Estimating the experiment time as a function of concentration on b600.

Estimating the concentration.

Tune on proton and take a proton16 spectrum of your sample.

Integrate the solvent peak (7.28 ppm in chloroform-d, 2.5 ppm in DMSO-d6, 7.15 ppm in benzene-d6 or 3.30 ppm in methanol-d4) and one of the peaks in the analyte. Calibrate the solvent integral to the value in the table below and read the integral value for one proton in the analyte. This is the estimate for the concentration.

chloroform 99.8%	13
dmso 99.9%	33
benzene 99.5%	148
methanol 99.8%	50

These concentrations are below the real ones, e.g. 25 mM for chloroform-d 99.8 %, because the protons in the deuterated solvent did not relax between scans. The analyte nuclei didn't relax also, even in the ratio of the integrals is correct. The underestimation of concentration is ca. 20 %, depending on T1s. These errors are acceptable for our goal, to estimate the time for further experiments based on the proton16 spectrum you already have.

Estimating the experiment time

A menthol sample with a concentration of 3.3 mM, determined as above, gives a SNR of 256 in the proton spectrum in 1.5 minutes, a minimal SNR of 6 in the carbon spectrum in 18 min and a minimal hsqc spectrum in 12 minutes (4 scans x 128 increments). Halving the concentration quadruples the experiment time.

The minimum concentration which will give a minimal carbon spectrum (SNR = 6) in 12 hrs is 3.3 / SQRT(12*60/18) = 0.5 mM.

The minimum concentration to run indirect detection overnight (2 hrs for hsqc and 10 hrs for hmbc) is 3.3 / SQRT(2*60/12) = 1 mM.

To run a cosy spectrum on a window of 5 ppm (3000 Hz) 1024 increments will be needed for seeing cross-peaks from couplings larger than 3 Hz, which will take 25 minutes in one scan. The proton spectrum in one scan would produce a SNR of 64. The concentration for a SNR=8 would be 3.3/8 = 0.4 mM for a minimal spectrum in 25 minutes. Overnight is roughly 16 times more time, so one could get a cosy from a sample 0.1 mM.