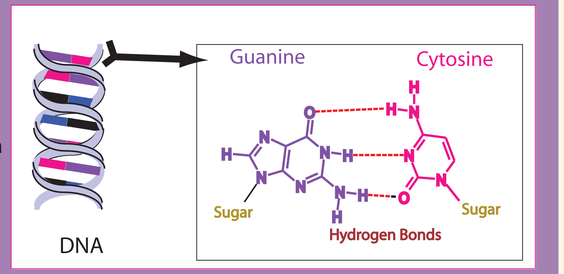
Elucidating Hydrogen-Bonding Structure in Novel Synthetic Guanosines by Solid-State NMR

Jenna Holder, Department of Physics
Supervisor: Dr. Steven Brown

The Role of Guanosine in DNA and RNA

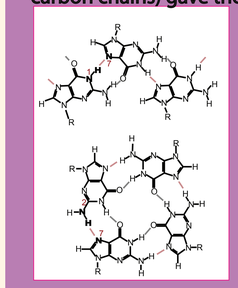
DNA contains guanine-cytosine base pairs, which are connected via hydrogen bonds.



Guanosine = guanine + ribose (sugar) - found in RNA
Deoxyguanosine = guanine + deoxyribose (sugar) - found in DNA

Predicted Structures

Two previously studied deoxyguanosine derivatives (with different lengths of carbon chains) gave these hydrogen-bonded macromolecular structures [1]:

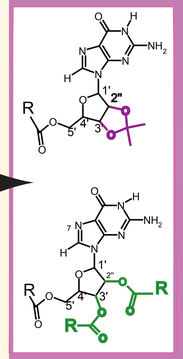


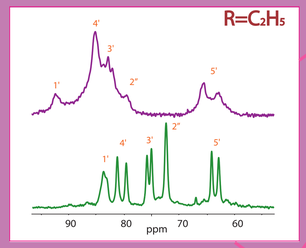
Ribbon Structure:
Two molecules per asymmetric unit cell (each spectral peak is split into two)
Typical of the short carbon chain derivative and of one polymorph of the long chain derivative.

Quartet Structure
One molecule per asymmetric unit cell.
Typical of the long chain derivative.

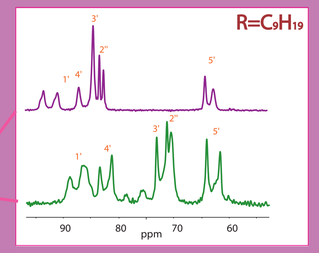
We can therefore say that if splitting is exhibitied, the molecular structure stands a good chance of being ribbon shaped.

13C Cross Polarisation Results



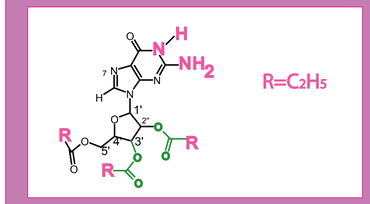


The lines here are too broad to determine whether there is splitting or not.



Ordered peaks, split into two. Possibly indicative of the ribbon macromolecular structure

2D 1H-1H Double Quantum Correlation Results



Intermolecular bonding - the two groups of atoms are not close to each other on the molecule so must be intermolecularly bonded.

Intramolecular bonding - the two groups of atoms are adjacent on the molecule

Next Step

Heteronuclear Correlation Experiments especially N-H as bonding tends to happen on the nitrogen-rich part of the molecules.

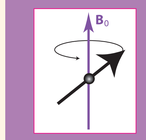
The URSS Experience

I had a really interesting time on my project and I can see that it will be extremely useful, not only in my final year project, but in any future research career. It is a worthwhile experience that I highly recommend to everyone. Many people connected to URSS were extremely helpful to me during my project, but I would particularly like to thank my supervisor, Dr. Steven Brown, as well as Amy Webber and everyone in the NMR Department for their time and support.

References:
[1] Pham et al, Identification of 15N Refocused INADEQUATE Solid-State NMR of Intermolecular Hydrogen Bonding that Directs Self-Assembly of Modified DNA Bases , *JACS*, **2005**

The NMR Process

NMR identifies chemical environments and gives spatial information within and between molecules.



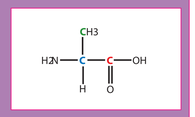
The sample is placed in a large magnetic field (B_0), which nuclei precess around with a characteristic frequency, ω_0

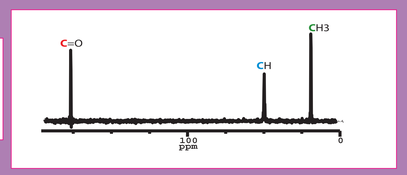
Electrons from adjacent atoms shield the nucleus from B_0, with shielding (σ)causing a shift in its precession frequency (to ω_0^S).

$$\omega_0^S = \omega_0(1-\sigma)$$

therefore each different chemical site precesses at a different frequency

E.g. alanine

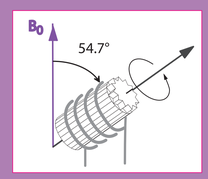




Where the shift value is given by:

$$\delta_{ppm} = \left[\frac{(\omega_0^{cs} - \omega_0^{ref})}{\omega_0^{ref}}\right] \quad 10^6$$

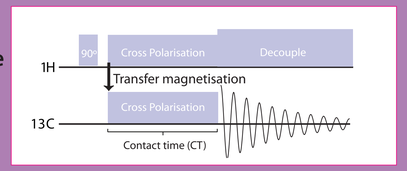
Each experiment provides spectral information on one type of atom, in this case carbon and then hydrogen. Samples are placed in rotors which are tilted at 54.7 ° (the magic angle) from B_0. This ensures that sharp spectral peaks are produced.



13C Cross Polarisation

For carbon, NMR is only able to obtain a signal from the 13C isotope. Unfortunately these only make up 1% of carbon atoms.

Solution to improve signal:noise ratio: Cross Polarisation- Transfer of magnetisation from 1H to 13C



2D Double Quantum Correlation Experiments

Between any two atoms exists what is known as a dipolar interaction, D:

$$D \propto \frac{1}{r^3}$$

D weakens considerably with increasing atomic separation, r, so if you can see an interaction, you know the atoms are in close proximity (<3A)

This interaction can be observed using a 2D spectrum

These two peaks emerge if atoms A and B are close to themselves

These two peaks show that atom A is close to atom B.