

Cannabinoid Modulations of Resting State EEG Theta Power and Working Memory Are Correlated in Humans

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Abstract

■ Object representations in working memory depend on neural firing that is phase-locked to oscillations in the theta band (4–8 Hz). Cannabis intake disrupts synchronicity of theta oscillations and interferes with memory performance. Sixteen participants smoked cigarettes containing 0.0, 29.3, 49.1, or 69.4 mg Δ^9 -tetrahydrocannabinol (THC) in a randomized crossover design and performed working memory and general attention tasks. Dose-dependent effects of THC were observed for resting state EEG theta and beta power, working

memory (per-item search time), and attentional performance (percent errors and RT). The THC effects on EEG theta power and memory performance were correlated, whereas other EEG and behavioral effects were not. These findings confirm and extend previous results in rodents and humans, and corroborate a neurocomputational model that postulates that temporal aspects of information processing in working memory depend causally on nested oscillations in the theta and gamma (>30 Hz) bands. ■

INTRODUCTION

Phase-locking of neuronal firing to oscillations of local field potentials is instrumental in neural representations of behaviorally relevant information (Singer, 1993). The short-term maintenance of behaviorally relevant information is called working memory. Information in working memory is coded by object-selective neural signals in, for example, sensory cortex (Super, Spekreijse, & Lamme, 2001). These signals are phase-locked to local oscillations in the 4–8 Hz theta frequency band (Lee, Simpson, Logothetis, & Rainer, 2005). Accordingly, theta power in working memory tasks has been shown to increase during this task, both at the scalp (Ilan, Gevins, Coleman, ElSohly, & de Wit, 2005; Ilan, Smith, & Gevins, 2004; Jensen & Tesche, 2002; Klimesch, 1999; Gevins et al., 1998) and intracranially (Raghavachari et al., 2001).

According to a current neurocomputational model, the (repetitive) search through multiple items in memory would be phase-locked to theta oscillations. Individual items in memory would be separated by phase-locking to gamma oscillations nested within the theta rhythm (Jensen & Lisman, 1998). The model describes the reaction time (RT) distribution in a Sternberg working memory task, in which subjects decide whether a probe stimulus was part of a memorized set or not (Sternberg, 1975).

Cannabinoids such as Δ^9 -tetrahydrocannabinol (THC; the main psychoactive ingredient of cannabis) have long

been known to disrupt working memory (in rodents, e.g., Hampson & Deadwyler, 2000; and in man, reviewed in Ranganathan & D'Souza, 2006). Activation of CB1 cannabinoid receptors inhibits synaptic transmission at hippocampal pyramidal neurons. Cannabinoids consequently disrupt the synchronization of hippocampal theta oscillations in local field potentials in rodents, both in rest and during the performance of a delayed alternation (working) memory task. Recently, it has been shown that the cannabinoid modulations of theta synchronization and working memory performance in rodents are correlated (Robbe et al., 2006).

In humans, local decreases in synchronicity of local field potentials, that is, postsynaptic potentials, become manifest as a decrease of power in scalp-recorded EEG (Bastiaansen & Hagoort, 2006; Klimesch, 1999). In accordance with results in rodents, acute cannabis intake in humans induced a decrease in EEG theta power both in rest and during a working memory task, and led to a deterioration of working memory performance (Ilan et al., 2004, 2005). Here we tested whether a decrease in resting state theta power is correlated with deterioration of memory performance in humans.

The neurocomputational model holds that theta synchronization is a necessary condition for working memory performance, namely, memory search and comparison, in the Sternberg task. This causal proposition cannot be tested by demonstrating simultaneous pharmacological modulations of theta synchronization and memory performance because these would not exclude the alternative proposition that theta synchronization is an epiphenomenon of working memory operations. Yet, if it would be demonstrated

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that THC-induced theta desynchronization in a baseline condition (i.e., during resting state) correlates with the deterioration in working memory performance, then this would corroborate that theta desynchronization is causally related to memory performance. If theta power synchronization is only an epiphenomenon of memory performance, then THC effects on baseline theta synchronization would not correlate with working memory performance.

Furthermore, the neurocomputational model is specific for working memory. Therefore, the relationship between resting state theta power and working memory performance should also be specific and should not be related to general effects of THC intake, such as sedation. This specificity was addressed at various levels. First, to the extent that both effects are dose-related, they should not only be correlated over different subjects, but also within subjects over doses. To optimize the chances of observing dose-dependent relationships, the maximum dose tested (69 mg THC) was much higher than in previous studies. On the other hand, this dose is still ecologically valid as it approximates the maximum THC content of cannabis cigarettes in Great Britain (Potter, Clark, & Brown, 2008), The Netherlands (Niesink, Rigter, & Hoek, 2004), and North America (El Sohly, 2004). Second, we extended previous findings by employing a memory task that enables the estimation of per-item search time, which specifically depends on the working memory scanning stage, and not on other psychomotor stages (Sternberg, 1975). Third, the specificity of possible correlations was assessed exploring correlations with power changes in other EEG bands and with performance in a general attention task as well.

METHODS

Participants

Sixteen male volunteers (age = 18–45 years) participated in this study. They were recruited through advertisements in (local) newspapers. They were selected on the basis of their average cannabis use (between 2 and 9 cannabis cigarettes per month) to exclude both novice users and users with considerable tolerance to effects of cannabis (Perez-Reyes et al., 1991). Participants reported no history of psychiatric diseases, and no respiratory diseases, liver conditions, or cardiovascular problems. They neither use medications (chronically) nor take excessive amounts of alcohol. The study protocol was approved by the medical-ethical committee of the Utrecht University Medical Centre. Each participant was informed about the possible risks and signed an informed consent form. The study was conducted following the guidelines for Good Clinical Practice.

Design and Procedures

Cannabis Intake

The hypothesis was tested in a double-blind, placebo-controlled, crossover design. Participants smoked one of

four cannabis cigarettes (a mixture of 700 mg tobacco and 300 mg of cannabis), referred to as joints, on each testing day, separated by a washout period of at least 7 days. The four joints differed in THC levels, with the low dose corresponding to 29.3 mg THC, the medium dose to 49.1 mg, and the highest dose to 69.4 mg. A cannabis batch containing less than 9 µg THC was used to fabricate a placebo cigarette. Subjects were instructed to smoke the cigarettes according to a computer-paced procedure (i.e., 3 sec for getting ready, 2 sec for inhalation, 3 sec for breath holding, and 32 sec for normal breathing and relaxation). The whole cigarette was smoked in about 22 min.

Behavioral Tasks

Participants completed several tasks at various times after cannabis intake (Figure 1). Here we report on the memory-search task and on a general attention task that was recorded closely in time with the EEG. Working memory performance was assessed using the Sternberg's memory scanning test, performed 60 min after the onset of cannabis intake. Participants had to memorize four sets of digits, increasing in length (i.e., 2 to 5 digits), generating four increasing levels of memory load. Presentation of the memory set was terminated by the subject. Subsequently, on memorization, a series of probe digits was presented. Probe presentation was terminated by the response, or after 2000 msec, and followed by 500 msec presentation of a small square that was presented on the side of the correct response and served as feedback. The interstimulus interval between feedback and the next probe was 1000 msec. If a test item belonged to the memorized set and was a target, participants had to press the right button; if it was a lure and did not belong to the memorized set, they had to press the left button. In total, 56 targets and 56 lures were presented. Performance (in)accuracy was expressed as

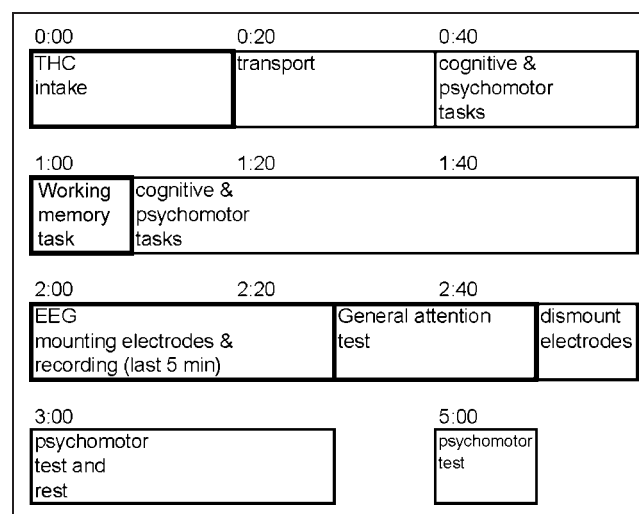


Figure 1. Time schedule of one testing day. Bold boxes indicate when data for the present analyses have been recorded.

percentage errors (including misses). The average RTs were further broken down into a per-item search time and a constant. Per-item search time characterizes the time needed to compare a probe to the items in memory. For each individual and each condition, a regression function was fitted to the RT values for the four different memory loads (memory set sizes 2 to 5), consisting of $y = ax + b$, where a = per-item search time, x = memory load, and b = the constant (or intercept; Jensen & Lisman, 1998; Sternberg, 1975).

The general attention task was performed 150 min after the onset of cannabis intake. Participants responded to one of four different visual stimuli by pressing a button, while ignoring the other three (Kenemans, Kok, & Smulders, 1993). The visual stimuli consisted of square-wave gratings, varying in fundamental spatial frequency (high or low, 0.6 and 4.8 cycles per degree of visual angle, respectively) and orientation (horizontal or vertical). The gratings subtended 6.67° of visual angle and were presented in the center of a computer screen, against a gray background. Each grating was presented for 50 msec. Eight consecutive blocks of 128 trials (stimulus onset asynchrony = 750 to 950 msec) differed with respect to target stimulus and responding hand. Behavioral-dependent measures were RT, its standard deviation (sdRT; reflecting possible lapses of attention), and the number of errors (omissions and commissions).

On the evening prior to the first exposure, subjects were individually trained to familiarize themselves with the tasks. Full data on all tasks are reported elsewhere (Böcker et al., submitted; Hunault et al., 2009).

EEG Measurement and Analysis

EEG electrodes were applied and the EEG was recorded just prior to the general attention task, during a 1-min interval in which the participants sat quietly with eyes closed. EEG was recorded from six midline (AFz, Fz, Cz, Pz, POz, Oz) electrodes using an electrocap. The right mastoid was used as a reference and a ground electrode was attached to the forehead. Electrodes placed above and below the right eye and at the outer canthi of both eyes were used to measure the vertical and horizontal EOG, respectively. Impedances were kept below 5 k Ω . Signals were amplified using Ampligraph (EEG Technology International BV, Levereij, the Netherlands) amplifiers with an on-line 100 Hz low-pass filter and were sampled at a rate of 250 Hz using Neuroscan Acquire software (Neuroscan, El Paso, TX).

The raw EEG signals were analyzed using BrainVision Analyzer software (Brain Products, Munich, Germany). Off-line filtering was applied (30 Hz low-pass, 12 dB/oct and high-pass, time constant 1 sec at 24 dB/oct) and the data were epoched for 50% overlapping 2-sec intervals, after which artifacts were removed and the data were corrected specifically for ocular artifacts (Gratton, Coles, & Donchin, 1983). Next, root-mean-square (RMS) power was calculated

for each interval using fast Fourier transform, after applying a 10% Hanning window, and these values were averaged over all intervals. The resulting resolution was 0.5 Hz. Average power in discrete EEG bands was submitted to statistical analysis. To assess the validity of a possible change in band-average theta power as reflecting a real theta power change, time–frequency plots were evaluated. Alternative possibilities are among others that the band-average power decreased due to a change in peak theta frequency, with some of the power at the sides of the peak extending partly beyond the predefined band. Alternatively, a change in band-average power might be secondary to a power change and/or peak narrowing or widening in a neighboring band. The time–frequency plots were derived by application of Morlet wavelets that resolved the power between 0.5 and 25 Hz into 10 slightly overlapping frequency bands, including 5–8.5 Hz.

Statistical Analyses

Both behavioral and EEG-power data were submitted to a repeated measures MANOVA to assess the effect of THC dose. The relationship between behavioral and EEG power measures was assessed in two ways. First, correlations between significant changes (69 mg placebo condition) in EEG power and behavioral measures were calculated. For significant power–behavior correlations, the individual consistency of this relationship was assessed, in turn, by calculating the correlation between EEG power and the behavioral measure concerned at each of the four doses for each subject separately. Subsequently, it was tested whether the group-average correlation differed from zero.

RESULTS

Performance

The RTs and percentage of errors in the Sternberg working memory task are shown in Figure 2. The MANOVAs of both RT and percentage of errors data showed main effects of dose [$F(3, 13) = 10.24, p < .005$ and $F(3, 13) = 4.34, p < .05$, respectively] and memory load [$F(3, 13) = 38.87, p < .0005$ and $F(3, 13) = 9.50, p < .005$], but no interaction [$F(9, 7) = 1.17, ns$ and $F(9, 7) < 1, ns$]. The linear effects of dose and memory load, indicating increasing RT and increasing numbers of errors with increasing doses of THC and increasing loads, were all significant [all $F(1, 15) \geq 12.7$, all $p < .005$]. After breaking down the RTs into per-item search time and a constant (see Figure 3), only the former revealed a cannabis effect [dose main effect: $F(3, 13) = 4.01, p < .05$ and $F(3, 13) < 1, ns$, respectively]. The per-item search time increased linearly with THC dose [$F(1, 15) = 12.6, p < .005$].

The mean RT, its standard deviation (sdRT), and the percentage of errors in the general attention task are shown in Figure 4. The MANOVA showed significant effects of THC

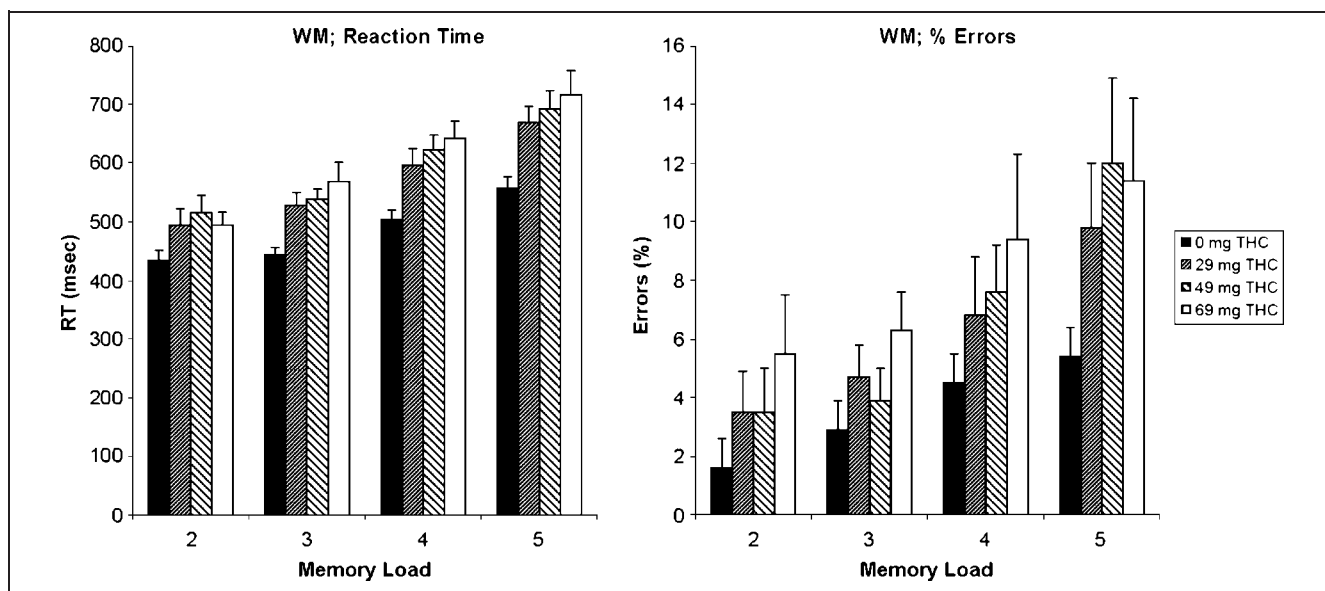


Figure 2. Working memory (WM) performance deteriorated after cannabis intake, both with respect to RT (left) and proportion of errors (right). Both measures increase with the length of the memory set.

dose on RT [$F(3, 13) = 4.42, p < .05$] and percentage errors [$F(3, 13) = 4.01, p < .05$], and was marginally significant for sdRT [$F(3, 13) = 3.20, p = .06$].

EEG Power

Figure 5 shows the RMS power in the different frequency bands. Dose-related effects were found in theta [$F(3, 13) = 9.18, p < .05$] and beta bands [12–30 Hz, $F(3, 13) = 3.51, p < .05$], but not in the delta [0.05–4 Hz, $F(3, 13) = 1.58, ns$] and alpha bands [8–12 Hz, $F(3, 13) < 1, ns$]. Theta

band power was smaller for each active dose compared to placebo (all $p < .005$). Furthermore, theta power was smaller after 69 mg than after smoking 29 mg THC intake ($p < .05$). The linear, quadratic, and cubic relationships between THC dose and theta power were all significant [all $F(1, 15) \geq 4.99$, all $p < .05$]. THC beta band power showed a quadratic relationship to THC dose [$F(1, 15) = 8.60, p < .05$]. Power in this band was larger after 49 mg than after smoking 69 mg THC intake ($p < .01$). Both theta and beta power were maximal at electrode Fz [at the midline between both hemispheres, over the frontal

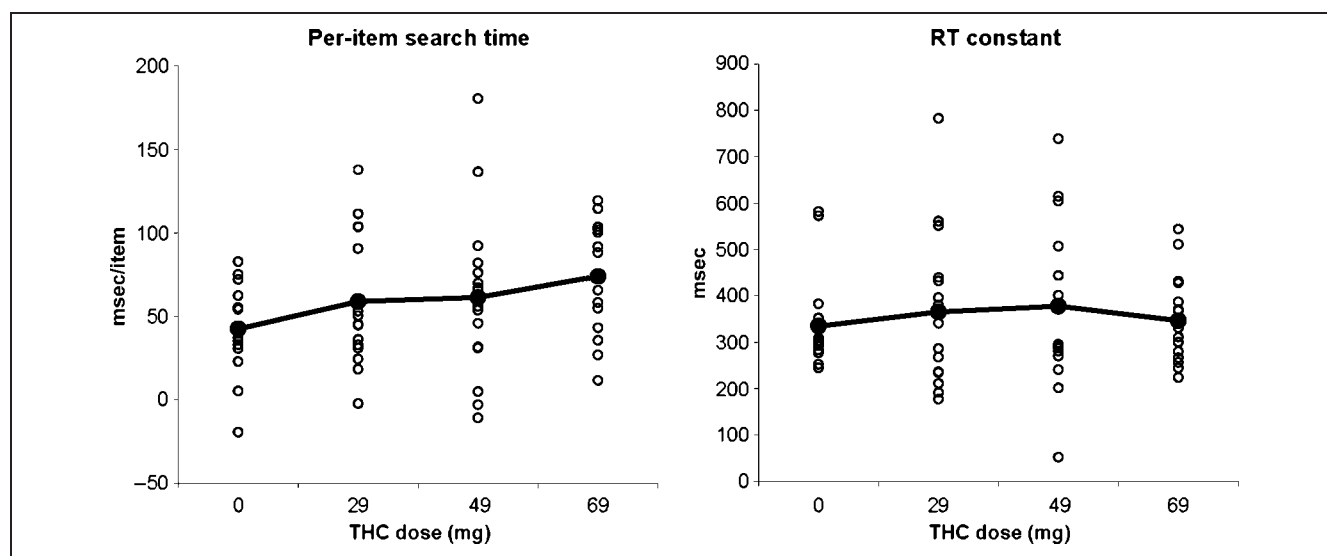


Figure 3. Individual and grand-average (bold) working memory performance after different doses of THC. RT for comparing probes to the memory set in the Sternberg task was broken down into per-item search time (left) and a constant (right). One hour after cannabis intake, only the former showed a statistically significant increase ($p < .05$).

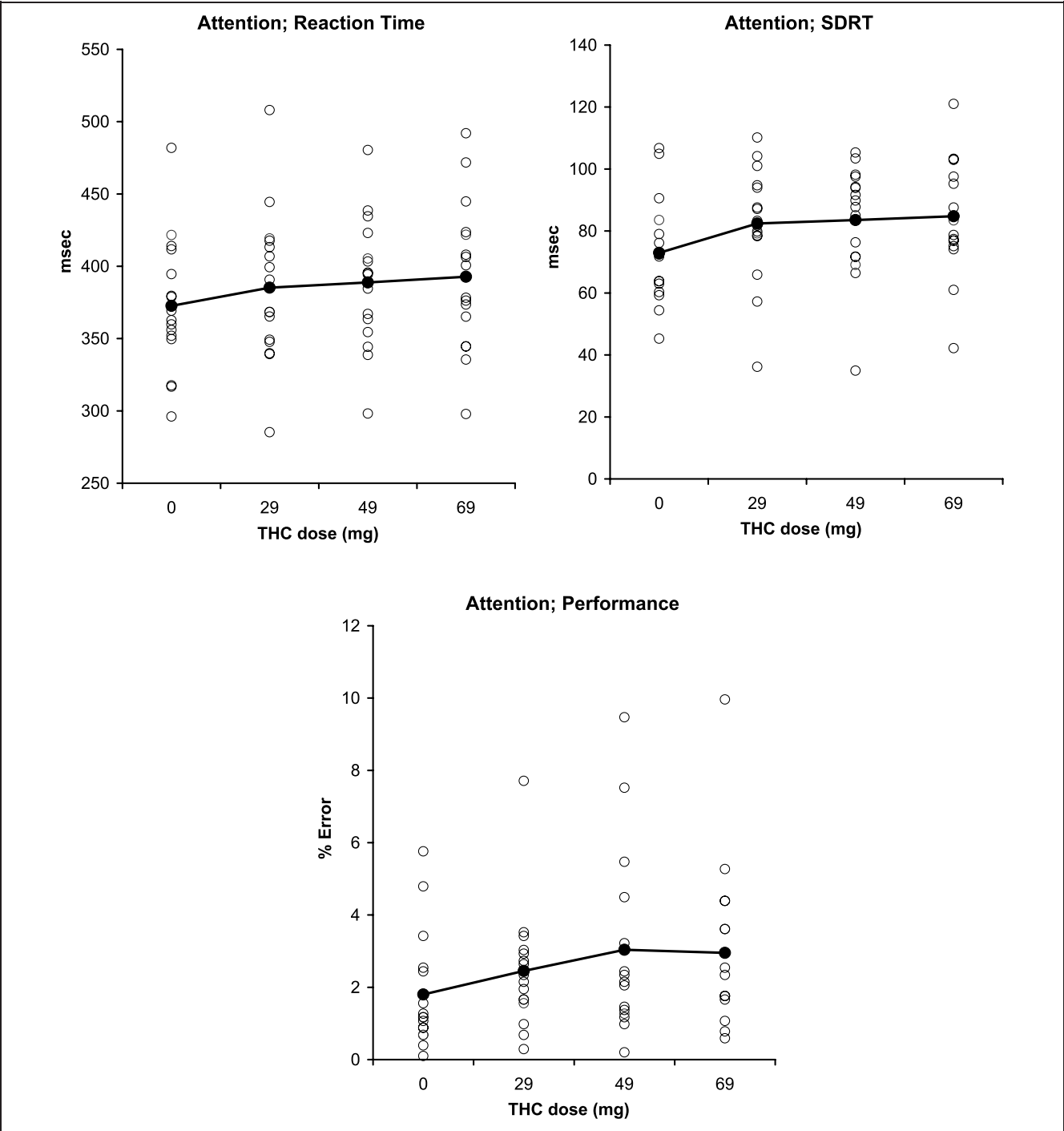


Figure 4. Individual and grand-average (bold) general attention performance after different doses of THC. Attention tended to deteriorate after cannabis intake with respect to RT (upper left panel) and proportion of errors (lower panel), and tended to do so for the standard deviation of the RT (SDRT; upper right panel).

lobes, main effect of electrode: $F(5, 11) = 56.98, p < .0005$ and $F(5, 11) = 98.14, p < .0005$, for theta and beta power, respectively]. Figure 6 shows the power spectrum of the EEG at this electrode.

To check the validity of a change in band-average theta power as a measure of a change in power of theta oscillations, individual time–frequency plots were inspected. Twelve of the 16 subjects showed theta oscillations at Fz,

which were independent of oscillations in neighboring bands for at least part of the time (Figure 7A and B show data of two of those subjects). In an attempt to quantify this subjective classification, 10 of these 12 subjects could be identified quantitatively by relatively high average theta power in the theta band under placebo conditions ($>0.80 \mu V$) and a relatively low multiple R for the regression of delta and alpha power over time on theta power over

time ($R < .25$). In the remaining participants, the theta power and its modulations seemed to reflect these in neighboring bands to a larger extent.

Correlations between Performance and EEG Power

Because both theta and beta power were maximal at electrode Fz, all correlational analyses were performed for data from this electrode. The correlation between THC modulations of theta power and per-item search time was

significant at the group level ($r = -.58, p < .05$; Figure 8). Furthermore, it was found to be consistent over cannabis doses at the individual level as well [mean correlation between individual theta power and per-item search time over doses, $r = -.30 \pm .14$ ($M \pm SEM$, after Fischer Z' transformation $t(15) = -2.35, p < .05$]. In contrast, the correlation between THC modulations of theta power and the number of errors on the working memory task was not significant ($r = -.34$).

To assess the specificity of the correlation between the increase in per-item search time and the decrease in theta

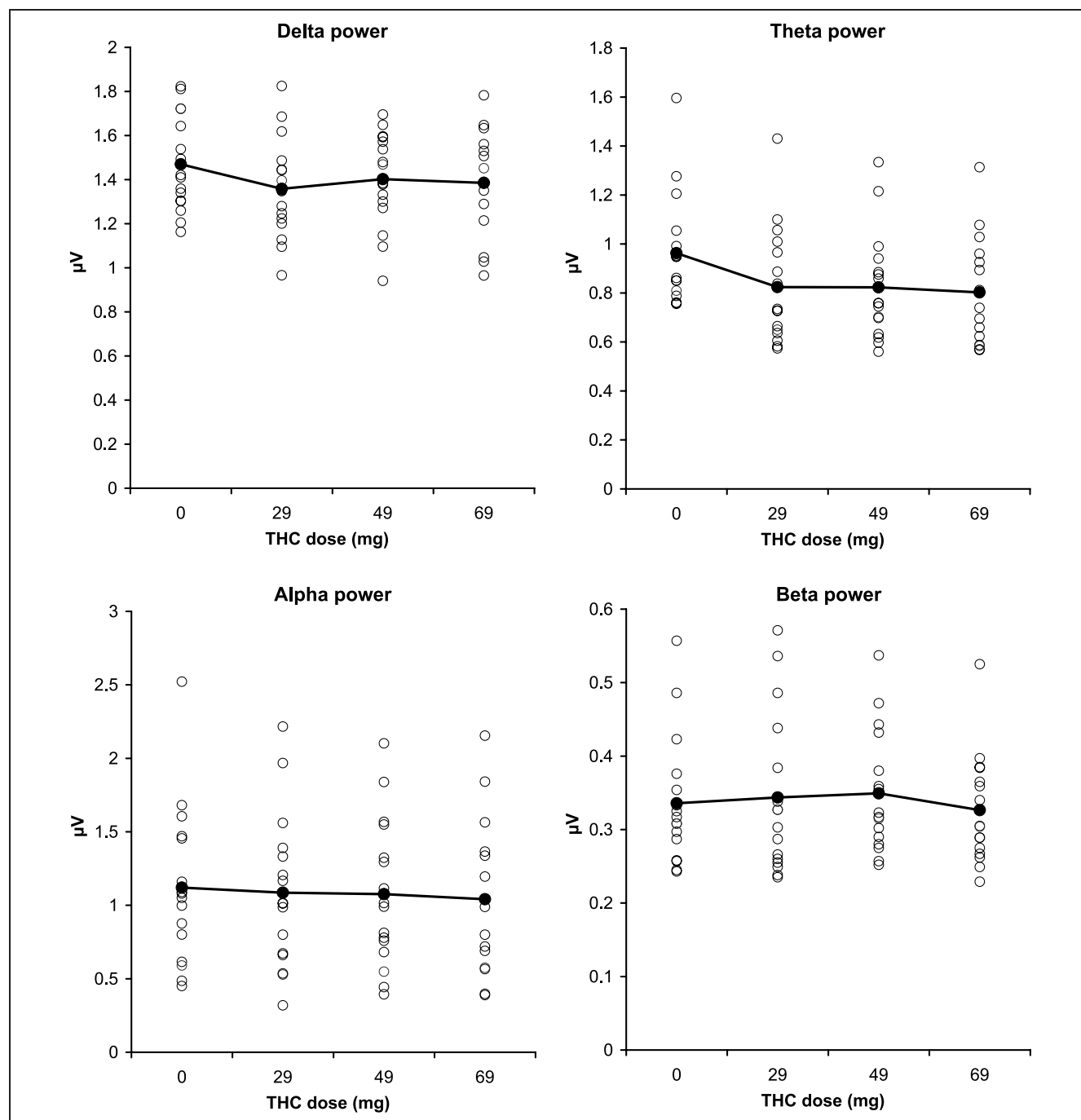


Figure 5. Individual and grand-average RMS power in each of the different frequency bands at electrode Fz.

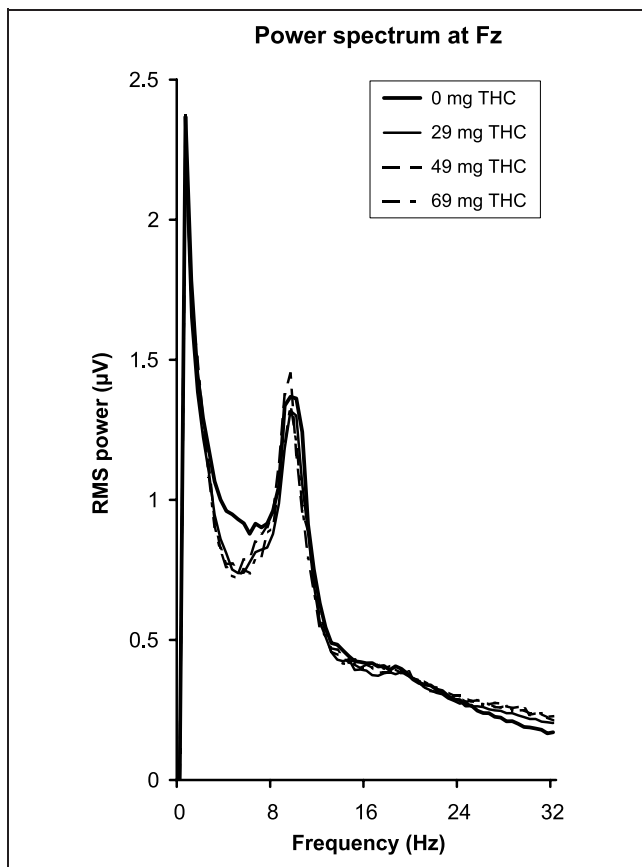


Figure 6. RMS EEG power in the theta band (4–8 Hz, average of 60 consecutive 2-sec intervals, eyes closed) decreased 2:20 hr after administration of a cannabis cigarette containing 29, 49, or 69 mg of THC compared to placebo ($p < .005$). Electrode Fz (at the midline between both hemispheres, over the frontal lobes) is shown. Theta power was maximal at this location.

power, correlations with dose effects in the beta band, as well as for performance on the general attention tasks, were calculated. All dose effects were expressed as contrasts between the highest dose and placebo. The correlation between the drug effects on beta power (at electrode Fz) and memory search time was $-.035$ and not significant. The correlations between theta power and performance measures from the general attention task were $-.25$ for RT, $-.19$ for sdRT, and $-.13$ for error percentage, all nonsignificant.

Finally, the group-level correlation between THC modulations of theta power and per-item search time was computed for the subgroup of 10 subjects that clearly showed independent theta oscillations according to the quantitative criteria outlined above. This amounted to $r = -.80$ ($p < .01$).

DISCUSSION

The present translational study set out to replicate and extend the finding that cannabinoid modulation of working memory and of synchronicity in theta oscillations were correlated. It was found that, in humans, THC intake de-

creased both theta power and working memory performance speed. With the extended dose range that was tested, both effects showed linear dose–response relationships. Moreover, these effects were correlated, replicating and extending the finding in rodents (Robbe et al., 2006), whereas other cannabinoid modulations were not correlated. A final indication of the specificity of the present findings was the observation that, among the performance measures derived from the Sternberg working memory task, only the per-item search time was affected by THC, not the constant psychomotor time.

The main aim of the study, a translational replication of the correlation between theta synchronicity and working memory performance, has been attained. The translational replication of the present brain–behavior correlation was not self-evident given the differences in theta generation in rodents and man. These differences also lead to different inferences regarding the physiological underpinning of the observed correlations. In rodents there is strong coupling between hippocampal and cortical theta during memory operations (Jones & Wilson, 2005; Siapas, Lubenov, & Wilson, 2005) that can also be observed in man (Gallinat et al., 2006). This model would explain the presently observed decrease in theta power as being a consequence of decreased firing synchronicity in the hippocampus, such as the desynchronization that followed application of cannabinoids in rats (Robbe et al., 2006). However, in man, cortical theta has been shown to have a larger contribution from local generators during sleep (Cantero et al., 2003) and working memory operations (Raghavachari et al., 2006). Therefore, the presently observed theta power decrease could also represent the effects of THC on synchronicity of local cortical generators of theta oscillations. As a consequence of THC-induced desynchronization of either distributed or local theta generators, object-specific firing patterns at gamma frequencies (above 30 Hz; Jensen, 2006; Jensen & Lisman, 1998) that are phase-locked to cortical theta oscillations might be expected to become desynchronized as well (Canolty et al., 2006; Lee et al., 2005). This would have compromised the quality of the memory representations. Therefore, the chances decrease that classification of a probe stimulus and the related response decision have completed at the end of a given search through the items in memory (taking one full theta cycle; Jensen, 2006; Jensen & Lisman, 1998). This would necessitate waiting for another memory search (i.e., theta cycle) before being able to decide and respond on some of the trials. In this way, performance would slow down and become more error-prone on average, as has been observed in the present study.

Results of intracranial EEG recordings also have significance for mechanistic interpretations of the present results. These recordings have shown that working memory-related theta modulations reflect phase-resetting of spontaneous theta (Rizzuto et al., 2003), in accordance with the neurocomputational model (Jensen, 2006; Jensen & Lisman, 1998). It could be assumed that THC-induced desynchronization of ongoing theta interferes with phase-resetting,

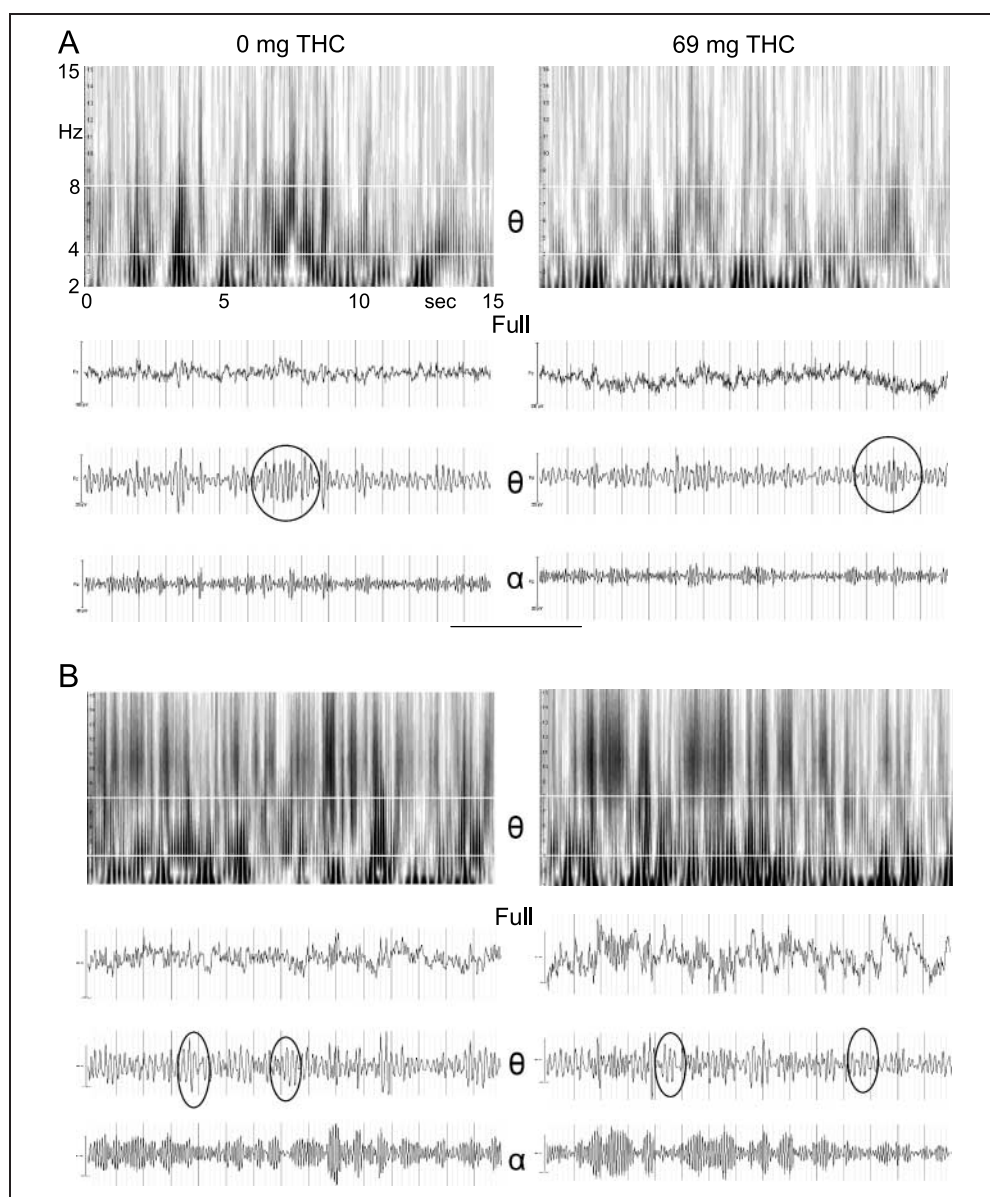
which again compromises the memory representations and slows down average performance. Alternatively, THC-induced theta desynchronization might also directly interfere with phase-locking by slowing down this process. To relate this slowing of phase-locking to the observed increase in working memory speed, it remains to be explained how this slowing would depend on memory load.

The present study also replicated previous studies into the effect of cannabinoids on working memory and EEG in humans. These also found a decrease in working memory speed and accuracy, as well as a decrease in EEG theta power, both in rest and concurrent with performance of the task (Ilan et al., 2004, 2005). In comparison with previous studies, the dose range was extended from 0–27 mg (Ilan et al., 2005) to 0–69 mg THC in the present study. Using the latter dose range, significant linear trends for performance speed and EEG theta power were observed. Given these dose-dependent effects, it is more probable

that the effects are receptor-specific rather than of a general nature. The dose dependency of both effects was also observed at the individual-subject level. On average, the correlations between theta power and working memory speed over doses were significantly positive.

Specificity was observed at several other levels. First, the correlations were specific with respect to frequency band. Per-item search time did not correlate with the observed beta power modulations. Second, specificity with respect to task was observed in that THC modulations in the general attention task did not correlate with the decrease in theta power. Third, within the task, only the per-item search time, and neither the constant part nor the number of errors, did correlate significantly with theta power. Likewise, theta phase-resetting in intracranial EEG was more pronounced after presentation of a probe stimulus than after presentation of a to-be-memorized item (Rizzuto et al., 2003). This phase-reset was interpreted as a reflection

Figure 7. (A) Time–frequency plot showing the power distribution from 2 to 15 Hz at electrode Fz over time. Shown are 15 sec of data under placebo and the highest THC dose. The white horizontal lines are at 4 and 8 Hz, respectively (i.e., at the borders of the theta band). Darker compared to lighter areas indicate time intervals with larger power at a certain frequency. Raw and filtered EEG in the theta and alpha bands is shown as well. Ellipses indicate the clearest instances of theta oscillations, which are clearly independent of either alpha or delta waves. This subject shows a marked reduction in theta power in the THC condition. (B) Corresponding data of a second subject, with marked amplitude fluctuations in the alpha band and relatively constant power in the theta band.



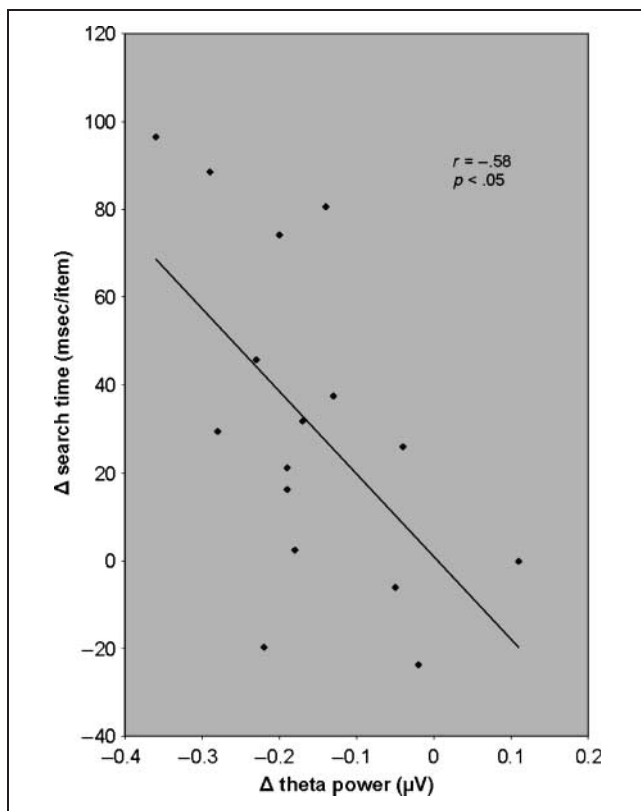


Figure 8. Scatterplot showing the correlation between the theta power decrease (69 mg–0 mg conditions) in Figures 5 and 6 and the increase in per-item search time (item) in Figure 3 (left) after cannabis intake. The more negative was the theta power difference, that is, the larger the power decrease, the larger was the elongation of per-item search time.

of memory comparison operation, such as the per-item search time (Sternberg, 1975). The alternative possibility put forward by Rizzuto et al. (2003), that theta phase-resetting was related to motor preparation, could be ruled out in the present study by the absence of a correlation between theta and the RT constant. The latter would have reflected modulations in the duration of general sensory and motor processing stages. The latter two levels of specificity are also difficult to reconcile with the conclusion that the THC-induced decrease in theta power would reflect increased arousal, and that the performance decrease in working memory would result from cannabinoid modulations of attention as suggested by Ilan et al. (2004, 2005). However, a firm conclusion would necessitate a study that combines the strong aspects of both studies (e.g., extend the present set-up with physiological manifestations of arousal and attention), and would take the relationship between attention and working memory into account.

An opposite account of the present data states that theta power increase is related to decreased activity in a so-called default mode network (Scheeringa et al., 2008), and vice versa. The default mode network consists of areas that are less active during goal-directed behavior. The core network includes the posterior cingulate, retro-

splenial, lateral temporal, and medial prefrontal cortices (Buckner, Andrews-Hanna, & Schacter, 2008). The latter area probably generates more scalp-recorded EEG theta when the default mode network is inactive (Scheeringa et al., 2008). With respect to the present results, an interpretation of cannabis modulations of theta power in terms of an increase in default mode network is strengthened by the finding that not only did theta power decrease, but at least with 29 and 49 mg THC, beta power increased. Similarly, default mode network activity has been shown to correlate not only with theta power decreases but also with beta power increases (Laufs et al., 2003). From this perspective, the observed correlation between theta power decrease and per-item search time would stem from a cannabis-induced increase in default mode network activity. Some of the effects of cannabis, such as disorganized thought, together with cognitive decline (D'Souza et al., 2004), could be explained by a cannabis-induced increase of default mode network activation that has been shown to be related to mind wandering, or stimulus-independent thought (Mason et al., 2007). Again, this is difficult to reconcile with the specificity of the observed correlation that was only significant for per-item search time and not for changes in attention as assessed by the general attention task. The present authors agree with Scheeringa et al. (2008) that more experimental work is needed to be able to conclude whether scalp EEG theta oscillations are specific pacemakers for scanning working memory or more generally related to decreases in default mode network activity.

The default network account of the present data, and of theta power in general, also defines the functional significance of resting state theta oscillations. The function of theta oscillations is generally defined in terms of active reentrant or looping information processing (as reviewed in Mitchell, McNaughton, Flanagan, & Kirk, 2008). The repetitive serial search through multiple items in working memory is an example of such reentrant processing. According to a default mode network account of theta oscillations in baseline EEG, these oscillations could be a free running rhythmicity, with theta power serving as a state parameter of the involved brain systems. High theta power would indicate increased preparedness for reentrant or looping processing. Recently, there has been much interest in resting state brain activity. fMRI recordings of resting state have been shown to consistently reflect fluctuations in the activity of 10 networks (Damoiseaux et al., 2006), including the default mode network. Their relationships with fluctuations of resting EEG oscillations are only starting to be explored (Laufs, 2008).

A potential limitation of the current study is that the recordings of theta power and memory performance were more than one hour apart. However, this did not obscure the hypothesized relationship, and provided a first test of causality, which was reinforced by the specificity of the present results at various levels. A causal role of theta synchronicity in memory function has also been deduced from recent findings in the rat. These showed that not only does

a disruption of theta synchronicity disrupt spatial memory, but moreover, that restoring the synchronicity (at theta frequency) by external electrical stimulation reinstates memory function (McNaughton, Ruan, & Woodnorth, 2006). This does not exclude, for instance, that in the present study the theta power decrease during baseline is an epiphenomenon of a possible THC-induced decrease in baseline working memory activity. However, the hypothesis of a causal theta–working memory relationship, that would clarify the mechanism of action of THC on working memory at least, seems a fruitful hypothesis for future testing. Given the limitations of behavioral tasks in rodents, and of pharmacological and physiological manipulations and recordings in man, such studies would probably require future translational studies of phase-resetting in monkeys (cf. Lee et al., 2005), or more advanced analysis in man (Onton, Delorme, & Makeig, 2005). Such an advanced analysis would also shed light on the dynamics of EEG theta oscillations during rest (as illustrated in Figure 7), and within and over trials during task performance, which is missing from average fast Fourier transform power values. Additional analysis of the present data confirmed that the main result, the correlation between THC-induced modulations in per-item search time and theta power, was indeed driven by subjects that showed robust theta oscillations, independent of those in the alpha and delta bands.

A second limitation of the present study was the use of a small number of exclusively midline electrodes. This precluded the use of EEG mapping, ICA or dipole modeling, or a combination of these techniques (e.g., Scheeringa et al., 2008) to assess the intracranial origin of the present findings. When such analysis techniques were employed, a fronto-central theta rhythm was indeed observed (Onton et al., 2005; Gevins, Smith, McEvoy, & Yu, 1997). The Fz maximum in the present data at least suggested that cannabis affected the frontal midline theta rhythm (Mitchell et al., 2008), as it did in a previous study (Ilan et al., 2004) which employed 40 electrodes.

In conclusion, THC modulations of EEG theta synchronicity and working memory speed are correlated humans as well as in rodents. These findings connect results from invasive neurophysiological experiments in animals with neurocomputational models of working memory based on analyses of working memory speed in man.

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REFERENCES

- Bastiaansen, M. C. M., & Hagoort, P. (2006). Oscillatory brain dynamics during language comprehension. In W. Klimesch & C. Neuper (Eds.), *Event-related dynamics of brain oscillations*. Amsterdam: Elsevier.
- Böcker, K. B. E., Gerritsen, J., Kruidenier, M., Hunault, C. C., Mensinga, T. T., & Kenemans, J. L. (submitted). Acute effects of cannabis with high Δ^9 -THC contents on visual selective attention; an event-related potential study.
- Buckner, R. L., Andrews-Hanna, J. R., & Schacter, D. L. (2008). The brain's default network: Anatomy, function, and relevance to disease. *Year in Cognitive Neuroscience 2008, Annals of the New York Academy of Sciences*, 1124, 1–38.
- Canolty, R. T., Edwards, E., Dalal, S. S., Soltani, M., Nagarajan, S. S., Kirsch, H. E., et al. (2006). High gamma power is phase-locked to theta oscillations in human neocortex. *Science*, 313, 1626–1628.
- Cantero, J. L., Atienza, M., Stickgold, R., Kahana, M. J., Madsen, J. R., & Kocsis, B. (2003). Sleep-dependent theta oscillations in the human hippocampus and neocortex. *Journal of Neuroscience*, 23, 10897–10903.
- Damoiseaux, J. S., Rombouts, S. A. R. B., Barkhof, F., Scheltens, P., Stam, C. J., Smith, S. M., et al. (2006). Consistent resting-state networks across healthy subjects. *Proceedings of the National Academy of Sciences, U.S.A.*, 103, 13848–13853.
- D'Souza, D. C., Perry, E., MacDougall, L., Ammerman, Y., Cooper, T., Wu, Y. T., et al. (2004). The psychotomimetic effects of intravenous delta-9-tetrahydrocannabinol in healthy individuals: Implications for psychosis. *Neuropsychopharmacology*, 29, 1558–1572.
- El Sohly, M. A. (2004). *Quarterly report potency monitoring project # 85*. National Center for Natural Products Research, University of Mississippi.
- Gallinat, J., Kunz, D., Senkowski, D., Kienast, T., Seifert, F., Schubert, F., et al. (2006). Hippocampal glutamate concentration predicts cerebral theta oscillations during cognitive processing. *Psychopharmacology*, 187, 103–111.
- Gevins, A., Smith, M. E., Leong, H., McEvoy, L., Whitfield, S., Du, R., et al. (1998). Monitoring working memory load during computer-based tasks with EEG pattern recognition methods. *Human Factors*, 40, 79–91.
- Gevins, A., Smith, M. E., McEvoy, L., & Yu, D. (1997). High-resolution EEG mapping of cortical activation related to working memory: Effects of task difficulty, type of processing, and practice. *Cerebral Cortex*, 7, 374–385.
- Gratton, G., Coles, M. G., & Donchin, E. (1983). A new method for off-line removal of ocular artifact. *Electroencephalography and Clinical Neurophysiology*, 55, 468–484.
- Hampson, R. E., & Deadwyler, S. A. (2000). Cannabinoids reveal the necessity of hippocampal neural encoding for short-term memory in rats. *Journal of Neuroscience*, 20, 8932–8942.
- Hunault, C. C., Mensinga, T. T., Böcker, K. B. E., Schipper, C. M. A., Kruidenier, M., Leenders, M. E. C., et al. (2009). Cognitive and psychomotor effects in males after smoking a combination of tobacco and cannabis containing up to 69 mg delta-9-tetrahydrocannabinol (THC). *Psychopharmacology*, 204, 85–94.
- Ilan, A. B., Gevins, A., Coleman, M., ElSohly, M. A., & de Wit, H. (2005). Neurophysiological and subjective profile of marijuana with varying concentrations of cannabinoids. *Behavioural Pharmacology*, 16, 487–497.
- Ilan, A. B., Smith, M. E., & Gevins, A. (2004). Effects of marijuana on neurophysiological signals of working and episodic memory. *Psychopharmacology*, 176, 214–222.

- Jensen, O. (2006). Maintenance of multiple working memory items by temporal segmentation. *Neuroscience*, *139*, 237–249.
- Jensen, O., & Lisman, J. E. (1998). An oscillatory short-term memory buffer model can account for data on the Sternberg task. *Journal of Neuroscience*, *18*, 10688–10699.
- Jensen, O., & Tesche, C. D. (2002). Frontal theta activity in humans increases with memory load in a working memory task. *European Journal of Neuroscience*, *15*, 1395–1399.
- Jones, M. W., & Wilson, M. A. (2005). Theta rhythms coordinate hippocampal–prefrontal interactions in a spatial memory task. *PLoS Biology*, *3*, 2187–2199.
- Kenemans, J. L., Kok, A., & Smulders, F. T. (1993). Event-related potentials to conjunctions of spatial frequency and orientation as a function of stimulus parameters and response requirements. *Electroencephalography and Clinical Neurophysiology*, *88*, 51–63.
- Klimesch, W. (1999). EEG alpha and theta oscillations reflect cognitive and memory performance: A review and analysis. *Brain Research Reviews*, *29*, 169–195.
- Laufs, H. (2008). Endogenous brain oscillations and related networks detected by surface EEG-combined fMRI. *Human Brain Mapping*, *29*, 762–769.
- Laufs, H., Krakow, K., Sterzer, P., Eger, E., Beyerle, A., Salek-Haddadi, A., et al. (2003). Electroencephalographic signatures of attentional and cognitive default modes in spontaneous brain activity fluctuations at rest. *Proceedings of the National Academy of Sciences, U.S.A.*, *100*, 11053–11058.
- Lee, H., Simpson, G. V., Logothetis, N. K., & Rainer, G. (2005). Phase locking of single neuron activity to theta oscillations during working memory in monkey extrastriate visual cortex. *Neuron*, *45*, 147–156.
- Mason, M. F., Norton, M. I., Van Horn, J. D., Wegner, D. M., Grafton, S. T., & Macrae, C. N. (2007). Wandering minds: The default network and stimulus-independent thought. *Science*, *315*, 393–395.
- McNaughton, N., Ruan, M., & Woodnorth, M. A. (2006). Restoring theta-like rhythmicity in rats restores initial learning in the Morris water maze. *Hippocampus*, *16*, 1102–1110.
- Mitchell, D. J., McNaughton, N., Flanagan, D., & Kirk, I. J. (2008). Frontal–midline theta from the perspective of hippocampal “theta”. *Progress in Neurobiology*, *86*, 156–185.
- Niesink, R. J. M., Rigtter, S., & Hoek, J. (2004). *THC-concentraties in wiet, nederwiet en hash in Nederlandse coffeshops, 2003–2004* [THC concentrations in cannabis, home-grown cannabis and hashish in Dutch coffeeshops, 2003–2004]. Utrecht, The Netherlands: Trimbos Institute.
- Onton, J., Delorme, A., & Makeig, S. (2005). Frontal midline EEG dynamics during working memory. *Neuroimage*, *27*, 341–356.
- Perez-Reyes, M., White, W. R., McDonald, S. A., Hicks, R. E., Jeffcoat, A. R., & Cook, C. E. (1991). The pharmacological effects of daily marijuana smoking in humans. *Pharmacology Biochemistry and Behavior*, *40*, 691–694.
- Potter, D. J., Clark, P., & Brown, M. B. (2008). Potency of Delta(9)-THC and other cannabinoids in cannabis in England in 2005: Implications for psychoactivity and pharmacology. *Journal of Forensic Sciences*, *53*, 90–94.
- Raghavachari, S., Kahana, M. J., Rizzuto, D. S., Caplan, J. B., Kirschen, M. P., Bourgeois, B., et al. (2001). Gating of human theta oscillations by a working memory task. *Journal of Neuroscience*, *21*, 3175–3183.
- Raghavachari, S., Lisman, J. E., Tully, M., Madsen, J. R., Bromfield, E. B., & Kahana, M. J. (2006). Theta oscillations in human cortex during a working-memory task: Evidence for local generators. *Journal of Neurophysiology*, *95*, 1630–1638.
- Ranganathan, M., & D’Souza, D. C. (2006). The acute effects of cannabinoids on memory in humans: A review. *Psychopharmacology*, *188*, 425–444.
- Rizzuto, D. S., Madsen, J. R., Bromfield, E. B., Schulze-Bonhage, A., Seelig, D., Aschenbrenner-Scheibe, R., et al. (2003). Reset of human neocortical oscillations during a working memory task. *Proceedings of the National Academy of Sciences, U.S.A.*, *100*, 7931–7936.
- Robbe, D., Montgomery, S. M., Thome, A., Rueda-Orozco, P. E., McNaughton, B. L., & Buzsaki, G. (2006). Cannabinoids reveal importance of spike timing coordination in hippocampal function. *Nature Neuroscience*, *9*, 1526–1533.
- Scheeringa, R., Bastiaansen, M. C. M., Petersson, K. M., Oostenveld, R., Norris, D. G., & Hagoort, P. (2008). Frontal theta EEG activity correlates negatively with the default mode network in resting state. *International Journal of Psychophysiology*, *67*, 242–251.
- Siapas, A. G., Lubenov, E. V., & Wilson, M. A. (2005). Prefrontal phase locking to hippocampal theta oscillations. *Neuron*, *46*, 141–151.
- Singer, W. (1993). Synchronization of cortical activity and its putative role in information-processing and learning. *Annual Review of Physiology*, *55*, 349–374.
- Sternberg, S. (1975). Memory scanning: New findings and current controversies. *Quarterly Journal of Experimental Psychology*, *27*, 1–32.
- Super, H., Spekreijse, H., & Lamme, V. A. F. (2001). A neural correlate of working memory in the monkey primary visual cortex. *Science*, *293*, 120–124.