Clinical pharmacology of cannabinoids in early phase drug development

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5 Evaluation of THC-induced tachycardia in humans using heart rate variability

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Abstract

Cannabis use induces tachycardia, but its mechanism is unexplained. Heart rate variability (HRV) can provide information concerning effects of drugs on parasympathetic and sympathetic tone. HRV data of healthy male volunteers were used from two separate double-blind and placebo-controlled studies. Rising doses of pure THC were administered by inhalation with or without co-administration of the selective CB1 antagonist AVE1625. After THC administration, significant dose-related changes compared to placebo were seen in the 'time domain' on heart rate and SDSD. In the 'frequency domain' dose-related changes were seen on total power, low frequency power and high frequency power. Overall, normalized LF and HF and the LF/HF ratio did not change significantly. However, with the two highest THC doses, average values increased for LF and decreased for HF, leading to an average increase in LF/HF ratio. Co-administration of the selective CB1 antagonist AVE1625 had no effect on HRV under placebo conditions, but completely antagonized THC-induced effects on HRV. This indicates that HRV is mediated by CB1 receptors. These findings confirm the involvement of CB1 receptors in THC-induced tachycardia and suggest that the increase in heart rate caused by acute THC administration may be caused by a peripheral mediated reduction in the vagal tone.

Introduction

Delta-9-tetrahydrocannabinol (THC), a CB1/CB2 agonist, is the most abundant and major psychoactive cannabinoid identified in the plant *Cannabis sativa* L. Cannabis causes a pronounced increase in heart rate,[1,2] but its mechanism has not been fully elucidated.

An increase in heart rate can be established by direct or indirect effects. In the case of THC-induced tachycardia it may be caused by direct stimulation of CB1 receptors in human atrial muscle.[3] Indirect adaptation of heart rate is mainly mediated by the central nervous system and involves a change in the interaction between sympathetic and parasympathetic stimulation on heart rate. Although CB1 receptors are located in human atrial muscle,[3] CB1 receptors are predominantly situated in the brain with the highest densities in the hippocampus, cerebellum and striatum, which account for the well-known effects of cannabis on motor coordination and short term memory processing.[4-6] However, CB1 receptors are also expressed at low levels in the brainstem[6] where the cardiovascular control centers are located, making it possible that THC exerts its effects on heart rate via this route.

Propranolol, a non-selective beta-adrenergic blocking agent without sympaticomimetic activity, is able to antagonize tachycardia induced by THC.[7,8] This suggests that THC may increase heart rate by activation of the

sympathetic nervous system either centrally or peripherally. Pretreatment with atropine, a parasympatholytic drug, attenuates THC-induced tachycardia as well.[8] Pretreatment with both propranolol and atropine abolishes THC-induced tachycardia completely.[8] These findings suggest that both anticholinergic and beta-adrenergic effects contribute to the increase in heart rate after THC administration.[8] However, these data do not elucidate the balance between the parasympatic and sympatic nervous system involved in the increased heart rate. In addition, it cannot be determined from these experiments if the tachycardia is controlled by centers in the brain and spinal cord or that a direct effect on CB1 receptors in the heart is involved in THC-induced tachycardia.

Heart Rate Variability (HRV) analyses can provide information concerning the effects of drugs on parasympathetic and sympathetic tone.[9] In this study, the sympathovagal balance in THC-induced tachycardia was evaluated. In addition, co-administration of the selective CB1 antagonist AVE1625 will elucidate the role of CB1 and CB2 receptors in THC-induced tachycardia.

Methods

DESIGN

The heart rate variability (HRV) data originate from two double-blind and placebo-controlled studies. For both studies pure HRV was purified from *Cannabis sativa* according to GMP-compliant procedures and administered by inhalation using a Volcano® vaporizer (Storz-Bickel GmbH, Tüttlingen, Germany).[10]

Study 1[10]: Twelve healthy males (average 23 ± 2 years, range 21-27) with a history of mild cannabis use for at least one year were included in the study. On one study day, rising doses of THC (2, 4, 6 and 8 mg) were administered at 90 minute intervals. On a separate occasion, vehicle was administered in the same way, as double-blinded placebo. This study is described in full detail by Zuurman *et al.*[10] Heart rate was measured at baseline and 10, 35, 45, 55 and 85 minutes after each THC administration. HRV measurement was performed at baseline and 25 minutes and 85 minutes after each THC administration.

Study 2[11]: Thirty-six healthy males (average 21 ± 3 years, range 18-31) with a history of mild cannabis use for at least one year were included in the study. During each study period a single oral dose of the CB1 receptor antagonist AVE1625 (20, 60 or 120mg) or matching placebo was administered three hours prior four consecutive rising doses of THC (2, 4, 6 and 6 mg) or placebo were administered by inhalation at 60 minute intervals using a Volcano® vaporizer. Each subject received four out of the six available treatment

combinations. The treatment combination 'placebo AVE1625 + placebo THC' was used as a negative control (24 subjects received this treatment). All three single AVE1625 doses (20, 60 and 120 mg) were administered in combination with the rising doses of THC, but only the highest dose of 120 mg AVE1625 was administered in combination with 'placebo THC' to study the effects of the antagonist itself. This study is described in full detail by Zuurman *et al*.[11] Heart rate was measured at baseline and 11, 23, 32 and 43 minutes after each THC administration. HRV measurement was performed at baseline, after AVE1625 administration and 25 minutes after each THC administration. After the last THC administration 3 additional measurements were performed.

HEART RATE

Heart rate was measured in sitting position after a rest of approximately 5 minutes, twice pre-dose and repeatedly post-dose at fixed time-points. All measurements were carried out with an automated sphygmomanometer (Nihon Kohden, Life Scope EC, Tokyo, Japan).

HEART RATE VARIABILITY

Five-minutes ECG recordings using lead II were made using a CardioPerfect ECG machine (Welch Allyn, Delft, The Netherlands). Recordings were made at baseline, 32, 92 and 152 minutes after oral administration of AVE1625 and 37 minutes after each consecutive dose of THC. In addition, after the last THC administration two additional ECG recordings were made to study the decline of the effects. The recordings were analyzed using the software provided with the device which employs the methodology as described by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology.[9]

The parameters in the time-domain were the average RR-interval (corresponding to heart rate) and SDSD (standard deviation of consecutive RR-intervals, a measure of vagal activity).[9] For the frequency-domain we analyzed the total power (TP, total variability), low frequency power (LF, 0.01-0.08 Hz, measure of sympathetic activity), high frequency power (HF, 0.15-0.5 Hz, associated with respiratory sinus arrhythmia, and almost exclusively due to parasympathetic activity), and LF/HF (measure of sympathovagal balance).[9]

STATISTICS

All parameters were summarized by treatment and time, and were presented graphically as mean over time, with standard deviation as error bars. The parameters were analyzed separately by mixed model analyses of

variance (using SAS PROC MIXED, SAS for Windows V9.1.2, SAS Institute Inc., Cary, NC, USA) with treatment, period, time and treatment by time as fixed effects, with subject, subject by time and subject by treatment as random effect, and with the (average) baseline value as covariate.

Treatment effect was estimated using the average response of the values obtained in the 90 minutes after the final administration of THC. Contrasts were reported along with 95% confidence intervals. All HRV parameters were analyzed after log-transformation. Log-transformed contrasts were back-transformed resulting in geometric mean ratios with associated confidence intervals. These were expressed as percentage change from placebo.

Results

After THC administration, dose-related changes compared to placebo were seen in heart rate (Figure 1), SDSD (Figure 2), Total Power (TP), Low Frequency power (LF) and High Frequency power (HF) (Table 1). In the first study (n = 12), normalized LF power and normalized HF power and the LF/HF ratio did not change (Table 1). However, with the two highest THC doses, average values increased for LF and decreased for HF, leading to an average increase in LF/HF ratio (Table 1). In the second study normalized LF power increased with THC (Table 1). Normalized HF and the LF/HF ratio (Figure 3) did not change in study two (Table 1). Comparable to study one, with the two highest THC doses in study two, average values increased for LF and decreased for HF, leading to an average increase in LF/HF ratio (Table 1).

Co-administration of the selective CB1 antagonist AVE1625 in study two did completely antagonize THC effects on heart rate variability (Table 2). AVE1625 did not have an effect on heart rate variability itself (Table 2).

Discussion

This study evaluated the sympathovagal balance in THC-induced tachycardia using heart rate variability (HRV) analysis. THC (a CB1/CB2 agonist) increased heart rate in a dose-related manner in comparison to placebo (Figure 1). After the initial increase, heart rate decreased rapidly after each dose.

Time domain analysis showed that this increase in heart rate was associated with a decreased standard deviation of consecutive RR-intervals. This indicates a reduction in vagal tone.[12] The analyses of the frequency domain were more ambiguous, and both studies only showed average changes with the highest two THC doses. However, both studies demonstrated an increase in LF/HF ratio, a measure of sympathovagal balance. Although these

changes did not reach statistical significance, this finding also suggests that a lowering of vagal tone occurred. This is in agreement with findings from Newlin *et al.*,[13] who used vagal tone index, which is another non-invasive measure of tonic vagal inhibition of the heart. These investigators showed that heart rate was significantly increased and vagal tone significantly decreased at 5 and 30 minutes after smoking a 2.7% marijuana cigarette. This dose is comparable to the cumulative THC dose in our studies. The 5 minutes recording yielded a much greater decrease in vagal tone compared to the 30 minutes recording. The lack of a significant effect in the present studies may be due to the timing of the measurement of the vagal activity. Scheduling the HRV measurement closely after THC administration, e.g. after 5 minutes instead of after 25 minutes, may provide stronger evidence that THC-induced tachycardia is established by a reduction in the vagal tone.

Although our data indicate a reduction of the vagal tone, a direct effect of THC on the heart cannot be excluded. Also other indirect mechanism by THC cannot be excluded although these are unlikely. For instance, THC may induce orthostatic hypotension and accompanying sign of vasodilatation like facial flushing and conjunctival reddening.[1,14,15] In young healthy males a decrease in blood pressure may be directly compensated by an increase in heart rate. In these two studies blood pressure did not change and other observable signs of vasodilatation were not observed.[10,11] Another indirect effect by which THC may induce tachycardia could be via activation of PPARγ receptors for which THC is a ligand. Activation of these receptors leads to vasorelaxation through increased bioavailability of nitric oxide and hydrogen peroxide production.[16,17] This effect is mediated by a nuclear receptor resulting in altered gene expression. However, tachycardia in our study appeared to be a direct receptor-mediated pharmacological effect, because heart rate increased very rapidly in response to changing THC concentration. In the rat isolated aorta, these vasorelaxant effects were not inhibited by the selective CB1 antagonist rimonabant, but were inhibited by the selective CB2 receptor antagonist SR144528.[16] In the present study the selective CB1 antagonist AVE1625 completely antagonized THC-induced effects on HRV parameters. These observations demonstrate the involvement of the CB1 receptor in THC-induced tachycardia. However, it does not elucidate if they are involved in a direct or an indirect regulatory mechanism on cardiac functioning. Together with the observation that CB1 receptors are expressed at only low levels in the brainstem[6] where the cardiovascular control centre is located, the above mentioned observations favors an indirect and peripheral mediated regulatory mechanism involved in THC-induced tachycardia.

In summary, our findings confirm the involvement of CB1 receptors in THC-induced tachycardia and suggest that the increase in heart rate caused by acute THC administration may be caused by a peripheral mediated reduction in the vagal tone.

Figure 1 Least square means graph (SD) of heart rate time profile. AVE1625 administration at T = 0. THC administration: 2 mg at T = 3 hours; 4 mg at T = 4 hours; 6 mg at T = 5 hours; 6 mg at T = 6 hours. Arrows indicate drug administration.

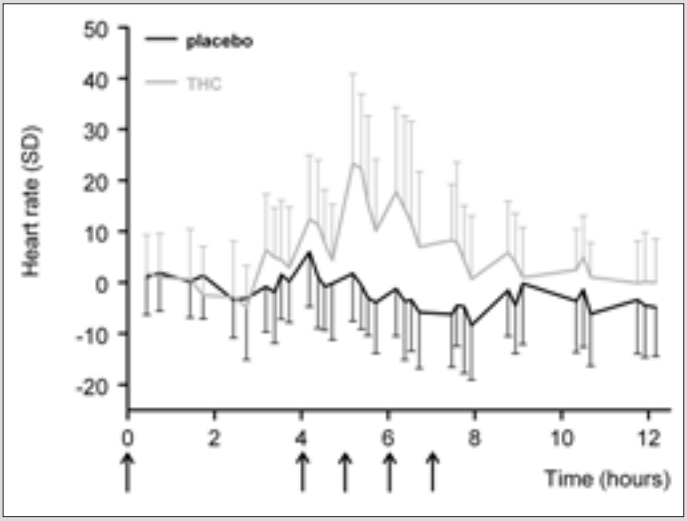

Figure 2 Least square means graph (SD) of SDSD time profile. AVE1625 administration at T = 0. THC administration: 2 mg at T = 3 hours; 4 mg at T = 4 hours; 6 mg at T = 5 hours; 6 mg at T = 6 hours. Arrows indicate drug administration.

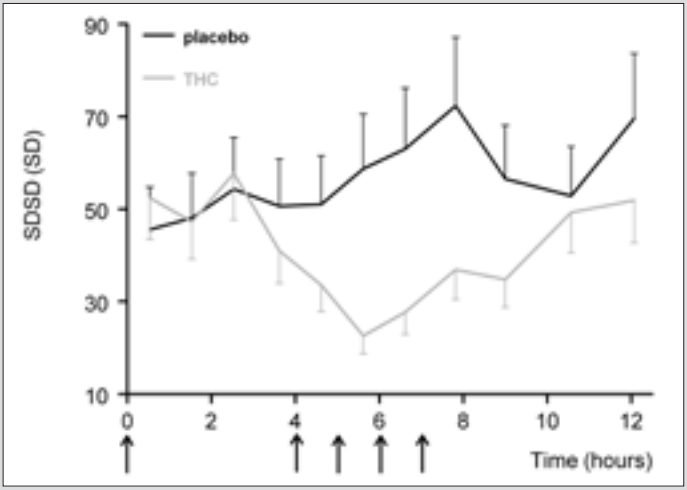

Figure 3 Least square means graph (SD) of LF/HF time profile. AVE1625 administration at T = 0. THC administration: 2 mg at T = 3 hours; 4 mg at T = 4 hours; 6 mg at T = 5 hours; 6 mg at T = 6 hours. Arrows indicate drug administration.

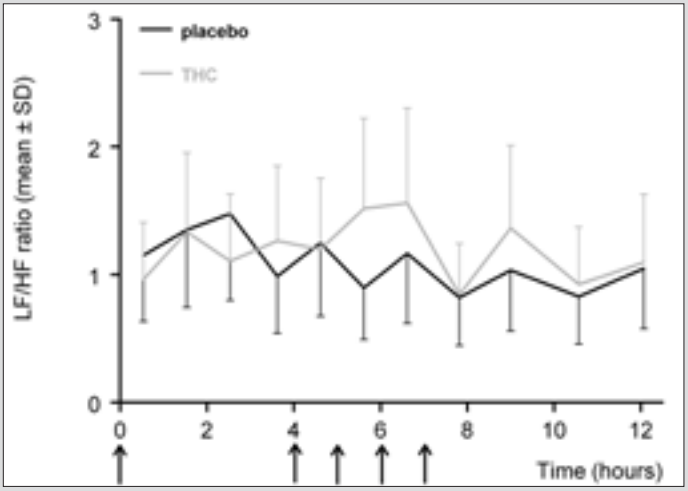

Table 1 Heart rate variability measurements after THC administration with 95% CI. Bold indicates a significant result.

Description	Variable	Study 1 (n=12)	Study 2 (n = 36)
	Heart rate (bpm)	**+19 (+13, +26)**	**+15 (+12, +19)**
Total variability	Total Power (ms^2)	**-71 (-81, -57)**	**-54 (-65, -40)**
Vagal activity	SDSD (ms)	**-49 (-58, -38)**	**-33 (-41, -24)**
Sympathetic activity	Low Frequency power (LF) (ms^2)	**-70 (-81, -52)**	**-55 (-66, -39)**
	Normalized Low Frequency power	+7 (-14, +34)	**+16 (+0.2, +35)**
Parasympathetic activity	High Frequency power (HF) (ms^2)	**-80 (-88, -67)**	**-67 (-77, -53)**
	Normalized High Frequency power	-20 (-42, +9)	-10 (-24, +6)
Sympathovagal balance	LF/HF ratio	+33 (-20, +121)	+29 (-5, +75)

Table 2 Heart rate variability measurements after co-administration of THC and AVE1625 with 95% CI (data from study 2). Bold indicates a significant result.

Description	Variable	Placebo + 120 mg AVE1625	THC + 120 mg AVE1625
	RR-interval (bpm)	+1% (-2, +5)	**+14% (+11, +19)**
Total variability	Total Power (ms^2)	+10% (-14, +41)	**+138% (+85, +206)**
Vagal activity	SDSD (ms)	+1% (-10, +14)	**+51% (+34, +70)**
Sympathetic activity	Low Frequency power (LF) (ms^2)	+1% (-23, +34)	**+124% (+70, +196)**
	Normalized Low Frequency power	-4% (-16, +11)	**-17% (-28, -4)**
Parasympathetic activity	High Frequency power (HF) (ms^2)	+7% (-25, +52)	**+224% (+128, +359)**
	Normalized High Frequency power	+6% (-10, +25)	**+18% (+0, +39)**
Sympathovagal balance	LF/HF ratio	-9% (-32, +22)	**-29% (-48, -6)**

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