Association between increased retinal background noise and co-occurrent regular cannabis and alcohol use

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ABSTRACT

BACKGROUND: Cannabis consumption is widespread across the world, and the co-occurrence of cannabis use and alcohol consumption is common. The study of background noise - resting-state neural activity, in the absence of stimulation - is an approach that could enable the neurotoxicity of these substances to be explored. Preliminary results have shown that delta-9-tetrahydrocannabinol (Δ9-THC) causes an increase in neural noise in the brain. Neurons in the brain and the retina share a neurotransmission system and have similar anatomical and functional properties. Retinal function, evaluated using an electroretinogram (ERG), may therefore reflect central neurochemistry. This study analyses retinal background noise in a population of regular co-occurrent cannabis and alcohol consumers.

METHODS: We recorded the flash ERGs of 26 healthy controls and 45 regular cannabis consumers, separated into two groups based on their alcohol consumption: less than or equal to 4 glasses per week (CU≤4) or strictly greater than 4 glasses per week (CU >4). In order to extract the background noise, the Fourier transform of the pseudo-periodic and sinusoidal signals of the 3.0 flicker-response sequence was calculated. This sequence represents the vertical transmission of the signal from cones to bipolar cells. The magnitude of the background noise is defined as the average of the magnitudes of the two neighbouring harmonics : harmonic -1 (low frequency noise) and harmonic +1 (high frequency noise).

RESULTS: The magnitude of harmonic -1 was significantly increased between the groups CU>4 (6.78 (+/-1.24)) and CU≤4 (5.69 (+/-1.80)) among regular users of cannabis and alcohol. A significant increase in the average magnitude of the two harmonics was found between the groups CU>4 (5.12 (+/-0.92)) and CU≤4 (4.36 (+/-1.14)). No significant difference was observed with regard to the magnitude of the harmonic +1.

CONCLUSIONS: The increase in background noise may reflect the neurotoxicity of cannabis, potentiated by alcohol consumption, on retinal neurons dynamic. This neural disruption of the response generated by retinal stimulation may be attributable to altered neurotransmitter release.

Keywords: retinal background noise; cannabis; alcohol; retina; electroretinogram

1. Introduction

Cannabis is the third most commonly used psychoactive substance in the world after alcohol and tobacco (1). Co-occurrence of alcohol consumption in regular consumers of cannabis is common (2). Cannabis and alcohol are both neurotoxic substances that may potentiate each other's effects, justifying the investigation of their toxicity both alone and in combination (3). In particular, they are responsible for altered synaptic transmission (4, 5). The mechanisms of action of their neurotoxicity are the subject of many scientific papers in the field of neuroscience.

Studying background noise is an innovative approach to exploring this neurotoxicity. Background noise represents the neural electrical activity recorded without visual stimulation (6). In the brain, acute cannabis use increases neural noise, i.e. neural electrophysiological activity in the pre-stimulation period, disrupting the performance of cognitive tasks. The effects are mediated by the endocannabinoid system (7, 8). To our knowledge, the impact of alcohol on neural noise has not been studied.

This study aims to use the retina as a site for the indirect investigation of cerebral neurotransmission by means of retinal background noise in people who use cannabis and alcohol concomitantly. The retina is an anatomical and developmental extension of the central nervous system (9-11). It has a functional endocannabinoid system that is involved in the regulation of retinal neurotransmission (12, 13). In particular, its neurotransmission system shares similarities with the brain transmission system. The neurotransmission system comprises the principal neurotransmitters involved in substance consumption: glutamate, gamma-aminobutyric acid (GABA) and dopamine to name but a few (13). These neurotransmitters are involved in the vertical transmission of retinal signals, thus enabling the propagation of visual information captured by photoreceptors to its transmission to the visual processing centers within the brain (14). The retina may therefore offer functional markers for the abnormalities of cerebral neurotoxicity (15).

Retinal function may be measured using an electroretinogram (ERG). The ERG records electrophysiological signals responding to various types of light stimuli (16). The response

generated reflects the average electrical potential generated by populations of neurons and is associated with changes to the levels of their neurotransmitters (17). Regular cannabis users have delayed signal processing in the retina versus healthy volunteers, as a result of delayed ganglion and bipolar cell responses (18, 19). Alcohol-induced retinal toxicity might also cause delayed processing of retinal signals (20, 21). This is a consequence of changes to the organization of the bipolar cell layer (20). These anomalies - the consequences of cannabis and alcohol use - are supported by malfunctions of the synaptic transmission in the retina caused by regular use of these substances.

This is the first study assessing retinal background noise in regular, co-occurrent users of cannabis and alcohol. The objective of our study is to compare the background noise recorded using flash ERG of regular cannabis users divided into two populations according to their level of alcohol consumption, and a population of healthy volunteers. Our hypothesis is that background noise is increased in regular users of cannabis and on the basis of their level of alcohol consumption.

2. Methods and Materials

2.1. Population and ethics statement

Regular cannabis users (n=56) and matched healthy drug-naive controls (n=29) were recruited among the general population via a special press campaign and data were collected from February 11, 2014, to June 30, 2016. Among participants, data of 14 participants (11 cannabis users and 3 controls) were excluded because of lacking data or uninterpretable, then 45 cannabis users and 26 controls were included in this study. The 45 regular cannabis users were separated into two groups according to the median of the number of alcohol uses/week (=4), as follows: a group of 24 regular cannabis users with a number of alcohol uses/week strictly higher than 4 (CU>4) and a group of 21 regular cannabis users with a number of alcohol uses/week equal to or less than 4 (CU≤4). Prior to taking part in the study, volunteers provided their detailed psychoactive drugs and medical history, underwent a full psychiatric evaluation, and signed consent forms detailing all aspects of the research. All of the participants received payment in the form of €100 in gift vouchers. The study protocol met the requirements of the Helsinki Declaration and was approved by the Ethics Committee of Nancy University Hospital. This study is part of a bigger project, Causa Map, which is researching the impact of regular cannabis use on the visual system. All participants also underwent neuropsychological assessments and EEG recordings during several visual tasks.

2.2. Inclusion criteria, clinical and biological assessments

The inclusion criteria for the cannabis group were regular cannabis use at the rate of at least 7 cannabis consumptions per week over the past month, a positive urine toxicology screen for tetrahydrocannabinol (THC) metabolites, no other illicit substance use in the past month, a negative urine toxicology screen for other illicit substances, and no DSM-IV diagnosis of Axis I disorders. Since tobacco is regularly mixed with cannabis in joints, cannabis users may meet the criteria for tobacco dependence according to the Fagerström test. Cannabis users were required to present at

least 12 hours of abstinence of cannabis use so that there were no acute cognitive dysfunctions due to cannabis use. The inclusion criteria for the healthy control subjects were no history of illicit substance use, a negative urine toxicology screen for THC metabolites and other illicit drugs tested, and no history of DSM-IV diagnosis of Axis I psychiatric disorders. All participants were aged 18 to 35 years, had no history of neurological disease, no family history of schizophrenia or bipolar disorders, and were medication-free except for oral contraceptives in the case of women. They had no history of ophthalmological disease except for corrected refractive errors. All of them fared normally in an ophthalmic evaluation which included visual acuity and a fundoscopic examination. Importantly, visual acuity measured with the Monoyer Scale was at least 10/10 in each eye for all participants. None of the participants reported visual symptoms, and none was found to have any media opacities. If participants reported an alcohol dependence according to their score in the Alcohol Use Disorders Identification Test (AUDIT) they were excluded from the study. The Mini-International Neuropsychiatric Interview (M.I.N.I.) was administered to assess current and past history of psychiatric diseases and substance use. In addition, the Cannabis Abuse Screening Test (CAST), Fagerström Test, and AUDIT were performed to assess use, abuse or dependence with respect to cannabis, tobacco and alcohol, respectively. The extent of cannabis use was clinically assessed in an interview and a questionnaire as follows: age when regular cannabis use began, total years of cannabis use, average number of joints smoked daily and weekly over the past month, average number of grams smoked weekly**.** In order to obtain objective confirmation of cannabis consumption, urine drug screens (Nal von Minden, Moers, Germany) were performed for cannabis, buprenorphine, benzodiazepines, cocaine, opiates, amphetamines and methadone immediately before electroretinogram testing.

2.3. Experimental protocol

2.3.1. Flash electroretinogram (fERG) measurements

fERG was performed according to the International Society for Clinical Electrophysiology of Vision (ISCEV) standards for fERG (16). The MonPackOne system (Metrovision, Perenchies, France) was used for stimulation, recording and analysis. Electrical signals were recorded simultaneously from both eyes (averaged for analysis), on dilated pupils (fERG, Tropicamide 0, 5 %), with DTL electrodes (Metrovision, Perenchies, France) placed at the bottom of the conjunctival sac. The pupil's size was noted before and after fERG recordings and remained systematically constant during the whole testing period. Ground and reference electrodes were attached to the forehead and external canthi.

The standard fERG protocol comprises 5 sequences. This analysis will look at the flicker 3.0 response sequence, which specifically provides information on the status of the transmission of the cone response to their ON and OFF bipolar cells. In line with the ISCEV guidelines, the Flicker 3.0 ERG sequence was performed in light conditions and the standard flash was delivered at a temporal frequency of 30 Hz. This stimulation frequency means that the response of a specific neuron sub-population - L and M cones - can be isolated and their physiological properties exploited (Figure 1).

Participants were positioned 30 centimeters from the screen. They were then light-adapted for 10 minutes to a light background set at 30 candela/m² (cd/m²) provided by MonPackOne system before light-adapted fERG was performed. At least 16 responses were recorded for each participant and were extracted from the same sequence named the Flicker sequence of the flash ERG, as recommended by international guidelines.

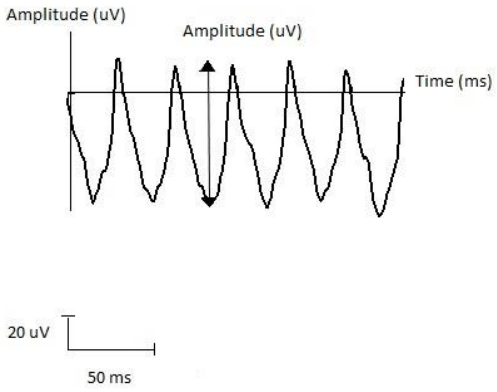

Figure 1. Typical fERG traces obtained when assessing the 3.0 flicker response. The arrow represents the wave amplitude.

2.3.2. **Analysis**

The recording duration for each test was 225ms, divided into 25ms in the pre-stimulation period and up to 200ms in the post-stimulation period. The sampling frequency was 1138 Hz. The tracing obtained was taken from the averaging of the electrophysiological activity across the tests. The final signal had a pseudo-periodic and sinusoidal appearance. Frequency analysis of the averaged signal was carried out using the software program MATLAB (MathWorks). On the basis of the literature concerning the distribution and spectral properties of background noise, we have calculated the Fourier transform of the averaged signal for each subject using the proposed mathematical methods (6). This gives a Time-Frequency spectrum with a frequency resolution of 4.46 Hz. The Fourier transform breaks down a periodic function into a sum of sinusoidal functions called harmonics, which have different magnitudes, measured in microvolts (µV). This enables the part of the signal at the dominant frequency of the stimulation - called fundamental frequency - to be distinguished from the background noise. It is therefore possible to carry out frequency analysis and selectively extract the dominant frequency of the stimulation and its harmonics. We thus extracted the magnitude of the spectrum at the frequency of the stimulus to evaluate the ratio of signal to noise on the recordings. The magnitude of noise was defined as the average of the magnitudes of the two

neighbouring harmonics at 30 Hz +/- 4.46 Hz, i.e. a harmonic ~10% higher and one ~10% lower than the stimulus frequency. The harmonic 10% lower was called harmonic -1 and the harmonic 10% higher was called harmonic +1. The signal-to-noise ratio (SNR) -which could also be named the signal-to-harmonics ratio- was thus calculated by dividing the amplitude of the fundamental by the average of the amplitudes of the adjacent harmonics (6) (Figure 2).

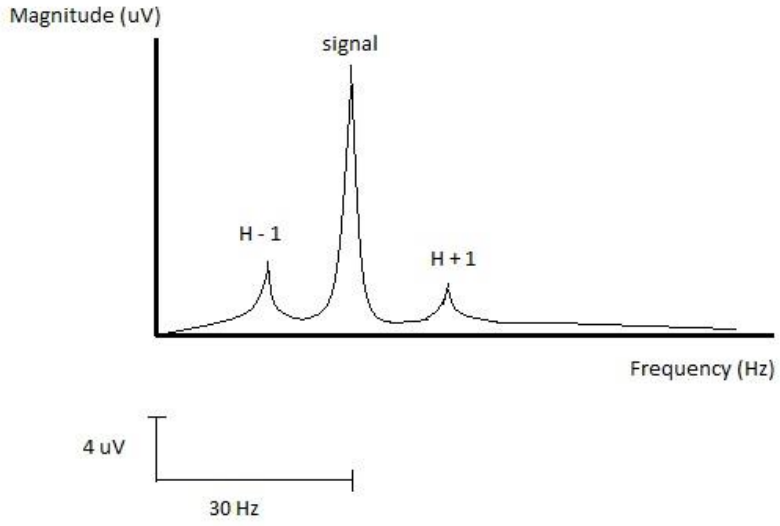

Figure 2. Trace obtained after Fourier analysis. The noise magnitude is defined as the average noise magnitude at the two neighbouring frequencies (H-1 and H+1).

2.4. **Statistical analysis**

Depending on the parametric distribution of variables included in the analyses, a Student test, Chi-square test, Analysis of variance (ANOVA) test and Post hoc comparison with Tukey test were used when appropriate to compare the three groups or to test the association between variables. We used a conservative level of significance in comparison with alpha <0.05%. Statistical analyses were performed using STATISTICA 8.0 (StatSoft, Inc.).

3. Results

3.1. Demographic and substance use characteristics

The demographic and substance use characteristics of the participants are described in Table 1. There was no significant difference between the 3 groups in terms of age (p=0,95), gender (p=0.19 for CU>4 vs CU<4; p=0.38 for CU>4 vs controls and p=0.63 for CU<4 vs controls, chi square test), but differences were noted between groups in terms of years of education (F(2,68)=41,38; p<0,05) and alcohol use (F(2,68)=7,42; p<0,05 for average alcohol consumption/week and F(2,68)=28,01; p<0,05 for AUDIT score). Post hoc analyses with Tukey test, when appropriate, were presented in Table 1.

Table 1. Demographic and substance use characteristics of the participants.

	Cannabis users with>4 alcohol uses/week (CU>4, n=24)	Cannabis users with≤4 alcohol uses/week (CU≤4, n=21)	Controls (n=26)
Gender (male/female)[a,d]	20/4	14/7	19/7
Age (years)[b,c]	25,6 (7,4)	25,1 (4,9)	25,2 (4,3)
Education (years)[b,c]	13,6 (1,5)	13,0 (2,4)	15,0 (1,7)
Average number of alcohol uses/week[b,c]	13,6 (8,2)	2,1 (1,3)	1,9 (2,7)
Alcohol Use Disorders Identification Test (AUDIT) scores[b,c]	8,9 (2,5)	4,4 (3,0)	3,2 (2,8)
Fagerström Test scores[b,e]	1,8 (2,0)	1,4 (1,7)	_
Average number of cigarettes/day[b,e]	6,6 (5,6)	4,6 (5,0)	_
Age of firts cannabis use[b,e]	16,1 (1,2)	16,0 (1,7)	_
Total years of cannabis use[b,e]	9,5 (7,3)	9,1 (5,1)	_
Average number of joints/week[b,e]	23,0 (15,7)	27,7 (23,7)	_
Cannabis Abuse Screening Test (CAST) scores[b,e]	4,2 (1,0)	3,7 (1,4)	_
Average number of grams of cannabis/week[b,e]	7,0 (8,7)	5,0 (4,3)	_

Categorical variable represented as frequencies[a]

Quantitative variable represented as mean and standard deviation[b]

Analysis of variance (ANOVA) test[c]

Chi-Square test[d]

Student test[e]

NS=non significant

Because tobacco is widely mixed with cannabis in joints, 39 in 45 cannabis users were also tobacco smokers, whereas all the controls were non-smokers. According to the Fagerström test, 32 in 45 cannabis users were not dependent on tobacco, 9 in 45 were slightly dependent, 4 in 45 were mildly dependent and 0 in 45 was highly dependent.

3.2. **ERG parameters**

The mean and standard deviation of the magnitude of the harmonic -1 was 6,07 (+/- 1,27) in controls versus 6,78 (+/-1,24) in CU>4 versus 5,69 (+/-1,80) in CU≤4. The magnitude of the harmonic -1 was significantly different between the 3 groups ($F(2.68)=3,87$, $p<0,05$, ANOVA test). Post hoc comparison with Tukey test showed that magnitude of the harmonic -1 significantly differed between CU≤4 and CU>4 ($p<0,05$), but it failed to show any difference between controls and CU>4 ($p=0.19$) and between controls and CU≤4 ($p=0.64$) (Figure 3).

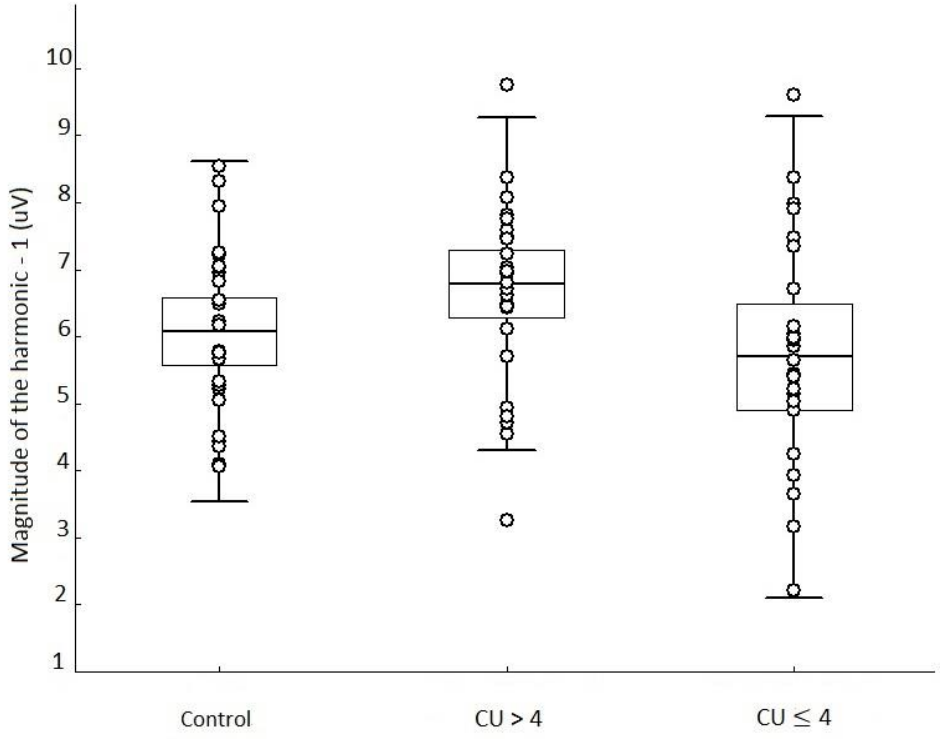

Figure 3. Box plot of magnitude of the harmonic -1 for cannabis users with > and ≤ 4 alcohol uses / week and

control with mean and standard deviation. For controls: n=26; mean: 6,07 µV; DS +/-1,27. For CU>4: n=24; mean: 6,78 µV; DS +/-1,24. For CU ≤4: n=21; mean: 5,69 µV; DS +/-1,80. Small disks represent the individual data points.

The mean and standard deviation of the magnitude of the harmonic +1 was 3,25 (+/-0,79) in controls versus 3,46 (+/-0,88) in CU>4 versus 3,04 (+/-0,79) in CU≤4. The magnitude of the harmonic +1 was not significantly different between the 3 groups (F(2.68)=1.4996, p=0.23, ANOVA test).

The mean and standard deviation of the background noise was 4,66 (+/-0,85) in controls versus 5,12 (+/-0,92) in CU>4 versus 4,36 (+/-1,14) in CU≤4. The magnitude of the background noise was significantly different between the 3 groups (F(2.68)=3.53, p<0,05, ANOVA test). Post hoc comparison with Tukey test showed that background noise significantly differed between CU≤4 and CU>4 (p<0,05), but it failed to show any difference between controls and CU>4 (p=0.22), and between controls and CU≤4 (p=0.55) (Figure 4).

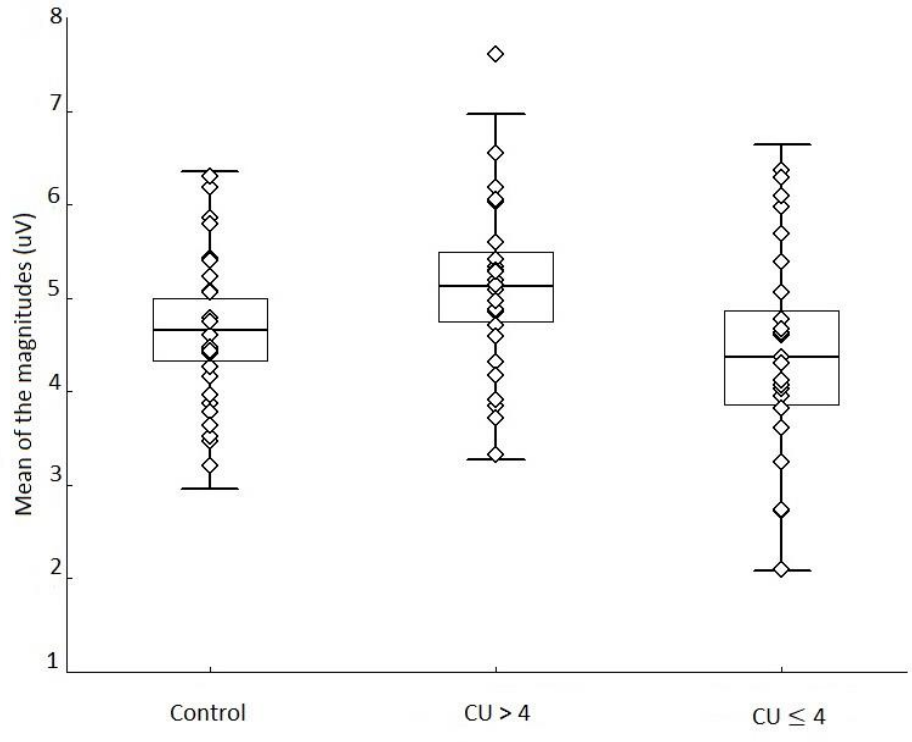

Figure 4. Box plot of magnitude of the retinal background noise for cannabis users with > and ≤ 4 alcohol uses / week and control with mean and standard deviation. For controls: n=26; mean: 4,66 µV; DS +/-0,85. For CU>4: n=24; mean: 5,12 µV; DS +/-0,92. For CU ≤4: n=21; mean: 4,36 µV; DS +/-1,14. Small diamonds represent the individual data points.

4. Discussion

The results of this study highlight an increase in retinal background noise at low frequency harmonic during stimulation, i.e. which could reflect a transient slow-down dynamic of the retinal neuronal response in users with co-occurrent consumption of cannabis and alcohol. This is an indicator of a background neural activity disturbance and a disruption of the retinal neurons cue following visual stimulation. This increase in background retinal noise is apparently an effect of the potentiation of the neurotoxic properties of cannabis and alcohol in a population where the subjects present co-occurrent consumption.

We have observed that the average magnitudes of the two harmonics is significantly increased between the CU>4 and CU≤4 groups, reflecting an increase in overall retinal background noise in regular concomitant users of alcohol and cannabis, throughout the cellular response. A significant increase in the magnitude of harmonic -1 is thus found between the CU>4 and CU≤4 groups, indicating an increase in retinal background noise during visual stimulation. But no significant difference was observed with regard to the magnitude of harmonic +1 between the three groups, which signifies that no disruption in neural activity at high frequency background noise was shown between the groups. The increase in low frequency noise indicates a possible hampering of the total neural activity. Furthermore, no significant difference was found between the control group and the CU>4 group, nor between the control group and the CU≤4 group, regardless of the variable studied. This could reflect an effect of the potentiation of the neurotoxicity of the two substances, alcohol and cannabis, on retinal neural activity rather than an effect of the neurotoxicity of cannabis alone.

The ERG is a test that could enable synaptic transmission anomalies in the retina as a result of regular cannabis use to be studied (5,9,10,15,18,19,22,23). Furthermore, using fERG and pattern-ERG (PERG, reversing checkerboard), our group showed a significant increase in both PERG N95 and fERG b-wave implicit time with no change in amplitude in regular cannabis users versus healthy volunteers. These results reflect slowed processing of retinal information at the level of the ganglion and ON-bipolar cells (18,19). These anomalies may be supported by malfunctions of the

synaptic transmission in the retina caused by regular cannabis consumption. Thus, THC, through direct action on the cannabinoid receptors found in the ganglion and ON-bipolar cells, could alter synaptic transmission and delay the cellular response. This delay could be the result of the anomalies in the neuronal firing that precedes the cellular response to visual stimulation presented here.

We have shown an increase at low frequency retinal background noise, indicating a possible transient slow down disturbance in retinal neuron activity. Cannabis and alcohol are both psychoactive substances that modulate the synaptic release of neurotransmitters to exert their effects (4,5). The results of our study may therefore be explained by the potentiation of the effect of the two substances, alcohol and cannabis, on neurotransmission, particularly glutamatergic and dopaminergic neurotransmission. Glutamate is one of the principal excitatory neurotransmitters detected in the retina (24). It is involved in the vertical transmission of the retinal signal transmitted from the photoreceptors to the bipolar cells and subsequently to the ganglion cells (13). Furthermore, the depolarization and hyperpolarization processes at the origin of the retinal neuronal response, and measured using flash electroretinography, are directly influenced by glutamate concentrations. In our study, a modulation of the polarization of the retinal neurons responsible for the background retinal neuron activity disruption could be a result of the effect of exogenous cannabinoids on glutamatergic transmission. Indeed, by binding to presynaptic CB1 receptors, THC disrupts the regulation of glutamate release caused by endocannabinoids, leading to a synaptic excess of glutamate. This results in an excess of calcium at the postsynaptic level, leading to a state of cellular hyperexcitability (4,13,14,25). Regular alcohol consumption also causes a state of cellular hyperexcitability by inducing hypersensitivity of the postsynaptic NMDA glutamate receptors (26,27). Thus, the state of cellular hyperexcitability caused by the two substances could affect the pseudo-periodic stability of the neural response which could be reflected by the observed increase in retinal background noise. Like glutamate, dopamine is a neurotransmitter found in the cones and bipolar cells, the source of the ERG flicker response, and plays a crucial role in retinal

processing of visual information (9,24,28,29). Regular cannabis consumption may inhibit presynaptic dopamine release in the cones and bipolar cells, as this effect is found under synthetic exogenous cannabinoids (30). Regular alcohol consumption is also associated with a lower synaptic dopamine rate via a reduction in the number of DRD2 receptors (31–33). A correlation between the reduction in retinal dopaminergic synaptic transmission and the increase in retinal background noise was shown by Bubl et al in patients with an attention disorder with or without hyperactivity (ADHD) (34). This effect is reversible under pharmacological dopaminergic treatment. This supports the hypothesis of a connection between the reduction in retinal dopaminergic synaptic transmission and the increase in retinal background noise (35–37). Thus, in our study, the increase in retinal background noise could also reflect a decline in synaptic dopamine release caused by the two substances.

Our study is faced with several methodological considerations and limitations that must be taken into account. A higher level of alcohol consumption is frequently observed in regular cannabis users versus healthy subjects (2). We have obtained results on a common effect, without being able to determine the role of each substance in the results obtained. This means that we cannot draw conclusions as to the isolated effect of each substance on background noise. Ideally, it would be pertinent to constitute a group of users of ‘cannabis only’ and a group of users of ‘alcohol only’ to evaluate precisely the impact of consumption of each of these toxic substances individually on retinal background noise. Constituting such groups is made difficult by the frequent co-occurrent consumption of these substances. Moreover, since deficits in cognitive functions are well known in cannabis users, it will be interesting to study correlations between alterations in retinal background noise and neuropsychological deficits in regular cannabis users.

Tobacco consumption is very common among frequent cannabis users, and tobacco is frequently used with cannabis to roll joints, particularly in France. Future studies should screen this bias with a control group including tobacco smokers. To our knowledge, the effect of nicotine administration on background noise has not been evaluated. Here, we did not find significant

correlations between retinal background noise and Fagerström score in the two populations of cannabis users.

Our results suggest that an increase in retinal background noise is a marker of the potentiation of cannabis and alcohol neurotoxicity, but they do not indicate a potential threshold effect. Studying a threshold effect could determine the minimum quantity under which background noise is not affected and the maximum quantity above which it is no longer affected. This could make it possible to clarify whether background noise alterations begin from the experimental phase, or if they are only found with regular consumption or beyond a certain threshold of consumption of the two psychoactive substances.

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Contributors
All the authors contributed to write the manuscript, concurred with the submission and have approved the final manuscript.

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Disclosures
All the authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Figure legends:

Figure 1. Typical fERG traces obtained when assessing the 3.0 flicker response. The arrow represents the wave amplitude.

Figure 2. Trace obtained after Fourier analysis. The noise magnitude is defined as the average noise magnitude at the two neighbouring frequencies (H-1 and H+1).

Figure 3. Box plot of magnitude of the harmonic -1 for cannabis users with > and ≤ 4 alcohol uses / week and control with mean and standard deviation. For controls: n=26; mean: 6,07 µV; DS +/-1,27. For CU>4: n=24; mean: 6,78 µV; DS +/-1,24. For CU ≤4: n=21; mean: 5,69 µV; DS +/-1,80. Small disks represent the individual data points.

Figure 4. Box plot of magnitude of the retinal background noise for cannabis users with > and ≤ 4 alcohol uses / week and control with mean and standard deviation. For controls: n=26; mean: 4,66 µV; DS +/-0,85. For CU>4: n=24; mean: 5,12 µV; DS +/-0,92. For CU ≤4: n=21; mean: 4,36 µV; DS +/-1,14. Small diamonds represent the individual data points.

Table legend:

Table 1: Demographic and substance use characteristics of the participants.

References

1. Degenhardt L, Chiu W-T, Sampson N, Kessler RC, Anthony JC, Angermeyer M, et al. Toward a global view of alcohol, tobacco, cannabis, and cocaine use: findings from the WHO World Mental Health Surveys. PLoS Med. 2008;5: e141.
2. Meier MH, Caspi A, Ambler A, Harrington H, Houts R, Keefe RSE, et al. Persistent cannabis users show neuropsychological decline from childhood to midlife. Proc Natl Acad Sci U S A. 2 oct 2012;109(40):E2657-2664.
3. Broyd SJ, van Hell HH, Beale C, Yücel M, Solowij N. Acute and Chronic Effects of Cannabinoids on Human Cognition-A Systematic Review. Biol Psychiatry. 1 avr 2016;79(7):55767.
4. Bossong MG, Niesink RJM. Adolescent brain maturation, the endogenous cannabinoid system and the neurobiology of cannabis-induced schizophrenia. Prog Neurobiol. nov 2010;92(3):37085.
5. Schwitzer T, Robert MP, Giersch A, Angioi-Duprez K, Ingster-Moati I, Pon-Monnier A, et al. Transient Retinal Dysfunctions after Acute Cannabis Use. Eur Addict Res. 2016;22(6):28791.
6. Meigen T, Bach M. On the statistical significance of electrophysiological steady-state responses. Doc Ophthalmol Adv Ophthalmol. 1999;98(3):20732.
7. Cortes-Briones JA, Cahill JD, Skosnik PD, Mathalon DH, Williams A, Sewell RA, et al. The psychosis-like effects of Δ(9)-tetrahydrocannabinol are associated with increased cortical noise in healthy humans. Biol Psychiatry. 1 déc 2015;78(11):80513.
8. Laprevote V, Bon L, Krieg J, Schwitzer T, Bourion-Bedes S, Maillard L, et al. Association between increased EEG signal complexity and cannabis dependence. Eur Neuropsychopharmacol. 2017;27(12):1216-22.
9. Schwitzer T, Lavoie J, Giersch A, Schwan R, Laprevote V. The emerging field of retinal electrophysiological measurements in psychiatric research: A review of the findings and the perspectives in major depressive disorder. J Psychiatr Res. nov 2015;70:11320.
10. Schwitzer T, Schwan R, Bubl E, Lalanne L, Angioi-Duprez K, Laprevote V. Looking into the brain through the retinal ganglion cells in psychiatric disorders: A review of evidences. Prog Neuropsychopharmacol Biol Psychiatry. 2 juin 2017;76:15562.
11. London A, Benhar I, Schwartz M. The retina as a window to the brain-from eye research to CNS disorders. Nat Rev Neurol. janv 2013;9(1):4453.
12. Hoon M, Okawa H, Della Santina L, Wong ROL. Functional architecture of the retina: development and disease. Prog Retin Eye Res. sept 2014;42:4484.
13. Schwitzer T, Schwan R, Angioi-Duprez K, Giersch A, Laprevote V. The Endocannabinoid System in the Retina: From Physiology to Practical and Therapeutic Applications. Neural Plast. 2016;2016:2916732.
14. Schwitzer T, Schwan R, Angioi-Duprez K, Ingster-Moati I, Lalanne L, Giersch A, et al. The cannabinoid system and visual processing: a review on experimental findings and clinical presumptions. Eur Neuropsychopharmacol J Eur Coll Neuropsychopharmacol. janv 2015;25(1):10012.
15. Schwitzer T, Schwan R, Bernardin F, Jeantet C, Angioi-Duprez K, Laprevote V. Commentary: Anatomical constitution of sense organs as a marker of mental disorders. Front Behav Neurosci. 2016;10:56.
16. McCulloch DL, Marmor MF, Brigell MG, Hamilton R, Holder GE, Tzekov R, et al. ISCEV Standard for full-field clinical electroretinography (2015 update). Doc Ophthalmol Adv Ophthalmol. févr 2015;130(1):112.
17. Holder GE, Celesia GG, Miyake Y, Tobimatsu S, Weleber RG, International Federation of Clinical Neurophysiology. International Federation of Clinical Neurophysiology: recommendations for visual system testing. Clin Neurophysiol Off J Int Fed Clin Neurophysiol. sept 2010;121(9):1393409.
18. Schwitzer T, Schwan R, Angioi-Duprez K, Giersch A, Lalanne L, Albuisson E, et al. Delayed bipolar and ganglion cells neuroretinal processing in regular cannabis users: The retina as a relevant site to investigate brain synaptic transmission dysfunctions. J Psychiatr Res. août 2018;103:75-82.
19. Schwitzer T, Schwan R, Albuisson E, Giersch A, Lalanne L, Angioi-Duprez K, Laprevote V.

Association Between Regular Cannabis Use and Ganglion Cell Dysfunction. JAMA Ophthalmol. 2017 Jan 1;135(1):54-60.
20. McKellar MJ, Hidajat RR, Elder MJ. Acute ocular methanol toxicity: clinical and electrophysiological features. Aust N Z J Ophthalmol. août 1997;25(3):22530.
21. Ikeda H. Effects of ethyl alcohol on the evoked potential of the human eye. Vision Res. 4 mars 1963;3(3):15569.
22. Laprevote V, Schwitzer T, Giersch A, Schwan R. Flash electroretinogram and addictive disorders. Prog Neuropsychopharmacol Biol Psychiatry. 2015;56: 264.
23. Laprevote V, Schwan R, Schwitzer T, Rolland B, Thome J. Is There a Place for Off-Label Pharmacotherapy in Cannabis Use Disorder? A Review on Efficacy and Safety. Curr Pharm Des. 2015;21: 3298–3305.
24. pubx_pdf_2004_marc_retinal _NTs.pdf [Internet]. [cité 19 déc 2017]. Disponible sur: http://prometheus.med.utah.edu/~marclab/pubx_pdf_2004_marc_retinal%20_NTs.pdf
25. Colizzi M, McGuire P, Pertwee RG, Bhattacharyya S. Effect of cannabis on glutamate signalling in the brain: A systematic review of human and animal evidence. Neurosci Biobehav Rev. mai 2016;64:35981.
26. Roberto M, Varodayan FP. Synaptic targets: Chronic alcohol actions. Neuropharmacology. 1 août 2017;122:8599.
27. Burnett EJ, Chandler LJ, Trantham-Davidson H. Glutamatergic plasticity and alcohol dependence-induced alterations in reward, affect and cognition. Prog Neuropsychopharmacol Biol Psychiatry. 4 févr 2016;65:30920.
28. Bodis-Wollner I. Retinopathy in Parkinson Disease. J Neural Transm Vienna Austria 1996. nov 2009;116(11):1493501.
29. Ghilardi MF, Marx MS, Bodis-Wollner I, Camras CB, Glover AA. The effect of intraocular 6-hydroxydopamine on retinal processing of primates. Ann Neurol. avr 1989;25(4):35764.
30. Yazulla S. Endocannabinoids in the retina: from marijuana to neuroprotection. Prog Retin Eye Res. sept 2008;27(5):50126.
31. Engel JA, Jerlhag E. Alcohol: mechanisms along the mesolimbic dopamine system. Prog Brain Res. 2014;211:20133.
32. Trantham-Davidson H, Chandler LJ. Alcohol-induced alterations in dopamine modulation of prefrontal activity. Alcohol Fayettev N. déc 2015;49(8):7739.
33. Thanos PK, Volkow ND, Freimuth P, Umegaki H, Ikari H, Roth G, et al. Overexpression of dopamine D2 receptors reduces alcohol self-administration. J Neurochem. sept 2001;78(5):1094103.
34. Bubl E, Dörr M, Riedel A, Ebert D, Philipsen A, Bach M, et al. Elevated background noise in adult attention deficit hyperactivity disorder is associated with inattention. PloS One. 2015;10(2):e0118271.
35. Forssberg H, Fernell E, Waters S, Waters N, Tedroff J. Altered pattern of brain dopamine synthesis in male adolescents with attention deficit hyperactivity disorder. Behav Brain Funct BBF. 4 déc 2006;2:40.
36. Volkow ND, Wang G-J, Tomasi D, Kollins SH, Wigal TL, Newcorn JH, et al. Methylphenidate-elicited dopamine increases in ventral striatum are associated with long-term symptom improvement in adults with attention deficit hyperactivity disorder. J Neurosci Off J Soc Neurosci. 18 janv 2012;32(3):8419.
37. Volkow ND, Wang G-J, Fowler JS, Telang F, Maynard L, Logan J, et al. Evidence that methylphenidate enhances the saliency of a mathematical task by increasing dopamine in the human brain. Am J Psychiatry. juill 2004;161(7):117380.

Background noise represents the neural activity recorded without stimulation.
An increased retinal background noise is observed in co-occurrent cannabis and alcohol users.
Retinal background noise could explore pathophysiology of addictive disorders.

Conflict of interest

All the authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Contributors

All the authors contributed to write the manuscript, concurred with the submission and have approved the final manuscript.

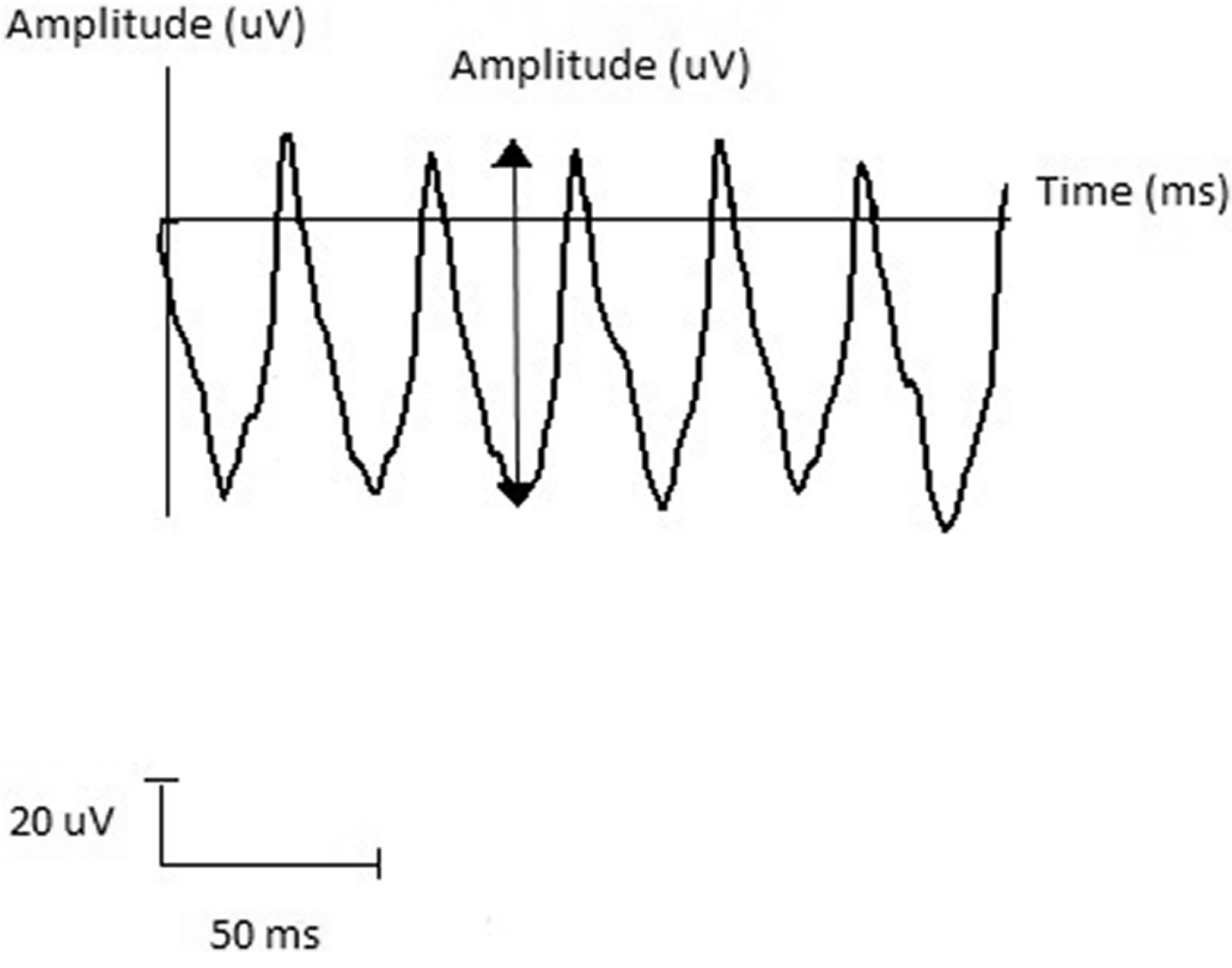

Figure 1

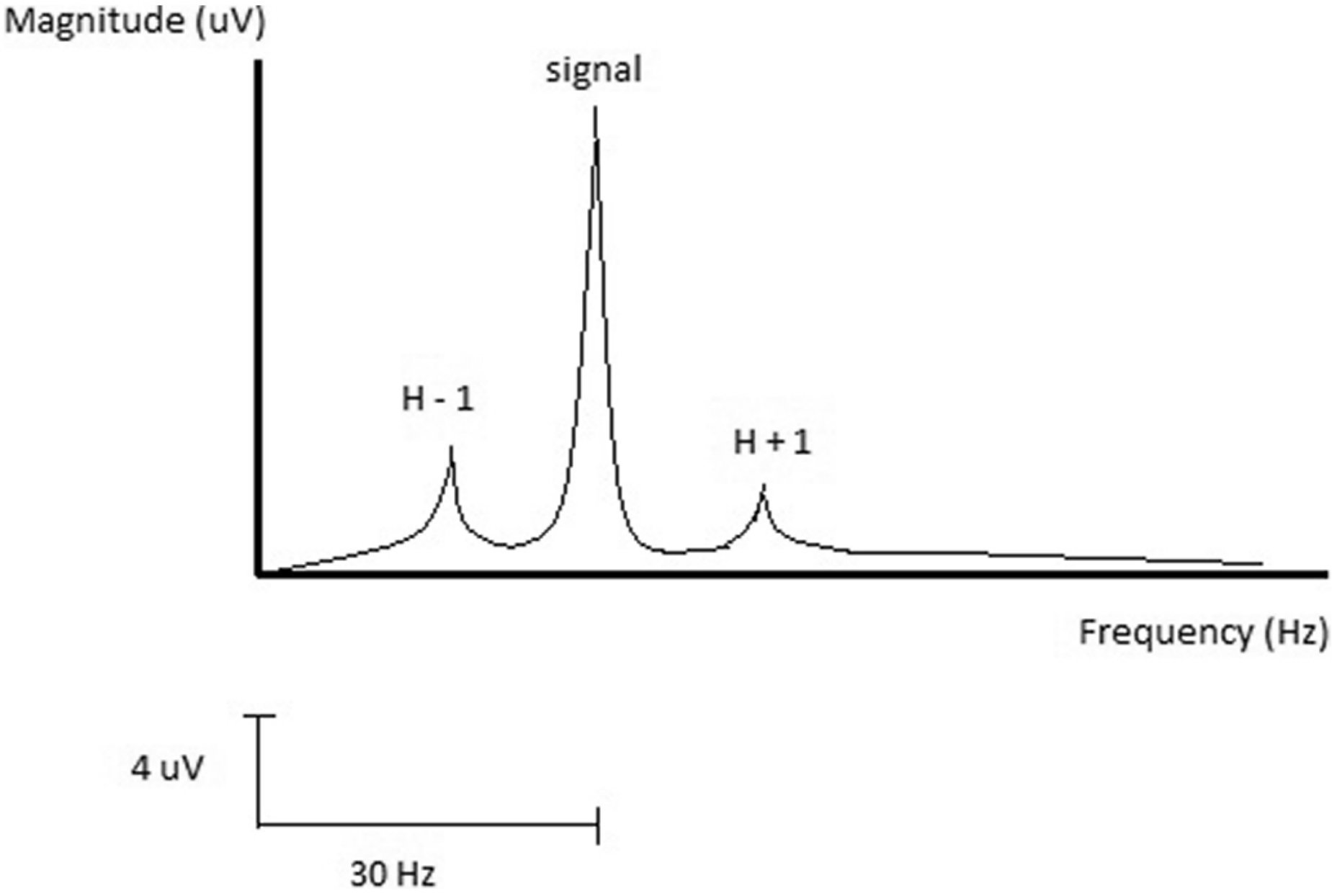

Figure 2

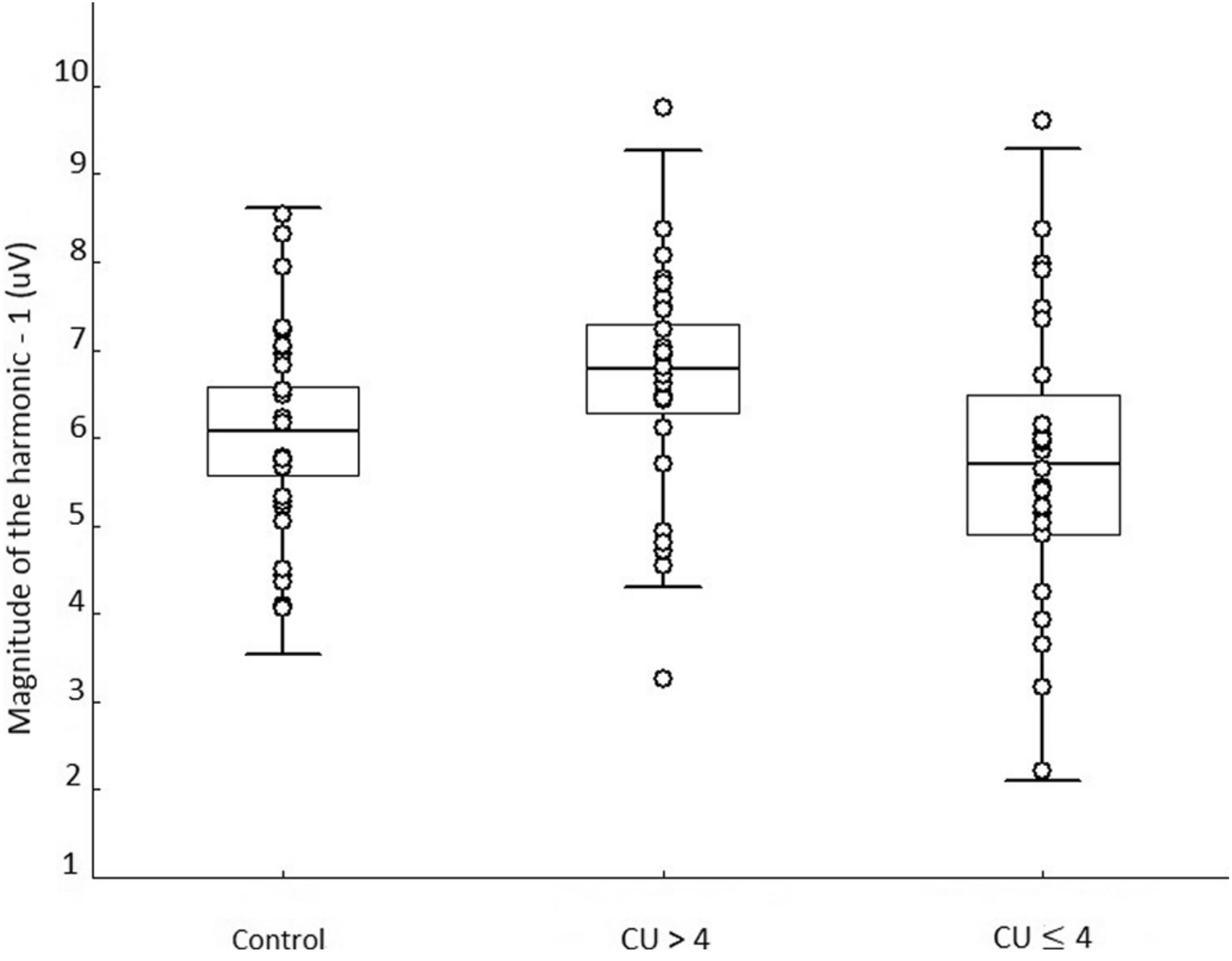

Figure 3

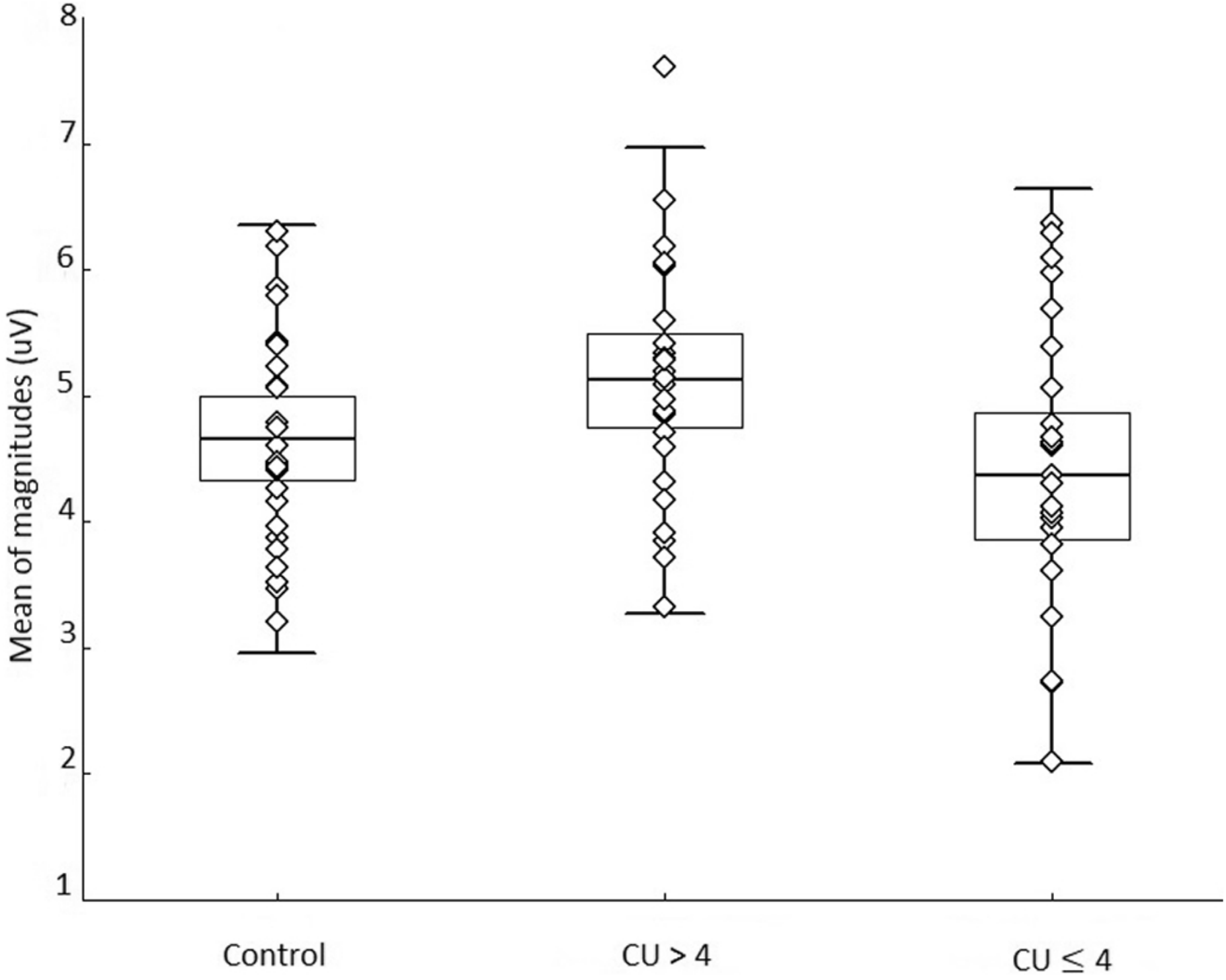

Figure 4