1990; Nesse et al. 1984; Albus et al. 1986; Schneider et al. 1987; Gurguis et al., 1991; Stein et al. 1992). Yet, PD patients have exaggerated cardiovascular responses to a variety of physiological or pharmacological challenges (Freedman et al. 1985; Taylor et al. 1986; Shear et al. 1987; Yeragani et al. 1990; Stein et al. 1992). Exaggerated cardiovascular responses in the presence of normal catecholamine levels in PD patients strengthen our inference and support peripheral postsynaptic adrenergic receptor supersensitivity as a possible explanation. More important at the molecular level, our finding of βAR supercoupling to G_S-protein may represent one of the underlying mechanisms of enhanced cardiac sympathetic tone that has been implicated in ventricular tachyarrhythmias sometimes associated with psychologic distress and the finding that β-blockers have a marked impact on both arrhythmias and the somatic symptoms of anxiety (Brodsky et al. 1987); the widely reported increased incidence of cardiovascular disease and mortality in patients with panic disorder (Coryell et al. 1982, 1986; Wissman et al. 1990; Allgulander and Lavori 1991); and a recent report documenting anxiety as the highest risk factor for developing fatal coronary artery disease and sudden cardiac death (Kawachi et al. 1994).

Of course, exaggerated cardiovascular responses in PD patients may partly reflect disturbances in the balance between the cardiac sympathetic and parasympathetic inputs. For example, a previous report from our laboratory suggested that decreased parasympathetic vagal input to the heart may account for some of the cardiac responses during panic attacks (George et al. 1989a). βAR supercoupling could explain increased cardiovascular responses to the reduction of vagal tone in patients with PD, even without a change in the presynaptic release of norepinephrine. Our finding of increased β_2AR density is consistent with observations of some (Albus et al. 1986), but not all, reports on PD (Brown et al. 1988; Maddock et al. 1993). Inconsistencies between reports may, however, be partly attributable to the fact that almost 50% of the patients in one study (Brown et al. 1988) had concurrent or a recent history of major depression. Low βAR density or function have been reported in the majority of studies on depression (Carstens et al. 1987; Ebstein et al. 1988; Halper et al. 1988; Pandey et al. 1989).

Our observations in AD/PD patients may suggest a pattern of βAR dysregulation that is different from patients with either AD or PD alone. For example, similar to PD patients, they had higher R_T than AD patients. However, unlike PD patients, AD/PD patients had higher R_L than Vs and AD patients. Finally, the K_L/K_H ratio of AD/PD patients fell along a continuum from Vs to AD to PD patients. A two-step process has been proposed in the regulation of βAR in response to agonist stimulation: phosphorylation and/or uncoupling into a low-affinity state followed by downregulation, internalization into the cytoplasm and later receptor degradation (Lefkowitz et al. 1983, 1984, 1990; Sibley and Lefkowitz 1985, 1987; Motulsky et al. 1986; Sibley et al. 1987). It is, therefore, possible that the increase of βAR density in the low-affinity conformation state may represent an adjustment to repeated intermittent stimulation associated with panic attacks. A similar trend for an increase of R_L was also seen in PD patients. Alternately, the increase of R_L in this group of patients may be due to lingering effects of ethanol. It has been reported to promote β_2AR uncoupling from AC and to shift βAR to a low-affinity conformational state (Valverius et al. 1987; 1988; 1989b). It is worth noting that although high K_L/K_H ratio is driven by a high K_L in PD, this ratio is driven by a low K_H in the AD/PD patients. A high K_L may suggest a posttranslational modification of the receptor, but the low K_H suggests an enhanced βAR-G_S interaction (Ostrowski et al. 1992). This interpretation remains preliminary given the small number of subjects in each group. Collectively, these observations support the notion that AD/PD patients may have a distinct pathophysiology that differs from that of patients with either disorder alone, as suggested by a lower responsiveness to a lactate infusion than in PD patients (George et al. 1989b).

In summary, the results of this investigation showed that β_2AR binding parameters in abstinent AD patients were not different from those found in Vs and that PD patients had higher β_2AR density and supercoupling of receptors to G_S-protein and AC. The data also suggest the possibility of a complex pattern of interaction between the pathophysiology of PD and AD. Further exploration of regulatory mechanisms controlling G_S-proteins and AC activity is warranted in mental disorders, alcoholism, and other substance abuse.

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