Cognitive impairments by alcohol and sleep deprivation indicate trait characteristics and a potential role for adenosine A_1 receptors

Short title: Trait Vulnerability to sleep loss and alcohol

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The other authors declare no conflict of interest.

Abstract:

Trait-like differences in cognitive performance after sleep loss put some individuals more at risk than others, the basis of such disparities remaining largely unknown. Similarly, inter-individual differences in impairment in response to alcohol intake have been observed. We tested whether performance impairments due to either acute or chronic sleep loss can be predicted by an individual's vulnerability to acute alcohol intake. Secondly, we used positron emission tomography (PET) to test whether acute alcohol infusion results in an upregulation of cerebral A_1 adenosine receptors (A_1AR), similar to the changes previously observed following sleep deprivation. Sustained attention in the psychomotor vigilance task (PVT) was tested in 49 healthy volunteers (26 ± 5 SD years, 15 females) under 1) baseline conditions, 2) after ethanol intake and after either 3A) total sleep deprivation (TSD, 35 h awake, n=35) or 3B) partial sleep deprivation (PSD, 4 nights with 5 h scheduled sleep, n=14). Ethanol versus placebo induced changes in cerebral A_1AR availability were measured in 10 healthy male volunteers (31 ± 9 years) with [^{18}F]CPFPX-PET. Highly significant correlations between the performance impairments induced by ethanol and sleep deprivation were found for various PVT parameters including mean speed (TSD r=0.62, PSD r=0.84). A_1AR availability increased up to 26 % in several brain regions with ethanol infusion. Our studies revealed individual trait characteristics for being either vulnerable or resilient to both alcohol and sleep deprivation. Both interventions induce gradual increases in cerebral A_1AR availability pointing to a potential common molecular response mechanism.

Significance statement:

Modern work environments favour working around the clock at the cost of an increased risk for fatigue related human error. Performance impairments caused by sleep loss show great variability among individuals and resemble the effects of ethanol intoxication. We found that there are individual trait characteristics that make individuals either vulnerable or resilient to both alcohol and sleep deprivation. We also provide evidence by molecular brain imaging pointing to the cerebral adenosine system as common molecular response mechanism to ethanol and sleep deprivation. Understanding the mechanistic link between cerebral adenosine and the vulnerability to performance impairment might help to identify countermeasures for fatigue related human error.

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Introduction:

Fatigue due to sleep loss and extended time awake is a major cause of accidents and incidents in the modern 24-h society (1–6). Surveys on sleep habits suggest that ~40% of the German population (7) and ~30% of the U.S. population (2, 8) sleep less than the recommended 7-h minimum sleep duration (9). Chronic sleep restriction is associated with increased sleepiness and cognitive performance impairment (10–13). The extent of impairment varies greatly among individuals, putting some more at risk than others (14, 15), whereas the physiological basis of this variation remains largely unknown. In contrast, performance varies relatively little across repeated exposures within an individual, indicating trait-like behaviour (16) and a substantial genetic influence (17). For a comprehensive review on the inter-individual neurobehavioral variation in the response to sleep deprivation and its involvement of adenosinergic, dopaminergic and cholinergic pathways see Tkachenko and Dinges (18). Similarly, trait-like differences exist in alcohol sensitivity and the vulnerability to performance impairment after alcohol intake (19). Ethanol is a psychoactive drug with depressant effects on the central nervous system. For example, alcohol facilitates sleep onset

and increases deep sleep in the first half of the sleep episode, whereas it disrupts sleep in the second half of the sleep episode (20, 21). It affects multiple neuroreceptor systems in the brain: It is a positive allosteric modulator at GABA receptors, agonist at the alpha 7 nicotinic acetylcholine and serotonin 3 receptors, antagonist at the NMDA, AMPA and Kainate receptors, and an inhibitor of various ion channels. Moreover, ethanol blocks the reuptake of adenosine and increases its formation. Adenosine is a nucleoside that plays a central role in energy transfer as it is formed by breakdown of ATP, the main energy source of cells. In the brain, adenosine acts as a neuromodulator and co-transmitter that inhibits the impact of excitatory neurotransmitters like glutamate. The physiological effect of adenosine is mediated by 4 types of receptors of which the A_1 subtype is most widespread in the human brain. The brain's adenosine system has emerged as a potential candidate mediating both the sleep inducing effects of alcohol as well as the increase in sleep propensity and sleepiness after sleep loss. The impairment of alertness and motor / cognitive performance by alcohol (22) is attributed to the inhibitory effects of increased adenosine concentration and to an increase of inhibitory A_1 adenosine receptor (A_1AR) availability (23–25). The increase in extracellular adenosine has been reported to inhibit wake-promoting cholinergic neurons in the basal forebrain, since blocking the A_1AR as well as lesioning of cholinergic neurons in the basal forebrain resulted in a reduction of the sedative effect of ethanol (26). Ethanol was found to increase extracellular adenosine levels up to 4-fold in cell culture and in vivo microdialysis experiments by stimulating its production and by uptake inhibition (27). Agonists at the A_1AR such as cyclohexyladenosine are known to accentuate the effects of ethanol whereas A_1AR antagonists like caffeine and theophylline attenuate them. Sleep deprivation increases A_1AR availability in the human brain as a function of elapsed time awake (28), thereby impairing brain functioning.

We hypothesized that 1) the individual performance degrading effects of sleep loss and alcohol correlate, i.e. that the response to alcohol predicts individual vulnerability to sleep loss, and that 2) ethanol induces a global increase in A_1AR availability in the human brain resembling the effects of sleep loss.

Three studies were conducted to test these hypotheses. First, 47 participants were exposed to alcohol and 38 h of wakefulness on separate days to compare the impairment of sustained attention on an individual basis. Second, individual impairment was compared in 16 participants who were exposed to alcohol and 4 days of partial sleep deprivation (5 h time in bed) on separate days. Third, cerebral A_1AR availability of 10 participants was measured in vivo using positron emission tomography (PET) comparing the impact of ethanol versus placebo infusion.

Results:

Alcohol intake, total sleep deprivation and 4 days of sleep restriction each resulted in impairment of sustained attention, measured as deviation from baseline in a psychomotor vigilance task (PVT); (Fig. 1). Individuals who showed good performance after alcohol intake proved to be more resistant to the effects of acute and chronic sleep loss, whereas individuals who showed large impairments after alcohol, were more vulnerable to the two types of sleep deprivation (Fig. 1 upper row). A median split was performed based on individuals' performance under the influence of alcohol. The resulting two groups differed significantly in mean speed during TSD and PSD (Fig. 2). While the resilient group recovered from sleep loss after one night of sleep, recovery remained incomplete in the vulnerable group.

Highly significant Spearman correlations (Table 1, Fig. 1 lower row) between performance impairments after alcohol intake and acute or chronic sleep deprivation were found for several PVT measures. In contrast, the impairment in the examined performance parameters under

alcohol or sleep deprivation did not depend on an individual's absolute performance at baseline (i.e. no significant correlation except for 10% slowest reaction times after sleep deprivation: $p=0.048$). Moreover, the small differences in blood alcohol levels (BAC) between subjects did not predict performance impairment ($r<0.36$, $p>0.2$).

Using quantitative [^{18}F]CPFPX positron emission tomography (PET), ethanol infusion rapidly increased A_1AR availability in various brain regions while placebo infusion had no such effect (Fig. 3 and 4A). Comparison between the post-treatment interval (i.e. 110 to 140 min after tracer infusion) and the baseline interval (50 to 90 min) revealed for the ethanol group a relative increase in A_1AR availability ranging from 20% ± 5% SEM (amygdala) to 26% ± 5% (cuneus), depending on the brain region. In comparison, under identical control conditions (placebo group) there were no significant differences in any brain region (variation of 3% ± 3% across examined regions). Details are displayed in Table 2. The increase in A_1AR availability showed large interindividual differences that were not correlated with BAC (Fig. 4B).

The biphasic alcohol effect scale (BAES), which is a measure of alcohol's stimulating and sedating effects, showed in the sedating subscale a significant correlation with the treatment-dependent relative changes in A_1AR in several brain regions (SI Appendix, Table S1). Exemplarily, the striatum (left and right, $r=-0.99$, $p=0.0001$) is depicted in Figure 4C. Importantly, BAES did not show a correlation with BAC ($r=-0.05$, $p=0.91$).

Discussion:

The present study shows that cognitive performance impairments due to ethanol intake and sleep loss correlate strongly across individuals, indicating the presence of an underlying trait vulnerability. The present study also reveals that acute ethanol administration is accompanied

by a rapid increase in A_1AR availability in the human brain and that the subjective sedating effects as assessed by the BAES correlate strongly with cerebral A_1AR availability. Given the previously observed upregulation of A_1AR availability with extended time awake (28, 29), the cerebral adenosine system might therefore represent a common pathway for both ethanol and sleep deprivation effects on performance. Behavioral studies have established a link between both stressors providing evidence for additive or even synergistic detrimental effects on cognitive performance (30-33).

The large inter-individual variation in the susceptibility of cognitive performance to acute and chronic sleep deprivation that we found here substantiates previous reports (15, 34) and is consistent with a genetic contribution (e.g. refs. (35, 36)). Large inter-individual variations have also been reported for the wakefulness and performance promoting effects of caffeine (37), which is known to act via adenosine receptor antagonism (38). Individual differences in caffeine sensitivity have been linked to genetic variations in the adenosine system (39, 40). In a recent study using PET we found that the degree of an individual's performance impairment after extended time awake was related to cerebral A_1AR availability (29). Taken together, the adenosine system appears to be subject to individual variations that in turn contribute to the differences in the vulnerability to sleep loss.

The increase in A_1AR availability in humans after ethanol exposure confirms previous reports of in vitro and in vivo experiments in rodents. Autoradiography experiments in rats showed that 1.5 g/kg ethanol led to a 40% increase in A_1AR availability after only 15 min (23). A 35 to 55% increase in distribution volume was measured 20 min after application of ethanol along with an adenosine kinase inhibitor (25). Recent results from mice suggest that sleep deprivation attenuates the sensitivity to alcohol via reducing extracellular accumulation of adenosine in a long-lasting manner (41). The magnitude of the 35% increase in cerebral

A_1AR availability after alcohol infusion in the present study is comparable to a respective decrease observed after 4 cups of espresso (42).

Caffeine has been reported to be an effective countermeasure against performance impairment in the PVT during sleep deprivation, sleep inertia (43–45), as well as circadian misalignment (43–45). Although numerous studies have investigated caffeine as antidote for alcohol-related performance decrements, outcomes remain equivocal (43–47). Moreover, there are differences in their neurochemical action, as the antagonist caffeine blocks receptor binding whereas ethanol rapidly increases A_1AR availability through a mechanism that has not been clarified so far. It is possible that alcohol slows down the turnover of cell-surface receptors and increases receptor trafficking to the cell surface. Mechanisms could be an enhancement of an intracellular receptor pool that involve receptor assembly and maturation or sensitization (at least in the basal ganglia) by A_1 adenosine- D_1 dopamin receptor heteromeres interaction (48, 49). Nonetheless, the present behavioral and PET studies indicate that alcohol and caffeine may represent at least in part antagonistic drugs as they appear to interact with a common underlying pathway.

In conclusion, these findings support the framework that there are individual trait characteristics for being either vulnerable or resilient to both alcohol and sleep deprivation. Based on previous studies on the effects of extended wakefulness (29) and the present findings on ethanol induced changes in cerebral A_1AR availability we provide evidence that both alcohol and sleep deprivation effects are mediated – at least partly – via the adenosine system of the brain. Important related research questions that need to be addressed to corroborate our conclusion are (i) whether individual changes in A_1AR availability after sleep deprivation correlate with those under ethanol influence, (ii) whether chronic sleep restriction induces alterations of A_1AR availability in the human brain, and (iii) whether chronic sleep

restriction modulates the effect of ethanol on neurobehavioral performance. Targeting the adenosine system might therefore help identify new countermeasures against compromised performance induced by both sleep deprivation and alcohol. Possible countermeasures could include individualized fatigue management systems and training. Furthermore, assessing the sensitivity to alcohol might be cautiously used as a means to identify/protect individuals who are at greatest risk to be impaired by sleep loss, e.g. in safety-critical environments (e.g. transportation, healthcare, and the military) that include extended duration shifts or night work. Especially in situations of high workload and stress, sleep loss can result from sleep disturbances (50) or from trading sleep for additional work hours; in such situations alcohol consumption is increased (51), and thus insight into putative cumulative effects of sleep loss, stress, and alcohol is highly relevant. A recent model that predicts cognitive performance after sleep loss based on putative changes of adenosine and the A_1AR (52) could be extended to the influence of ethanol on performance and be evaluated accordingly. Finally, our studies are important from a public health education point of view. Reports indicate that young drivers believe that driving drowsy is less of a serious problem than driving under the influence of alcohol (53). Our finding of a shared physiological basis raises awareness for the still underestimated danger of driving while sleep deprived.

Materials and Methods:

Alcohol, sleep deprivation, and performance (Study 1 and 2):

Participants: The studies were approved by the Ethics Committee of the North Rhine Medical Board. Written informed consent was obtained from all participants. Volunteers' physical and psychological health was confirmed with questionnaires and medical examination. They reported normal sleep-wake rhythmicity and conducted no shift-work. From a total of 63 healthy volunteers, data sets of 35 participants in study 1 (mean age ± SD 26 ± 5 years, 20

men/15 women) and 14 participants in study 2 (mean age ± SD 27 ± 4 years, 14 men) were included in the present analysis. 14 participants (12 from study 1/ 2 from study 2) were excluded (see below).

Study Design: The two studies spanned 12 days and 11 nights and were both carried out in the sleep laboratory of the Institute of Aerospace Medicine (German Aerospace Center). Eight participants at a time underwent the experiments, and they were constantly attended by at least two study staff members.

Study 1 – Total sleep deprivation: After an adaptation night and two baseline nights (sleep opportunity: 23:00-07:00 h) participants underwent three experimental conditions administered in a balanced cross-over design. The experimental conditions were 1) 38-h total sleep deprivation, 2) one night of partial sleep deprivation (i.e. sleep opportunity restricted to 00:00-04:00 h), and 3) alcohol administration at 16:00 h followed by partial sleep deprivation (sleep opportunity: 00:00-04:00 h). The experimental conditions were followed each by two nights of recovery sleep (all recovery nights 23:00-07:00 h except for the first recovery night after total sleep deprivation 21:00-07:00 h).

Study 2 – Partial sleep deprivation: Methodological details of the study were published elsewhere (13, 54). In brief, after an adaptation night and two baseline nights (sleep opportunity: 23:00-07:00 h) participants underwent one day with hypoxia exposure during daytime cognitive testing, one day with alcohol administration at 10:30 h (n=12) or 16:00 h (n=2), one recovery day/night followed by 4 nights with restricted sleep opportunities (00:00-5:00 h) and two recovery nights (sleep opportunity: 23:00-07:00 h) in a sequential design.

Alcohol administration: In study 1 and 2 a drink of vodka whose amount was calculated concerning sex, weight, height, and age was consumed within 5 min. Participants' blood alcohol concentration (BAC) was based on calibrated breath alcohol analyser (ALCOTEST of the Draeger company) results immediately before the cognitive testing session at 18:00 h or

12:00 h. A BAC of more than 0.06% immediately prior to the respective cognitive testing session was chosen as minimum for inclusion in the analyses. Data of 14 participants (i.e. 12 in study 1 and 2 in study 2) had to be excluded due to physical problems like nausea or vomiting or because they did not reach this threshold due to inability to drink the whole amount of vodka. In study 1 the dataset included 35 participants with a mean BAC of 0.074% ± 0.001% SD (min. 0.064%, max. 0.095%, 25% Quantile 0.065%, 75% Quantile 0.078%). In study 2 the 14 participants included in the present analysis had a mean BAC of 0.076% ± 0.009% SD (min. 0.060%, max. 0.094%, 25% Quantile 0.069%, 75% Quantile 0.080%).

Performance measurements: Sustained attention was measured with a 10-min PVT (16) at 3-h intervals during baseline, experimental and recovery days. Apart from the cognitive testing session, participants engaged in non-vigorous activities such as reading and watching TV.

Statistics: Impairment of cognitive performance in the experimental conditions was assessed by calculating the difference in the PVT measures (e.g. mean speed, etc.) from the respective values during the first baseline day. Spearman rank correlations were calculated to compare performance impairments due to sleep deprivation/restriction with those due to alcohol intake. Based on participants' impairment in mean speed after alcohol exposure a median split was performed. Unpaired t tests compared the mean speed of the two groups during baseline, sleep deprivation, sleep restriction, and recovery.

Alcohol PET study (study 3):

Participants: The study was approved by the Ethics Committee of the Medical Faculty of the University of Duesseldorf and the German Federal Office for Radiation Protection. The study was performed at the PET laboratory of the Institute of Neuroscience and Medicine (Forschungszentrum Jülich). Written informed consent was obtained from all participants. Physical and psychological health was confirmed by medical examination and questionnaires

before ten healthy male volunteers were assigned to the experimental (n=7; mean age ± SD 31 ± 9 years) or control group (n=3; mean age ± SD 31 ± 12 years) (non-randomized, single blinded).

PET: 3D data acquisition took place on a Siemens ECAT EXACT HR+ scanner (Siemens-CTI, Knoxville, TN, USA). Scan duration was 140 min with radiotracer injection as bolus followed by a constant infusion with a Kbol value of 63 min. Realignment, co-registration, segmentation, and normalization of PET data and corresponding MRI (acquired on a 3T Siemens Magnetom Trio with MPRAGE sequence) were done with PMOD (version 3.305, PMOD Group, Zuerich, Switzerland). Repeated arterialized venous blood sampling was scheduled at 2, 5, and 10 min, and every 10 min until 80 min and every 5 min subsequently. The total distribution volume V_T in the equilibrium (between 50 and 140 min) is represented by the formula $V_T = TAC / C_p$ with TAC being the tissue activity concentration and C_p the plasma activity（28）.

More details concerning the PET experiment are provided in SI Appendix.

Alcohol administration/subjective effects: From minute 80 to 110 of the radiotracer infusion, 40 g of ethanol in 1 L saline or solely 1 L saline solution as placebo were infused. In order to assess the subjective effects of the infusion we used a German version of the biphasic alcohol effect scale (BAES)（55） with a slightly modified instruction that did not imply that alcohol was consumed. Subjects were asked to rate how they feel right before the ethanol infusion and at the end of the PET scan.

Statistics: The alcohol response in the PET regional A_1AR distribution volumes (V_T; mL/mL) was quantified in reference to: (i) baseline (pre alcohol infusion) and (ii) post-infusion with a t tests for pairwise comparisons with a Bonferroni-corrected significance level. Spearman rank correlation was used to evaluate associations between adenosine receptor availability and: (i) BAC and (ii) BAES.

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Figure Legends

Figure 1: Comparison of psychomotor vigilance task (PVT) performance impairment by ethanol, total sleep deprivation (left column of diagrams) and partial sleep deprivation (right column).

A, B: Individual impairment in 10% slowest reaction times (RT; means; difference from baseline) after alcohol (diamonds) and sleep deprivation (circle), ordered according to impairment after alcohol. Intraclass correlation coefficients (ICC, two-way mixed effects model, absolute agreement) indicate moderate agreement. Note that degree of impairment did not vary due to differences in blood alcohol concentration (color-coded diamonds ranging from orange (low concentration) to black (high concentration)).

C, D: Significant spearman correlations between the impairment (difference from baseline) in mean speed under alcohol and after sleep deprivation (see also Table 1).

Figure 2: Time course of mean speed during (A) total sleep deprivation (TSD), after (B) repeated partial sleep deprivation (PSD) and after recovery (REC) sleep. Based on performance impairment under alcohol the subjects were divided into two subgroups (median split). Gray vertical bars represent sleep periods. Error bars indicate standard error of the mean (SEM). * $p<0.05$ for differences between subgroups (unpaired t-tests).

Figure 3: Effects of ethanol infusion on cerebral A_1 adenosine receptor availability. Spatially normalized averaged images of anatomy (A: MRI) and adenosine receptor availability (B+C: PET distribution volume) of the ethanol receiving subjects (n=7) during the baseline interval (average of 50-80 min after start of radioligand infusion) and during the post-treatment interval (110-140 min) are shown. Left column: axial, middle column sagittal and right column coronal view.

Figure 4: Effects of ethanol infusion on cerebral A_1 adenosine receptor availability. A: Time course of the normalized (to the baseline interval) distribution volume in the frontal cortex of two representative subjects during the steady state phase of radioligand delivery. 30-min ethanol infusion (0.53g/kg, black diamonds) or placebo infusion (shaded circles for reference) started at 80 min after start of the PET experiment during the steady state phase and continued for 30 min (33 ml/min infusion rate). B: No correlation between changes in A_1AR binding and ethanol dose. C: Significant correlation between changes in A_1AR binding and changes in ethanol induced sedating symptoms (biphasic alcohol effect scale).

Table 1: PVT performance impairment after total and partial sleep deprivation, and alcohol administration.

PVT parameters	TSD	Ethanol		PSD	Ethanol	
	mean Δ (SEM)	mean Δ (SEM)	Spearman r-value p-value	mean Δ (SEM)	mean Δ (SEM)	Spearman r-value p-value
Median RT (ms)	36.92 (10.03)	31.96 (4.91)	0.567 **0.0004**	4.10 (4.73)	23.51 (8.10)	0.862 **<0.0001**
Mean Speed (1/s)	-0.64 (0.11)	-0.47 (0.07)	0.620 **<0.0001**	-0.07 (0.10)	-0.41 (0.13)	0.842 **0.0002**
10th Percentile Speed (1/s)	-1.25 (0.23)	-0.64 (0.10)	0.603 **0.0001**	-0.16 (0.14)	-0.53 (0.16)	0.886 **<0.0001**
90th Percentile Speed (1/s)	-0.29 (0.06)	-0.32 (0.06)	0.614 **<0.0001**	0.05 (0.08)	-0.27 (0.10)	0.833 **0.0002**
Lapses (#)	5.14 (1.29)	1.60 (0.48)	0.508 **0.0018**	1.43 (0.72)	2.43 (1.15)	0.281 0.5410
Mean RT (ms)	29.99 (4.20)	33.08 (4.76)	0.659 **<0.0001**	4.89 (5.19)	24.00 (8.22)	0.846 **0.0001**
Standard deviation of RT (ms)	19.70 (2.59)	14.99 (2.08)	0.742 **<0.0001**	6.14 (2.78)	10.12 (3.28)	0.574 **0.0320**
Mean 10% slowest RT (ms)	74.73 (9.78)	65.58 (8.50)	0.662 **<0.0001**	17.00 (11.29)	42.49 (13.76)	0.666 **0.0093**
Mean 10% fastest RT (ms)	8.47 (2.08)	12.97 (2.80)	0.462 **0.0052**	-1.80 (3.53)	9.72 (4.16)	0.846 **0.0001**

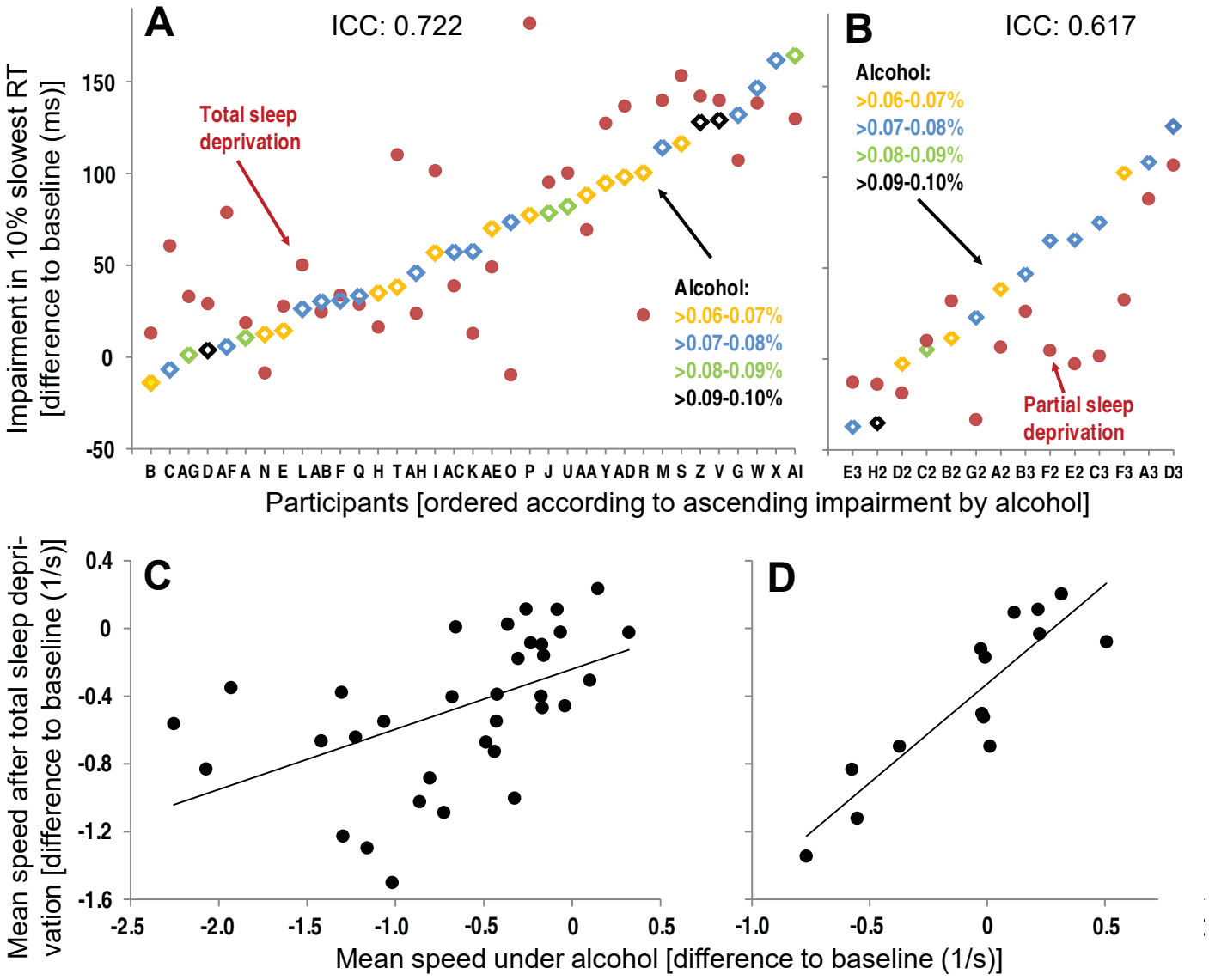
PVT, psychomotor vigilance task; RT, reaction time; TSD, total sleep deprivation (i.e. 35 h awake); PSD, partial sleep deprivation (i.e. after 4 nights with time in bed restricted to 5 h).

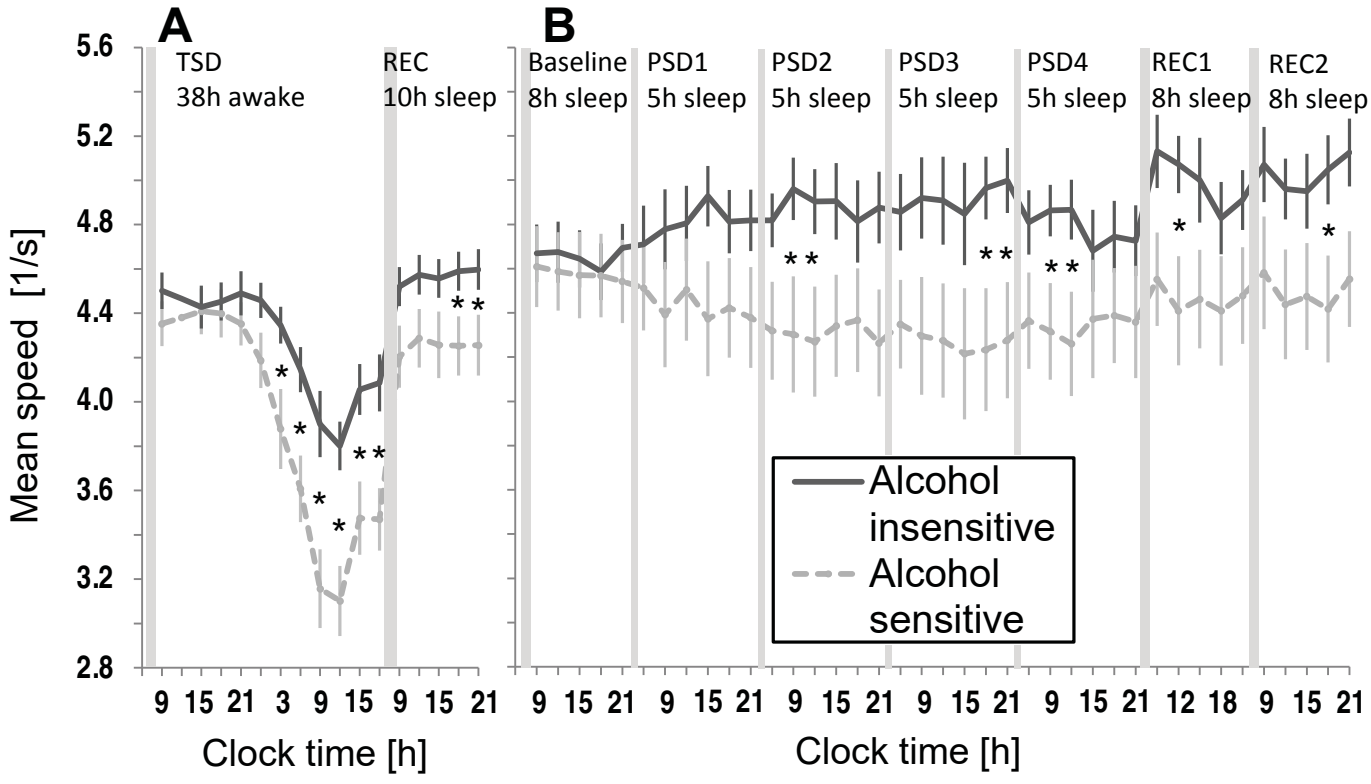
Median RT and parameters of speed include all RTs, Lapses are RTs ≥ 500 ms, other PVT parameters are based on RT excluding lapses. Mean Δ is the difference between experimental condition (TSD, PSD, Ethanol) and baseline at respective daytimes. 10th percentile refers to difference in slow response speed, 90th percentile to difference in fast response speed.

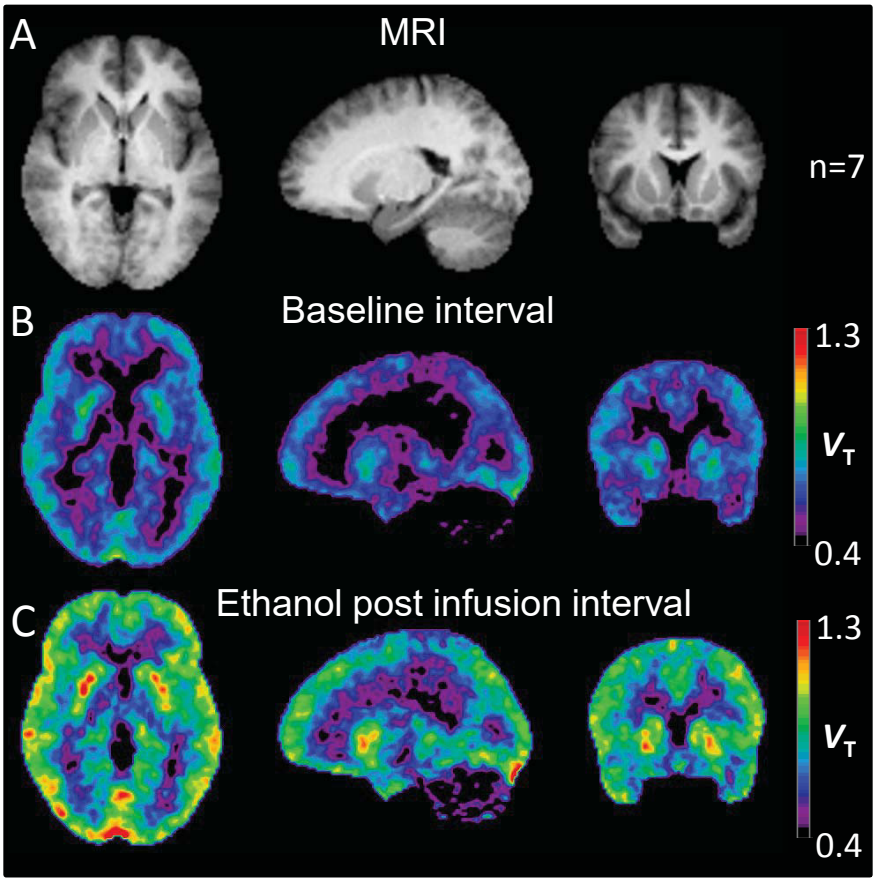
Table 2: Relative regional changes of cerebral A_1AR availability after ethanol or placebo administration.

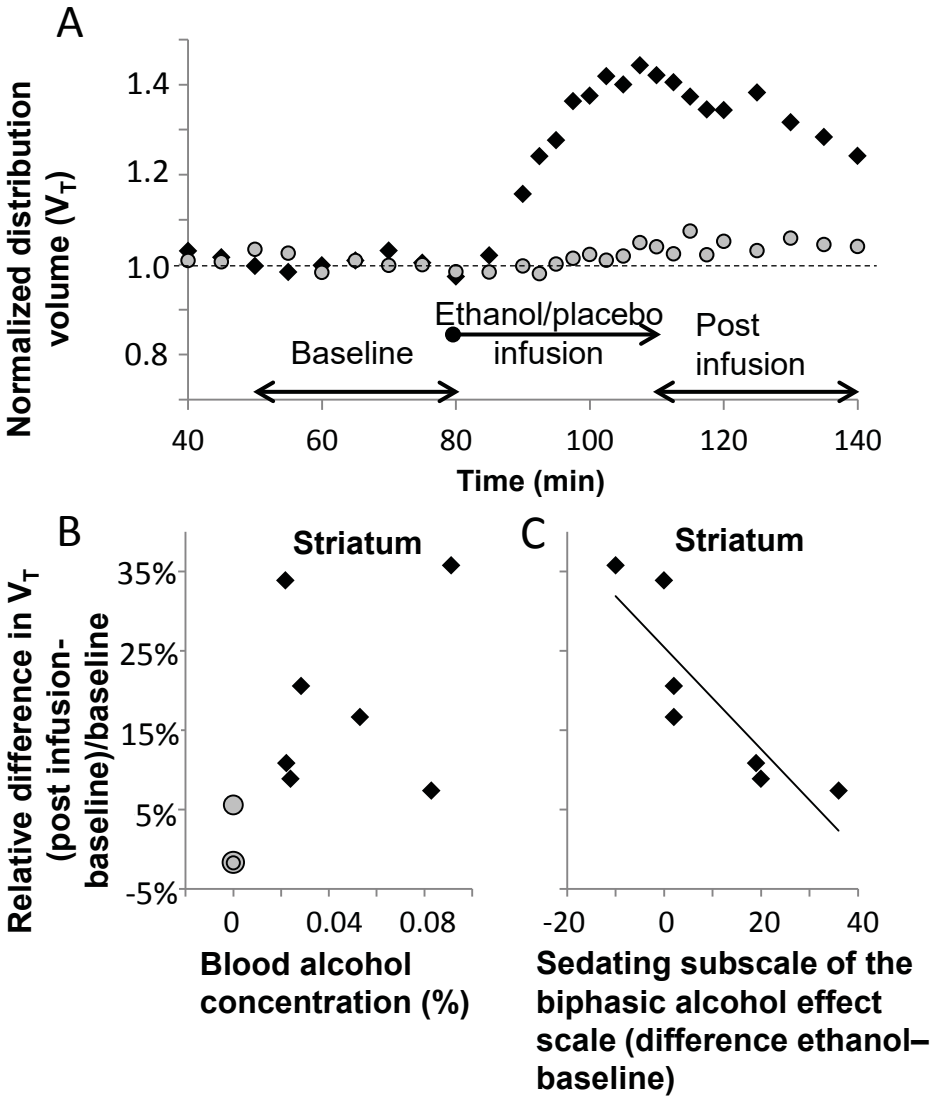
Brain regions	Relative increase in A_1AR availability (in %)			
	Ethanol group		Control group	
	mean relative Δ(SEM)	p-value	mean relative Δ(SEM)	p-value
Supp Motor Area	24 (5)	**0.001**	6 (2)	0.047
Frontal inf	23 (5)	**0.001**	3 (2)	0.269
Insula	23 (5)	**0.002**	0 (3)	0.905
Cingulum ant	25 (5)	**0.001**	2 (2)	0.434
Hippocampus	20 (4)	**0.002**	5 (4)	0.287
Amygdala	20 (5)	**0.002**	5 (6)	0.431
Cuneus	26 (5)	**0.001**	8 (3)	0.063
Occipital	23 (5)	**0.002**	3 (2)	0.124
Postcentral	24 (5)	**0.001**	3 (2)	0.288
Striatum	19 (5)	**0.002**	1 (3)	0.775
Pallidum	25 (6)	**0.003**	1 (3)	0.742
Thalamus	22 (5)	**0.003**	2 (2)	0.440
Temporal	23 (5)	**0.002**	3 (2)	0.185
Parietal	22 (5)	**0.001**	3 (2)	0.261

A_1AR, A_1 adenosine receptor; ant., anterior; Supp., supplementary. Significant p-values after Bonferroni correction (0.05/14=0.0035) are bold. Mean relative Δ is (post infusion-baseline)/baseline.


A
ICC: 0.722
B
ICC: 0.617
Impairment in 10% slowest RT [difference to baseline (ms)]
Total sleep deprivation
Alcohol:
>0.06-0.07%
>0.07-0.08%
>0.08-0.09%
>0.09-0.10%
Partial sleep deprivation
Participants [ordered according to ascending impairment by alcohol]
C
D
Mean speed after total sleep deprivation [difference to baseline (1/s)]
Mean speed under alcohol [difference to baseline (1/s)]


A
TSD
38h awake
REC
10h sleep
B
Baseline
8h sleep
PSD1
5h sleep
PSD2
5h sleep
PSD3
5h sleep
PSD4
5h sleep
REC1
8h sleep
REC2
8h sleep
Mean speed [1/s]
5.6
5.2
4.8
4.4
4.0
3.6
3.2
2.8
9 15 21 3 9 15 9 15 21
9 15 21 9 15 21 9 15 21 9 15 21 9 15 21 9 15 21 12 18 9 15 21
Clock time [h]
Clock time [h]
Alcohol insensitive
Alcohol sensitive


A
MRI
n=7
B
Baseline interval
1.3
V_T
0.4
C
Ethanol post infusion interval
1.3
V_T
0.4


A
Normalized distribution volume (V_T)
1.4
1.2
1.0
0.8
Baseline
Ethanol/placebo infusion
Post infusion
40
60
80
100
120
140
Time (min)
B
Striatum
Relative difference in V_T (post infusion-baseline)/baseline
35%
25%
15%
5%
-5%
0
0.04
0.08
Blood alcohol concentration (%)
C
Striatum
-20
0
20
40
Sedating subscale of the biphasic alcohol effect scale (difference ethanol–baseline)

Supplemental information PET imaging

This supporting information gives additional methodological details about the PET scans.

Participants: Detailed interviews ensured that volunteers did not have a history of neurological and psychiatric diseases, sleep disorders, shift work, jet-lag, night work, head injury, and alcohol or substance abuse. Only non-smoking subjects without any medication were included. Caffeine intake was restricted 36 h prior to the arrival at the laboratory which was controlled by blood plasma samples at the time of the PET scans (below detection limits in all subjects - < 0.5 mg/L). The average BMI of the subjects was 25.8 ± 4.8 kg/m². Daytime of scanning was 11:15 ± 1:23h.

Average amount of injected radioactivity was 264 ± 41 MBq corresponding to an average dose of 2.4 ± 1.2 nmol.

The free fraction of the parent compound in plasma (f_P) was determined 10 min before tracer injection and after 140 min (in 5 subjects additionally at 80 and 110 min). No significant differences were detected between time points, ethanol and placebo group and an independent group of previously investigated subjects. On average the free fraction difference between -10 and 140 min was 3% ($p = 0.87$) in the ethanol group.

The time course of the concentration of the parent compound of all subjects is displayed in Fig. S1.

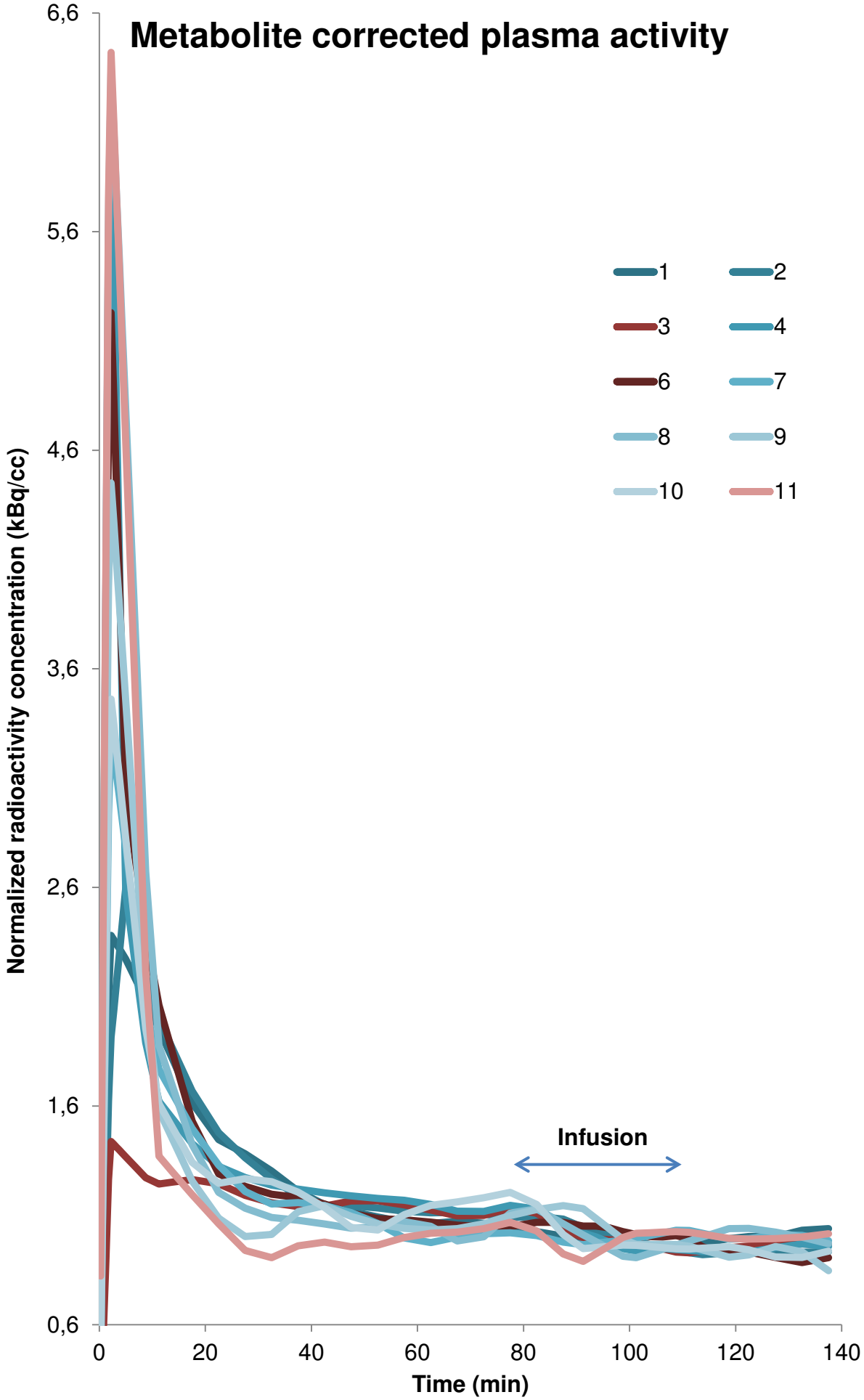


Figure S1: Overview of metabolite corrected plasma activity (normalized to the time span 60-140 min). Red colored lines represent the subjects which received a placebo infusion.

Table S1: Correlation of regional relative difference of cerebral A_1AR availability after ethanol with difference the sedating subscale of the biphasic alcohol effect scale.

Brain regions	r-value	p-value
Supp Motor Area	-0.76	0.0489
Frontal inf	-0.87	0.0120
Insula	-0.87	0.0120
Cingulum ant	-0.81	0.0269
Hippocampus	**-0.99**	**0.0001**
Amygdala	-0.85	0.0162
Cuneus	-0.90	0.0056
Occipital	**-0.96**	**0.0008**
Postcentral	**-0.96**	**0.0008**
Striatum	**-0.99**	**0.0001**
Pallidum	-0.68	0.0897
Thalamus	**-0.96**	**0.0008**
Temporal	**-0.96**	**0.0008**
Parietal	**-0.96**	**0.0008**

Significant p-values after Bonferroni correction (0.05/14=0.0036) are bold.