Appendix 1 (as submitted by the authors)

Methods

Changes in ethics review regarding prior cannabis use

Initially, to avoid exposing naïve subjects to cannabis smoke, only subjects who had used cannabis in the past were eligible, but subsequent ethical concerns about subjects having to have engaged in an illegal activity to be eligible for a research study led to this criterion being removed.

GCMS methodology for THC, OH-THC and THC-COOH assays

Sample preparation

1 ml of serum samples with deteurated internal standards of THC, OH-THC, THC-COOH, were first deproteinized and centrifuged. The supernatants were pH adjusted to 6, then loaded onto pre-washed and pre-conditioned Certify solid phase columns. After samples had passed through, columns were washed and THC was first eluted off the columns and collected into tubes. Columns were washed again before eluting the OH-THC and THC-COOH using different solvents into another set of tubes. These eluants were then evaporated to dryness and derivatized for GCMS analysis.

Instrumentation

A Thermo DSQ quadruple GCMS was used for the analysis. A 30 m DB5-ms column was used with a temperature gradient. Analysis was done in SIM mode. Ions 386 and 389 were used for THC and D3 THC respectively; 371 was used for OH-THC, THC-COOH and 374 was used for D3 OH-THC and D3 THC-COOH. Sample quantitation was calculated against a set of standards with known concentrations.

Additional statistical analyses

A general mixed model with repeated measures was fitted to test changes of pain intensity and heart rate over time on day 1 of each treatment across treatment groups. A dose-response effect was examined using ANOVA across treatment groups. For the EQ-5D, items were recoded to dichotomous variables because of the distribution of responses. For mobility, responses were recoded to "no problem" and "some problem and serious problem". Pain responses were recoded to "no pain and moderate pain" and "serious pain". State of health was recoded to "feeling better" and "much the same and worse". A conditional logistical regression model was then fitted.

Results

Plasma THC levels showed that single inhalation of 25mg 9.4% THC resulted in a mean peak plasma THC concentration of 45.3 ng/mL at 2 minutes (n= 7); mean peak plasma levels for 2.5% THC were 10.2 ng/mL at 1 minute (n=8), for 6.0% THC were 23.5 ng/mL at 1 minute (n=8). No THC was detected following the 0% THC dose.

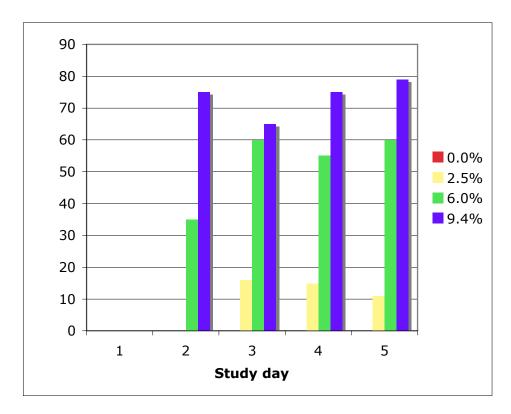Figure 3(S). Urine drug test results during on-drug phase

Figure 4(S). Plasma metabolites of THC following single inhalation of 9.4% THC