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Regional Brain Metabolic Response to Lorazepam in Subjects at Risk for Alcoholism

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The mechanisms underlying the blunted response to alcohol administration observed in subjects at risk for alcoholism are poorly understood and may involve GABA-benzodiazepine receptors. The purpose of this study was to investigate if subjects at risk for alcoholism had abnormalities in brain GABA-benzodiazepine receptor function. This study measured the effects of 30 μg/kg (iv) of lorazepam, on regional brain glucose metabolism using positron emission tomography and 2-deoxy-2[^{18}F]fluoro-D-glucose in subjects with a positive family history for alcoholism (FP) (n = 12) and compared their response with that of subjects with a negative family history for alcoholism (FN) (n = 21). At baseline, FP subjects showed lower cerebellar metabolism than FN. Lorazepam decreased whole-brain glucose metabolism, and FP subjects showed a similar response to FN in cortical and subcortical regions, but FP showed a blunted response in cerebellum. Lorazepam-induced changes in cerebellar metabolism correlated with its motor effects. The decreased cerebellar baseline metabolism in FP as well as the blunted cerebellar response to lorazepam challenge may reflect disrupted activity of benzodiazepine-GABA receptors in cerebellum. These changes could account for the decreased sensitivity to the motor effects of alcohol and benzodiazepines in FP subjects.

Key Words: Benzodiazepines, Positron Emission Tomography, Brain Glucose Metabolism, Alcoholism, Cerebellum.

THE IMPORTANCE of genetics in alcoholism has been demonstrated in epidemiological studies showing a higher level of concordance for alcoholism in identical twins than in fraternal twins and by studies documenting a 4-fold increased risk for alcoholism in children of alcoholics than in the general population.[1–5] The influence of genetics in the differential sensitivity to ethanol has also been demonstrated in studies documenting that subjects at risk for alcoholism, that is children of alcoholics who themselves are not alcoholics, show a blunted response to alcohol when compared with subjects with a negative family history for alcoholism (FN).[2,6] A blunted response to alcohol in family-positive subjects (FP) has been documented for the subjective experience of intoxication,[2,7,8] motor impairment,[9,10] neurohormonal changes,[11] and electrophysiological responses.[6,12,13] The relevance of genetics on the sensitivity to ethanol was also demonstrated in a study of fetal human tissue that showed that sensitivity to ethanol was variable and depended on the source of the donor tissue; grafts that were made from the same host all showed comparable sensitivities.[14]

The mechanisms underlying differences in sensitivity to alcohol could relate to pharmacokinetic factors (metabolism and bioavailability) and/or neurochemical differences of the brain. These variables in turn could modulate the reinforcing and toxic properties of alcohol, thus increasing or decreasing the likelihood of its abuse. The GABA-benzodiazepine receptor complex (GBRC) has been implicated as one of the molecular substrates underlying the differential sensitivity to the effects of alcohol.[15] Because the pharmacological actions of alcohol seem to be in part mediated by its facilitation of GABAergic neurotransmission at the GBRC,[16–18] one could postulate that the decreased sensitivity to alcohol in subjects at risk for alcoholism could reflect abnormal activity of the GBRC. In support of this hypothesis is a study documenting decreased sensitivity in FP subjects to the behavioral and eye movement effects of benzodiazepines.[19] Benzodiazepines, like alcohol, exert some of their pharmacological effects via the GBRC.[16]

Positron emission tomography (PET) is an imaging method that allows direct noninvasive measurement in the living human brain of metabolism, neurochemistry, pharmacology, and perfusion.[20] Brain glucose metabolism, typically measured with 2-[^{18}F]fluoro-D-glucose (FDG), is the most frequently applied measurement with PET and provides an index of regional brain function.[21] Regional brain metabolic measurements after acute and chronic drug administration have been used as indicators of the functional effects of drugs in the human brain. Acute alcohol administration was found to decrease regional brain glucose metabolism in a pattern that paralleled the regional distribution of benzodiazepine receptors in the human brain.[22] The regional metabolic response to acute[23,24] and chronic benzodiazepine agonists[25] have also been measured with PET. As for alcohol, the regional metabolic response to benzodiazepines paralleled the regional concentration of benzodiazepine receptors in human brain.[25] Lorazepam-induced

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changes in regional metabolism were reversed by the benzodiazepine antagonist flumazenil,[26] which indicates that benzodiazepine-induced metabolic changes reflect in part benzodiazepine receptor activity. We have therefore proposed the use of lorazepam as a pharmacological challenge to study the functional responsivity of the GBRC.[24] Using this strategy, we have shown that alcoholics have a blunted regional brain metabolic response to lorazepam that may be indicative of a decreased sensitivity of GBRC. Because the blunted brain metabolic response in alcoholics could reflect the chronic use of alcohol as opposed to a genetic trait associated with alcohol predisposition, we investigated a group of subjects with a family history of alcoholism. In this study, the regional brain metabolic response to lorazepam in FP subjects is compared with that of FN subjects.

MATERIALS AND METHODS

Subjects

Subjects at risk for alcoholism (family-positive) consisted of 12 right-handed males (30.5 ± 5 years of age) who had an alcoholic biological father (as per DSM-III-R) and at least two other first (parent or sibling)- or second-degree relatives (biological grandparents, uncles, aunts, or cousins) who were alcoholics. Subjects were excluded if they had a past or present history of abuse or dependence to alcohol and/or other drugs of abuse (except nicotine and caffeine) or any other DSM-III-R diagnosis for mental illness and/or neurological illnesses in themselves or in a first-degree relative (except for family history of alcoholism), if they had medical illnesses and/or if they were taking any medication. Nondrinkers (consumes alcohol less than once every year) were also excluded.

Controls (family-negative) consisted of 21 right-handed, healthy males 32.5 ± 10 years of age (age range 23–59) who had served as controls for a study that compared the response to lorazepam in normals and alcoholic subjects.[24] Subjects were excluded if they had a family history of alcoholism or drug abuse in first- and/or second-degree relatives. Otherwise, criteria were as for the subjects at risk. Right-handed subjects were elected to minimize variability from laterality. Table 1 provides demographic data for the two groups of subjects.

As part of the evaluation procedure, subjects had a physical, psychiatric, and neurological examination. Routine laboratory tests were performed as well as a random urine test to exclude the use of psychoactive drugs. Subjects were instructed to discontinue any over-the-counter medication 2 weeks before the PET scan and to refrain from drinking alcohol the week before the PET scan. Cigarettes, food, and beverages (except for water) were discontinued at least 4 hr before the study. This study was approved by the Human Subjects Research Committees of Brookhaven National Laboratory and the Northport Veterans Administration Medical Center. After explaining the procedure, written informed consent was obtained from each subject.

Table 1. Demographic Characteristics of Subjects

	FN (n = 21)	FP (n = 12)
Age	30.5 ± 5	32.5 ± 10
Education (yr)	15 ± 3	14 ± 2
Smokers	6	3
Alcohol use	3–4 ± 2	5–6 ± 3

Alcohol use corresponds to beers consumed/week. Groups did not show significant differences in years of education completed, in amount of alcohol consumed, or in percentage of smokers. Differences in caffeine intake could not be assessed, because they were not recorded.

PET Studies

PET measurements were conducted using a whole-body, high-resolution positron emission tomograph (6.5 mm × 5.9 mm full-width half-maximum at the center of the field of view, interslice distance 5.9 mm, 15 slices; Computer Technologies, Inc., CTI 931). Subjects were positioned in the gantry using an individual headholder and two sets of weak laser fan beams that illuminated the head surface along the canthomeatal line and along the sagittal line, respectively. Before radiotracer injection, each subject underwent a transmission scan performed with a ring filled with germanium 68/gallium 68 to allow the subsequent emission image to be corrected for attenuation. A catheter was placed in the antecubital vein for injection of radiotracer, lorazepam, and placebo and in a dorsal hand vein for "arterialized" blood sampling. The emission scans were taken 35 min following injection of 4–6 mCi of FDG (prepared according to the method of Hamacher[27]) for a total of 20 min. Arterialized blood was obtained throughout the procedure to measure plasma concentration of F-18, glucose, PO_2, pCO_2, and plasma lorazepam concentration (National Psychopharmacological Laboratory). For the first scan, subjects were injected with a placebo (3 ml of saline solution) given 40–50 min before FDG. For the second scan, subjects were injected with 30 μg/kg of lorazepam given 40–50 min before FDG. Subjects were blind to the drugs received. To ensure that the subjects would not fall asleep, they were monitored throughout the procedure and were asked to keep their eyes open. Subjects were scanned with their ears unplugged in a dimly lit room with noise kept to a minimum. The only intervention was the periodic assessment of the behavioral and cognitive effects of lorazepam or placebo. A detailed description of procedures, including calculation of metabolic "rates," has been published.[24] Each subject underwent two different PET procedures.

Behavioral, Cognitive, and Motor Evaluation

Before placebo or lorazepam and at 20 min, 60 min, and 2 hr after placebo or lorazepam administration, subjects were asked to evaluate on an analog scale (rated 0–100) their subjective sense of intoxication, desire for more drug, tiredness, and sleepiness, and were also evaluated with the Stroop Test, the Word Association test, Symbol Digit Modality test, and arithmetic calculations.[28] Before the scan, and at the end of the study, subjects were evaluated for motor coordination. Motor changes induced by lorazepam were rated with respect to baseline from 0 to 10 by scoring the following items: gait, rhythm (tapping the back and front of the hand with the other hand), equilibrium (eyes closed), rhomberg and subjective perception of incoordination as 0 = absent, 1 = minimal disruption, and 2 = marked changes. At the end of the study, subjects were asked whether they perceived the drug intervention (placebo or lorazepam) as pleasurable, neutral, or unpleasant. Blood samples were taken to quantify lorazepam concentration 20 min, 60 min, and 2 hr after its administration.

Image Analysis

Regions of interest (ROIs) were drawn directly on the transaxial PET images using the Matsui/Hirano atlas as reference.[29] Seventy-two ROIs were selected from 10 of the 15 images obtained with the tomograph. Weighted averages (to correct for difference in sizes) of the ROIs from different slices corresponding to the same anatomical areas were computed to obtain metabolic values in 10 "composite" brain regions. The location of the ROIs sampled and the ROIs that were included to obtain the 10 "composite" brain regions have been published.[24] A measure of "whole brain" metabolism was obtained by averaging metabolism in the 15 brain slices. "Relative" measures of regional brain metabolism were obtained using the ratio of the metabolic value in the "composite" brain regions to the metabolic value for the whole brain.

Statistical Analyses

Differences in regional metabolism between FP and FN subjects were tested with ANOVA. Differences in response to lorazepam between FP

Table 2. Behavioral and Cognitive Measures Obtained after Placebo and Lorazepam

	FN (n = 21)		FP (n = 12)	
Test	Placebo	Lorazepam	Placebo	Lorazepam
Desire	2.5 ± 7	3.5 ± 8	3.2 ± 1	3.6 ± 11
Intoxication	0.5 ± 1	35.2 ± 26	5.3 ± 8**	28.9 ± 18
Sleepiness	1.5 ± 5	57.0 ± 25	18.8 ± 20**	51.8 ± 21
Tiredness	14.0 ± 7	44.4 ± 29	17.5 ± 17	41.0 ± 19
Stroop-read	102.6 ± 14	77.4 ± 14	96.1 ± 19	73.2 ± 22
Stroop-XXX	74.0 ± 11	68.2 ± 17	64.36 ± 15**	53.8 ± 18*
Stroop-color	47.7 ± 9	40.7 ± 12	38.6 ± 8.5**	34.1 ± 10
SDMT	48.6 ± 4	36.2 ± 9	43.8 ± 9	32.7 ± 9
WA	15.1 ± 2	10.6 ± 5	12.9 ± 4	8.7 ± 5
Calculation	12.1 ± 1	10.6 ± 2	12.3 ± 2	10.9 ± 2
Motor	0	6.6 ± 3	0	4.6 ± 3*

Subjects were asked to evaluate in an analog scale (0–100) their desire for more drug, sleepiness, tiredness, and subjective sense of intoxication. Cognitive tests involved the Stroops [separate values are reported for the three sections: reading color names (read), describing the color (XXX), and reading color names colored with discrepant colors (color)], Symbol Digit Modality test (SDMT), word association (WA), and arithmetic calculations. Lorazepam impaired performance in both groups of subjects. Significance is only shown for those values where the two groups differed.

* $p < 0.05$; ** $p < 0.01$.

Table 3. Absolute Metabolic Values for the Different Brain Regions after Placebo and Lorazepam

	FN (n = 21)		FP (n = 12)	
Regions	Placebo	Lorazepam	Placebo	Lorazepam
Global	38.2 ± 3	33.2 ± 3	38.3 ± 4	34.3 ± 3
R prefrontal	45.0 ± 5	39.7 ± 5	44.0 ± 4	40.8 ± 3
L prefrontal	45.8 ± 4	39.5 ± 5	45.4 ± 4	41.5 ± 3
R frontal	54.5 ± 5	47.0 ± 5**	52.0 ± 5	46.5 ± 4
L frontal	55.1 ± 5	47.5 ± 6**	52.2 ± 5	46.9 ± 4
Cingulate G	52.6 ± 5	46.1 ± 5	50.1 ± 6	47.8 ± 4
R parietal	51.2 ± 4	45.0 ± 4**	49.1 ± 5	44.01 ± 5
L parietal	51.2 ± 2	44.8 ± 6**	49.6 ± 5	44.9 ± 3
R temporal	50.1 ± 4	45.6 ± 4**	49.9 ± 4	45.9 ± 4
L temporal	50.4 ± 5	45.3 ± 6**	49.1 ± 6	45.7 ± 4
Occipital	54.9 ± 6	44.4 ± 5**	52.8 ± 7	43.9 ± 5
Thalamus	52.7 ± 5	40.3 ± 4**	55.9 ± 8	44.7 ± 4
Basal ganglia	52.0 ± 6	45.3 ± 6**	51.8 ± 8	46.9 ± 3
R cerebellum	42.8 ± 5	36.7 ± 3	39.7 ± 5*	37.4 ± 5
L cerebellum	43.8 ± 4	36.5 ± 3	40.3 ± 5*	35.4 ± 3

Lorazepam significantly reduced brain glucose metabolism in all brain regions. (Significance is reported for comparison of baseline measures between FP and FN subjects, and for the magnitude of lorazepam-induced changes in regional brain.

* $p < 0.05$; ** $p < 0.001$.

and FN subjects were tested with a one-factor (FP versus FN) repeated measure (baseline versus lorazepam) ANOVA. Pearson product-moment correlation analyses were performed between the behavioral and the regional metabolic changes after lorazepam. Changes were computed by subtracting the baseline scores from the scores obtained during lorazepam administration.

In consideration of the "multiple comparison problem" incurred by analyzing values for 10 brain regions, we set the level of significance to $p \leq 0.01$. We chose this criterion of significance as being intermediate between the $p \leq 0.05$ value, considered significant for an individual variable and the $p < 0.005$ value required by the Bonferroni adjustment, because Bonferroni criterion assumes that variables are independent,[30] but regional metabolic values are highly dependent on one another.[31] Values of $p \leq 0.05$ are reported as trends.

RESULTS

Plasma lorazepam concentrations at 20, 40, 70, and 90 min postinjection for the FP subjects corresponded to 40.2 ± 9, 30.2 ± 5, 27.9 ± 9, and 29.6 ± 9, ng/ml, respectively, and were not significantly different from those of the FN that corresponded to 41.3 ± 12, 31.9 ± 10, 28.3 ± 6, and 28.1 ± 6 ng/ml, respectively. The behavioral, cognitive, and motor changes after lorazepam for the subjects at risk and for the normal controls are shown in Table 2. At baseline, both groups of subjects differed with respect to their scores in the Stroops and also in their subjective experience of intoxication after placebo. FP showed lower scores in the color subtest of the Stroops and had significantly higher ratings for the subjective experience of intoxication after placebo. In both groups of subjects, lorazepam significantly induced sleepiness and tiredness, impaired motor coordination, and affected cognitive performance in all tests except that of arithmetic calculations. The effects were comparable for both groups of subjects except for the motor incoordinating effects of lorazepam that showed a trend toward less of an effect in FP than in FN ($F = 4.22, df = 31, p < 0.05$) and for sleepiness, which also showed a trend toward less of an effect in FP than in FN subjects ($F = 4.78, df = 31, p < 0.05$). Lorazepam was experienced as pleasant by 43% of the FN and 58% of the FP, and as unpleasant by 24% of the FN and 25% of FP. The rest of the subjects described the experience as neutral.

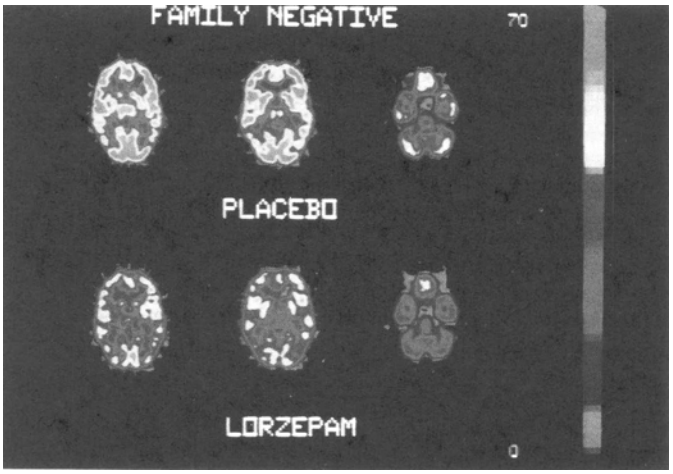

Fig. 1. Metabolic images from a FN subject tested after placebo and after lorazepam (30 μg/kg iv). Note the decrease in glucose metabolism in the cortical, subcortical, and cerebellar regions.

Whole-brain metabolic activity at baseline did not differ between FP and FN subjects. Table 3 shows the regional metabolic values for the FP and the FN after placebo and after lorazepam. There were also no significant differences in baseline metabolism between FP and FN for cortical and subcortical regions. Baseline metabolic values in right and left cerebellum showed a trend toward lower values in FP than in FN subjects. For both groups of subjects, lorazepam significantly decreased whole-brain (FN = −5 ± 2 μg/mol/g/min; FP = −3.9 ± 3 μmol/g/min) and regional brain metabolism (Table 3). Figures 1 and 2 show metabolic

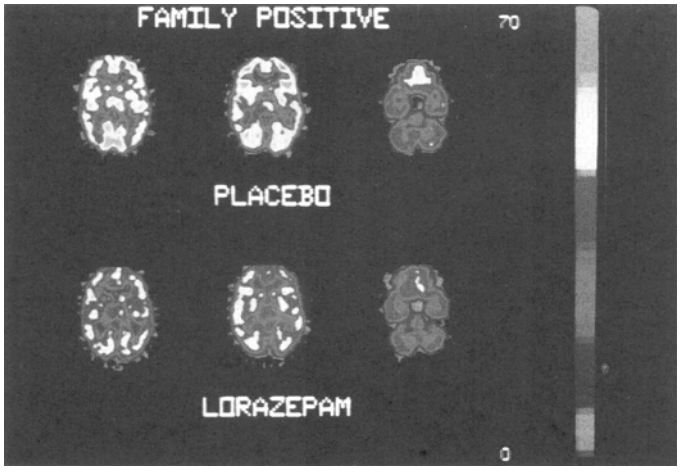

Fig. 2. Metabolic images from a FP subject tested after placebo and after lorazepam (30 μg/kg iv). Note the blunted cerebellar response after lorazepam.

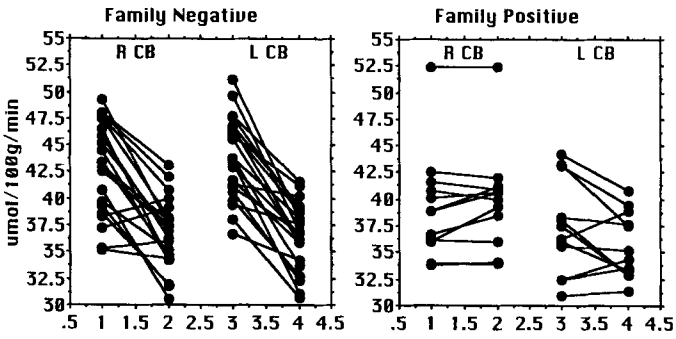

Fig. 3. Individual values for changes in right cerebellar (R CB) and left cerebellar metabolism (L CB) in FP and FN subjects. Lorazepam-induced differences in metabolism were significantly smaller in FP than in FN subjects in right cerebellum ($p < 0.01$) and showed a trend for left cerebellum ($p < 0.05$).

images after placebo and after lorazepam for a FN and a FP subject, respectively. For both groups of subjects, the largest decrements were in thalamus (FN = −12.2 ± 5 μmol/g/min; FP = −10.7 ± 8 μmol/g/min) and in occipital cortex (FN = −10.8 ± 5 μmol/g/min; FP = −8.9 ± 5 μmol/g/min) (Table 3). Although FP subjects showed smaller decrements in metabolism with lorazepam than those in FN, the differences were only significant for the right cerebellum (FN = −6.1 ± 4 μmol/g/min; FP = −2.1 ± 4 μmol/g/min; $F = 6.62$, $df = 1,31$, $p < 0.01$), and there was a trend in the left cerebellum (FN = −7.3 ± 3 μmol/g/min; FP = −4.6 ± 4 μmol/g/min; $F = 4.2$, $df = 1,31$, $p < 0.05$). Figure 3 shows the individual metabolic values for the right and left cerebellum at baseline and after lorazepam for the FN and the FP subjects. There was also a trend for lorazepam-induced changes in cingulate gyrus to be smaller in FP than in FN subjects (FN = −6.5 ± 4 μmol/g/min; FP = −2.7 ± 5.3 μmol/g/min; $F = 5.9$, $df = 1.31$, $p < 0.02$).

Correlation analyses between lorazepam induced changes in regional brain metabolism, and behavioral effects revealed a significant positive correlation between changes in cerebellar metabolism and motor impairment

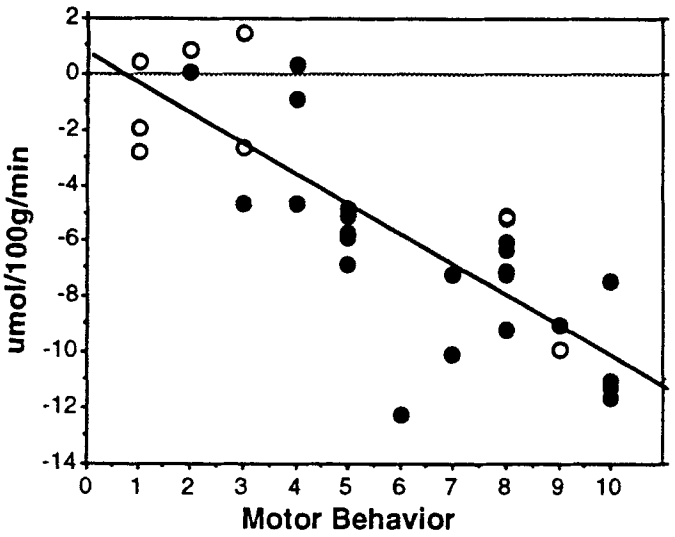

Fig. 4. Correlation analyses between lorazepam-induced changes in motor behavior and lorazepam-induced changes in cerebellar metabolism (placebo-lorazepam) ($r = 0.81$, $p < 0.001$). FP, open circles; FN, closed circles.

(Fig. 4) for FP subjects ($r = 0.83$, $df = 11$, $p < 0.001$) and FN subjects ($r = 0.77$, $df = 20$, $p < 0.001$), and between changes in thalamic metabolism and sleepiness in FN ($r = 0.60$, $df = 20$, $p < 0.005$) and a trend between changes in cerebellar metabolism and sleepiness in FN ($r = 0.52$, $df = 20$, $p < 0.02$). There were no other significant correlations between the regional changes in metabolism and the behavioral changes induced by lorazepam.

DISCUSSION

This study documents a blunted cerebellar response to lorazepam in subjects at risk for alcoholism. Furthermore, because lorazepam-induced changes in cerebellar metabolism were associated with the motor incoordinating effects of lorazepam, one could postulate that the decreased response of the cerebellum may account for the blunted response to the motor incoordinating action of alcohol previously reported in FP subjects.[9,10] It may also account for the blunted response in FP subjects to the effects of benzodiazepines in eye movements,[19] because the cerebellum is implicated in eye movement control.[32] Because both alcohol and benzodiazepine agonists facilitate inhibitory transmission at the GBRC by potentiating GABA-induced Cl^- flux,[33–38] one could postulate that decreased sensitivity of GBRC in cerebellum could explain the blunted motor response to alcohol and/or benzodiazepines. The fact that the blunted response to lorazepam was regionally specific (cerebellum) suggests that changes in GBRC are subtype-specific. The subtype specificity of the GBRC is given by its subunit composition (α, β, γ, and δ[39]). The subunit composition gives the GBRC its unique pharmacological properties and its regional brain heterogeneity.[39] The cerebellum is unique in that it is the only brain region that expresses the α6 subunit.[40] Receptors formed with the α6

subunit uniquely bind RO 154513, the benzodiazepine inverse agonist that inhibits the effects of alcohol.[40] RO 154513 has been found to reverse alcohol-induced motor impairment, anticonflict activity, and behavioral intoxication and to block its effects on GABA-induced Cl^- flux.[41–43] Hence, one could hypothesize that differences in sensitivity of this receptor subtype may explain the differences in sensitivity to alcohol. Evidence implicating a genetic involvement of this receptor subtype in sensitivity to alcohol was recently provided by a study in which injection of GBRC mRNAs from the brains of mice differentially inbred for their sensitivity to ethanol (Long Sleep and Short Sleep mice) into oocytes, resulted in differences in their sensitivity to ethanol and RO 154513.[44] The extent to which the antagonistic effects of RO 15453 are specific to ethanol and not generalizable to other drugs that interact with the GBRC is controversial.

It has been postulated that decreased GABA neurotransmission could predispose an individual to alcoholism and/or benzodiazepine abuse.[45,46] Genetically determined differences in the GBRC have been associated with sensitivity to ethanol in animals, including: (1) differences in sensitivity to chloride flux enhancement by ethanol[47]; (2) differences in concentration and affinity for benzodiazepine and GABA receptor ligands[48–50]; (3) differences in receptor properties[47,48,51,52]; and (4) differences in genes encoding for the GBRC or associated proteins.[44] In contrast to the extensive amount of work done in animals, there are relatively few studies addressing the role of the GBRC in genetics of alcoholism. Most of the studies have been done in alcoholics, and few studies have been conducted to evaluate the function of the GBRC in subjects at risk for alcoholism. Supporting the involvement of the GBRC in alcoholism are postmortem studies that have shown decreases in benzodiazepine receptors in the frontal cortex[53] and in the hippocampus of alcoholics,[54] and a preliminary PET study that documented a larger variability in the concentration of benzodiazepine receptors in alcoholics than in normal subjects.[55] Supporting this association is also the frequent abuse of benzodiazepines by alcoholics[56] and the lower cerebral spinal fluid GABA levels reported in alcoholics when compared with normal controls.[45] Furthermore, alcoholics report higher scores on items related to "drug liking" after benzodiazepines than normal controls (for review, see ref. 28).

Studies in FP subjects have predominantly evaluated their sensitivity to alcohol. In general, these studies have documented decreased sensitivity to the effects of ethanol.[2,6,7,9,10,12,13] A recent study that evaluated the effects of benzodiazepines in FP subjects reported decreased effects on peak saccadic velocity, average smooth pursuit gain, memory, and self-rated sedation.[19] Interestingly, these subjects reported significantly greater pleasurable effects for benzodiazepines than FN.[19] Although another study had found greater pleasurable effects to benzodiazepines in FP,[57] two others have not,[8,58] and we were also unable to document a significant difference between FP and FN subjects. Inconsistencies among investigators may reflect differences in sample populations as well as differences in the conditions at which the drug was given. For the current study, failure to document differences in pleasurable response to lorazepam between FP and FN may reflect the environment conditions during a PET experiment.

The current study documents decreased cerebellar metabolism at baseline in FP subjects, when compared with FN. These cerebellar changes could account for the baseline gait abnormalities documented in subjects at risk for alcoholism.[59] Because cerebellar functions have been predominantly associated with motor behaviors, it is hard to associate a change in cerebellar activity with a process as complex as genetic predisposition to alcoholism. However, there is evidence that the cerebellum is involved in regulating the sensitivity to some of the effects of alcohol[60] and that the genetic sensitivity to ethanol may be mediated in part by cerebellar neuronal circuitry.[61,62] There is also evidence implicating the involvement of the cerebellum with reinforcement.[63,64] The cerebellum could also regulate the response to alcohol indirectly via its projections to cortical and subcortical regions.[65–67] It is also possible that the cerebellar changes in FP may not involve a mechanistic interaction with vulnerability to alcoholism. From the current study, it is not possible to determine the extent to which the baseline cerebellar differences between FP and FN accounted for the differences in their cerebellar metabolic response to lorazepam.

Comparison of the metabolic response in the FP differs from that in the alcoholics in that the latter showed a blunted response that was most pronounced in basal ganglia and thalamus and not in cerebellum.[24] The fact that we were unable to document a blunted response to lorazepam in these subcortical brain regions in FP suggests that these changes in the alcoholics are associated with chronic alcohol exposure. Chronic ethanol exposure has been shown to reduce GABA-receptor-mediated Cl^- uptake[68,69] and to decrease the density of low-affinity binding sites for [^{3}H]GABA[70] and [^{3}H]muscimol.[71] In addition, chronic ethanol has been shown to affect the subunit composition of the GBRC.[72–74]

Different than other PET studies using ligands such as ^{11}C RO 15-1788,[75] ^{11}C suriclone,[76] and ^{11}C RO 15-4513[77,78] to measure benzodiazepine receptors, this study measured the regional brain metabolic consequences of the interaction of a benzodiazepine drug with these receptors. The regional metabolic response to a benzodiazepine drug probably reflects not only its direct interactions with benzodiazepine receptors, but also secondary effects. The extent to which the blunted response in subjects at risk reflects differences in receptors or secondary effects cannot be discriminated by this study. Direct evaluation of benzodiazepine receptors with the use of appropriate radioligands could help clarify this issue.

In interpreting the results from this study, one has to

keep in mind the small sample size that may have precluded the detection of differences between the groups. Small sample sizes in this type of study are particularly problematic, because the expression of the "predisposing factor and/or factors" may not be present in all subjects selected, and the degree of their expression may be environmentally related. Furthermore, inclusion of subjects >25 years of age may have decreased our chances of optimizing a high genetic load in our sample. The case could even be made that subjects at risk, which by 30 years of age are not alcohol-dependent, may possess "protective factors." Despite these limitations, the fact that we were able to document a difference between FP and FN subjects that corroborates previous studies documenting decreased motor response to alcohol[5,10] and benzodiazepines[19] in FP subjects suggests that the cerebellum may be directly or indirectly involved in the sensitivity to the effects of alcohol and benzodiazepines. The extent to which this cerebellar change is linked with vulnerability to alcoholism requires further study.

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