Name: ________________________

BCMB/CHEM 8190, BIOMOLECULAR NMR
MIDTERM – 10/09/02

Instructions: This is an open book, limited time, exam. You may use notes you have from class and any text book you find useful, but you must bring materials you plan to use with you to the exam; you cannot leave the room to get other materials. You may also use a calculator. You will have 50 minutes to complete the exam. Write your answers and name on the exam; turn it in at the end of the period.

Part I (50 pts). Give short answers to the following:

1) The four lines of the AB spectrum above have approximate relative intensities of 0.6, 0.4, 0.4, 0.6. What can you say about the sign of the coupling?

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2) We have a strong signal –100 Hz from the center of a quadrature spectrum collected with a total spectral width of 1000 Hz. If the two channels in our quadrature receiver amplify to slightly different extents, at what frequency would you expect to see an artifact?

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3) A three spin system has evolved so that its density matrix can be represented by the product operator I1zI2xSz. We apply a 90 pulse along the x axis that affects only I type spins. Represent the density matrix in terms of a new three spin product operator.

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4) In exponential multiplication what is the best line broadening value to use when natural line widths are about 5Hz (in Hz) and why?

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5) We run a TOCSY and a COSY on a peptide containing an alanine residue (amide proton, alpha proton and three protons on a beta methyl group). What crosspeaks

(connectivities) do you expect to see in the TOCSY that you don't see in the COSY? _

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6) In nucleic acids, imino protons are usually detected between 10 ppm and 15 ppm. Which value of spectral width (in Hz) would you use to detect these signals given that the carrier is positioned on water (4.773 ppm), you want to minimize the spectral width as much as possible, and you are working with a 500 MHz NMR spectrometer?

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7) A NOESY spectrum of an alpha helix shows cross peaks connecting an amide proton to both the amide protons of the sequential residues (i – 1 and i + 1) and the alpha proton of a residue 3 amino acids removed (i – 3). The distances to the i +1 amide is 2.8 Angstroms and the intensity ratios for the connectivitiy peaks are 1.0 : 1.0 : 0.3. What are the distances to the other protons? _

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8) In a homonuclear J-resolved spectrum which of the following do you expect to be removed from the indirect dimension, chemical shift offsets, line broadening from a heterogeneous magnetic field, homonuclear scalar couplings, heteronuclear scalar couplings?

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9) We achieve a signal to noise ratio for a peak of interest of 4:1 after collecting 16 scans. What do you expect for the signal to noise ratio after 256 scans?

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10) We are interested in ^{13}C chemical shifts of an oligosaccharide that has resonances spread over 20 ppm and want to measure shifts to approximately 1 ppm accuracy. Which would take less time to collect to a comparable S/N ratio, a 1D ^{13}C spectrum at 125 MHz or an indirectly detected (through ^{1}H) HSQC spectrum on the same spectrometer at 500 MHz? (Assume the same slow repetition rate allowing complete recovery of magnetization between acquisitions. Also assume no special tricks to enhance magnetization in the ^{13}C 1D spectrum).

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Part 2 (25 points) Answer the following showing your work.

The following is a density matrix for a pair of spin ½ nuclei that are weakly coupled (AX). In fact, the two spins are of different nuclear types (^{1}H, ^{15}N). Assume we have used the basis set order, αα αβ βα ββ, in writing our density matrix. A table showing matrix representations of some operators for a pair of spins , I, S, is included at the end of the exam.

0.1	0.0 + 0.1i	0.6 - 0.2i	-0.5 + 0.4i
0.0 - 0.1i	0.2	-0.2 + 0.5i	0.1 - 0.3i
0.6 + 0.2i	-0.2 - 0.5i	-0.1	-0.4
-0.5 - 0.4i	0.1 + 0.3i	-0.4	0.3

a) Using matrix methods find the magnetization observable on the y axis in the frequency range of the first nucleus (^{1}H).

b) What is the difference in populations of states in which the second spin (^{15}N) is beta versus alpha and the hydrogen spin is beta?

Part 3 (25 points) Answer the following showing your work.

A two quantum filtered COSY is often used to improve line shapes and eliminate non-coupled peaks from a COSY spectrum. The following shows a version to be applied to a system having a pair coupled protons (one at zero chemical shift offset) and a single isolated proton at zero chemical shift offset. Using a particular t1 point for which the time is equal to 1/(2J) and assuming the delay between the last two pulses is very short, show what happens to the coupled spin at zero offset and the isolated spin at zero offset using product operators and beginning with I1z and I3z.

Is the magnetization that began on spin 1 on spin 1 or spin 2? Is the magnetization that began on spin 3 observable in t2?