Experiences with a Nitrogen-cooled cryogenic probe

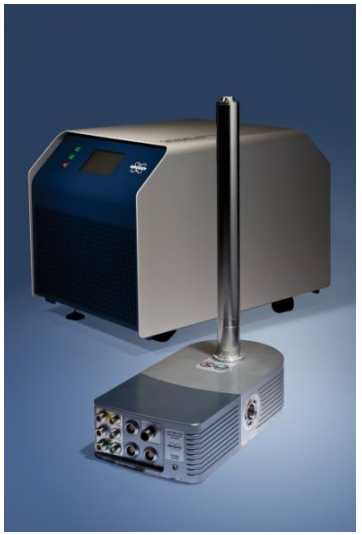
Tim Claridge

600 MHz N_2-cooled cryoprobe

- Installed Nov 2013
- System in use from Jan 2014
- 5 mm Multinuclear observe (BB)
- H/F outer coil
- VT 0-135 ^{o}C
- Auto tune/match

N_2 cooled probe
BB-H[F] @600 MHz

- Primary interest in H/C/F/P/N studies supporting “biomedical” research projects (Wellcome Trust)
  - Identifying & quantifying ^{13}C labelled metabolites
  - Following phosphorylation chemistry
  - ^{1}H and ^{19}F ligand binding assays
  - Ligand binding through ^{15}N HSQC protein screening
  - ^{19}F labelled proteins
  - Structural characterisation of synthetic “probes” of biological targets
- Special probe modification: no ^{19}F background
- No capacity to support additional He cryoprobes
  - Dual C/H
  - Triple H/C/N



(Bruker)

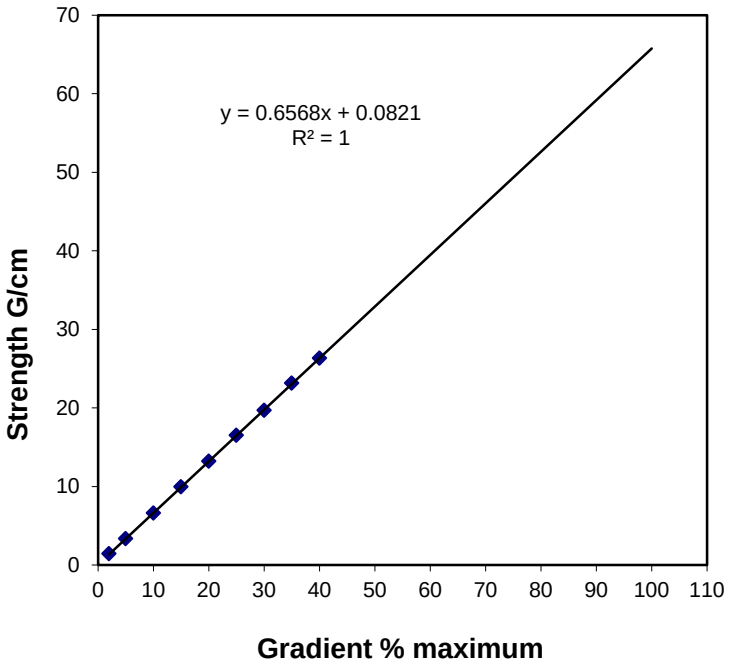
Sensitivity performance

	AVIII 600 Prodigy BB-H/F	AVIII 600 RT SMART BBF-H	AVIII 700 He TCI H/C/N	AVII 500 He Dual C/H	AVII 500 RT TXI H/C/F	AVIII 400 RT SMART BBF-H
^{1}H S/N (0.1 % EB)	1600:1	875:1	7800:1	2380:1	900:1	530:1
^{13}C S/N (ASTM)	1210:1	330:1	1560:1	1680:1	170:1	220:1
^{19}F S/N (0.05 % TFT)	1760:1	700:1	-	-	410:1	550:1
^{31}P S/N (48.5mM TPP)	728:1	250:1	-	-	-	210:1

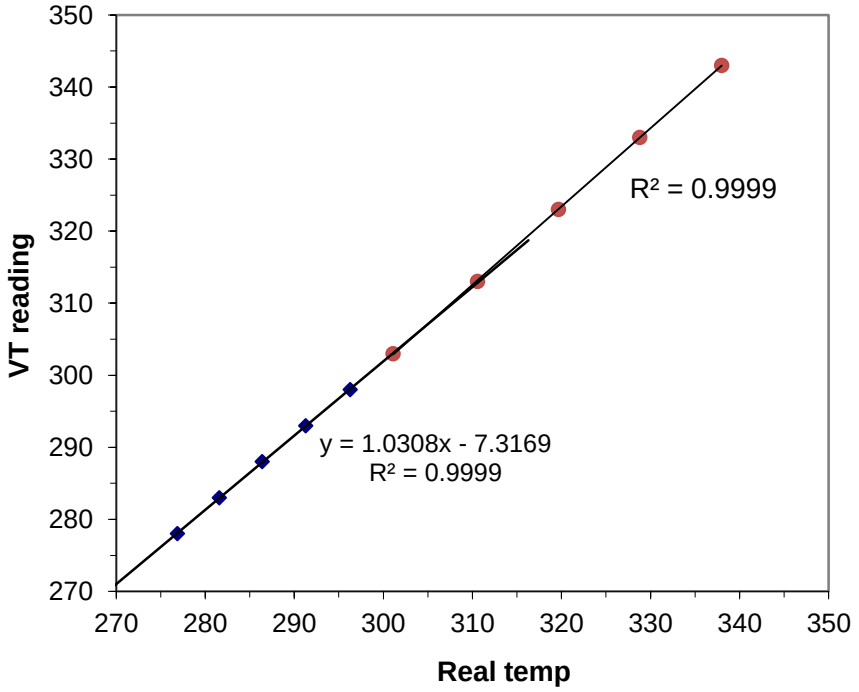
(Bruker specs)

Calibrations

Field Gradients
66 G cm^{-1}

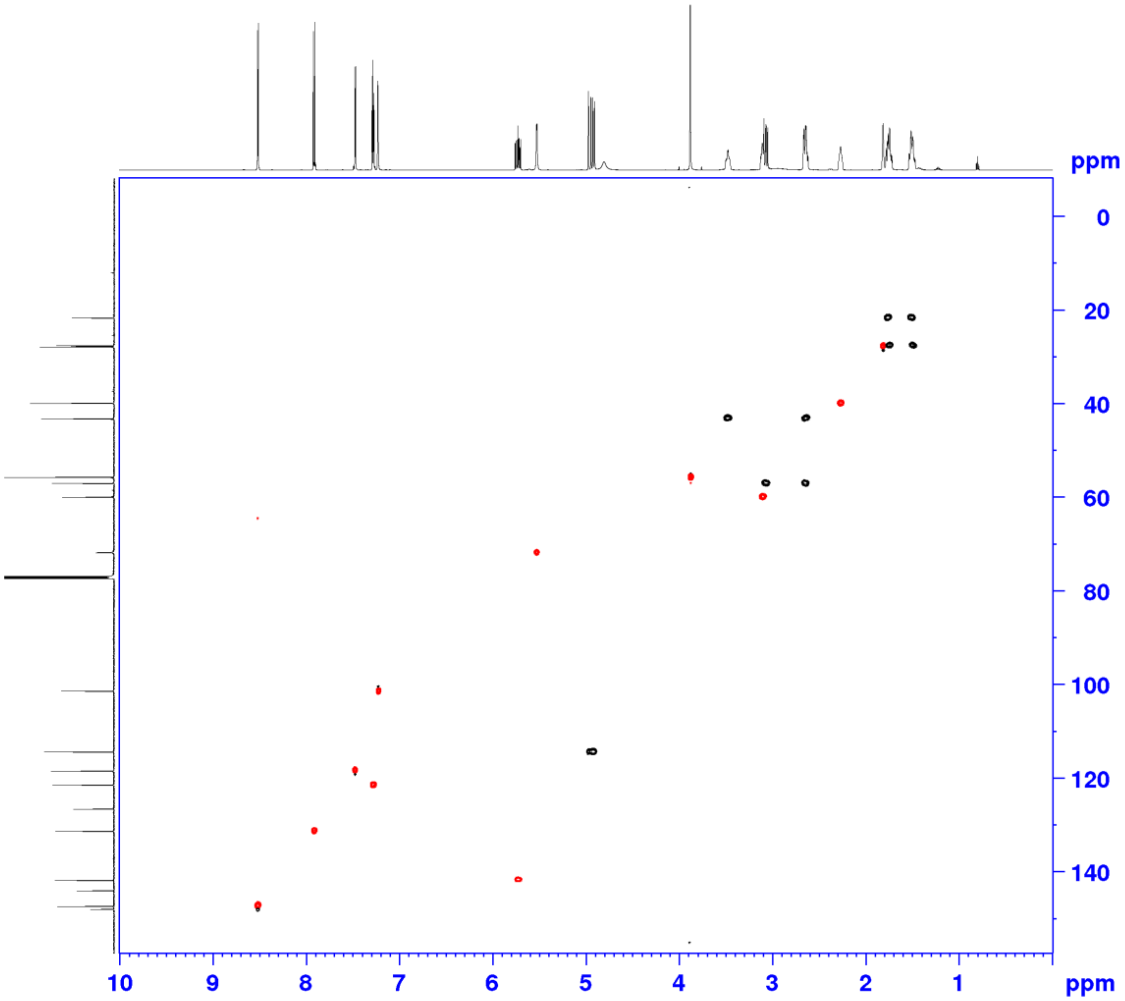


Temperature
MeOH, Ethylene glycol



Applications

Structure characterisation

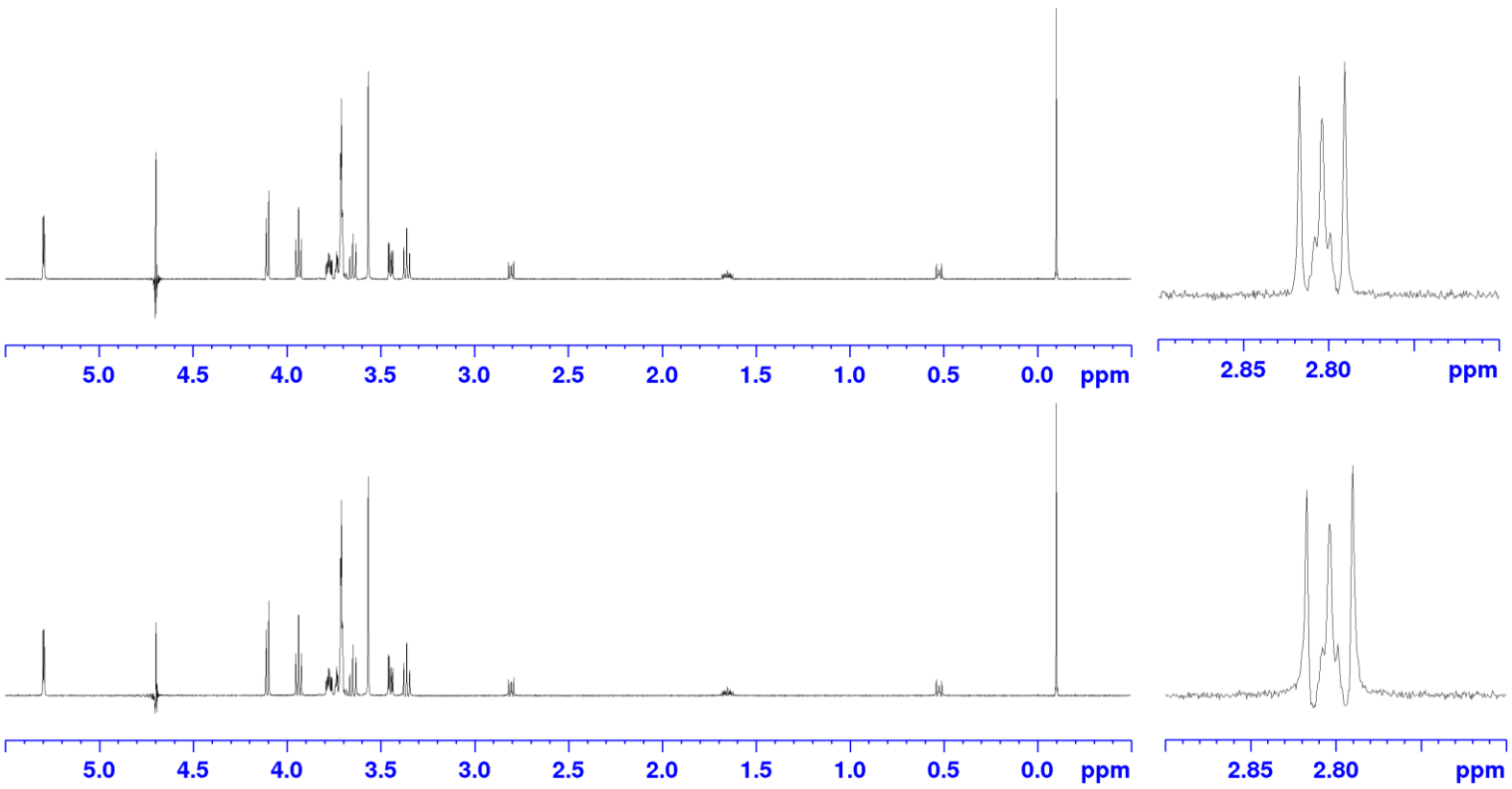


Quinine 50 mM
NS=1
5 min

Applications

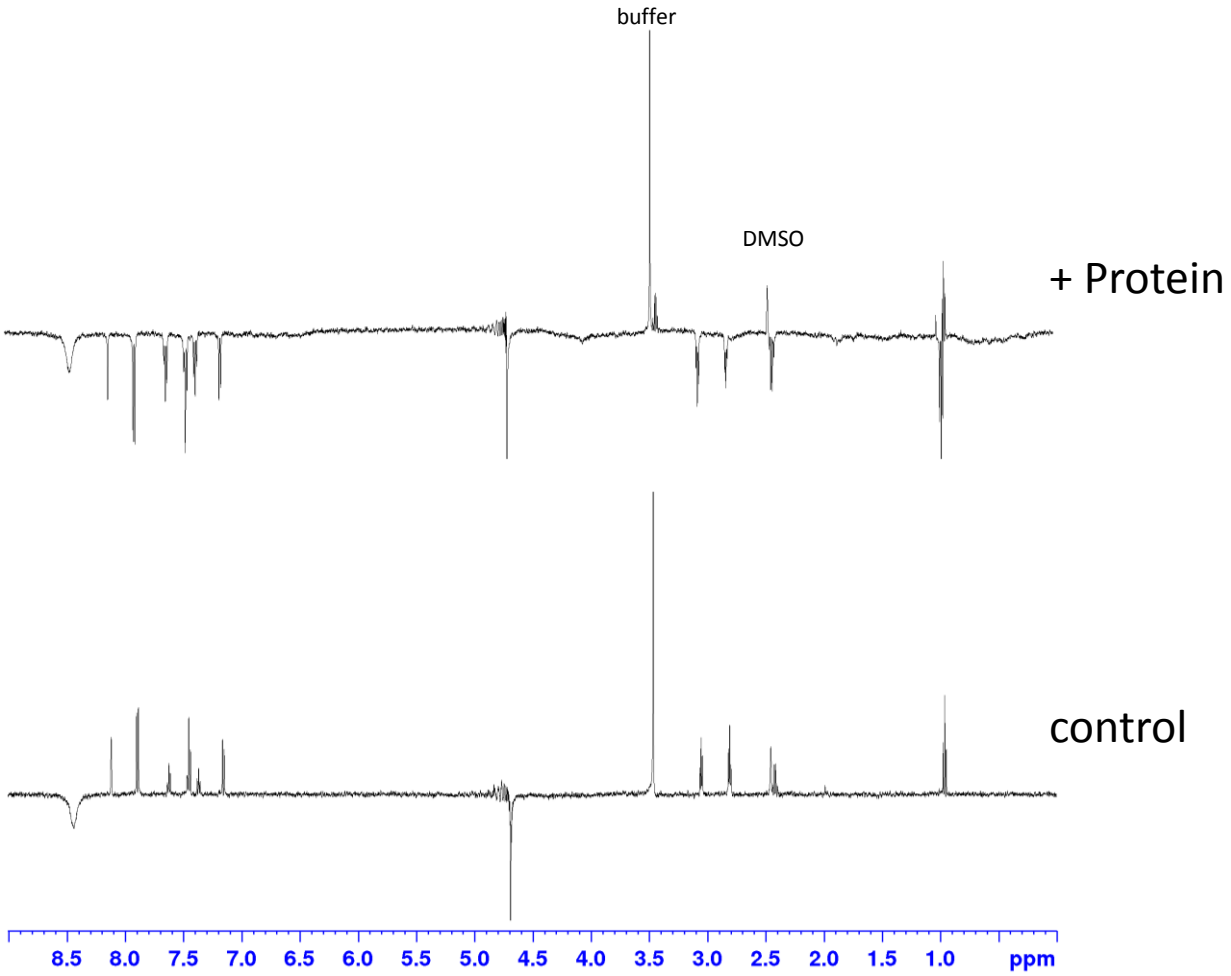
Water suppression: 2 mM sucrose

Excitation Sculpting and perfect-echo ES



Applications

Ligand screening via WATERLOGSY



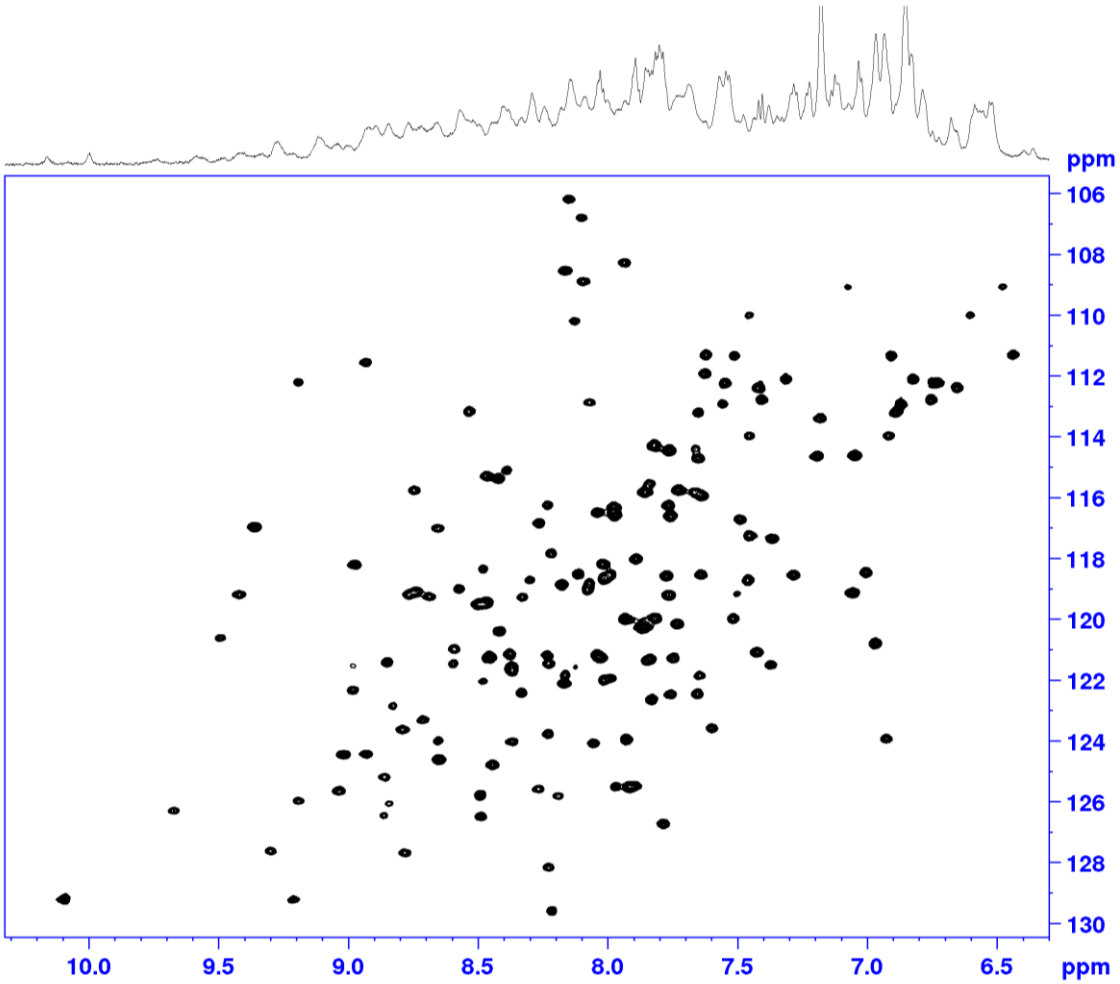
90:10 H_2O:D_2O
10 µM protein
500 µM ligand
283K

1s mixing
256 transients
7.5ms Gaussian Inversion

Automation

Applications

Protein characterisation ^{15}N HSQC



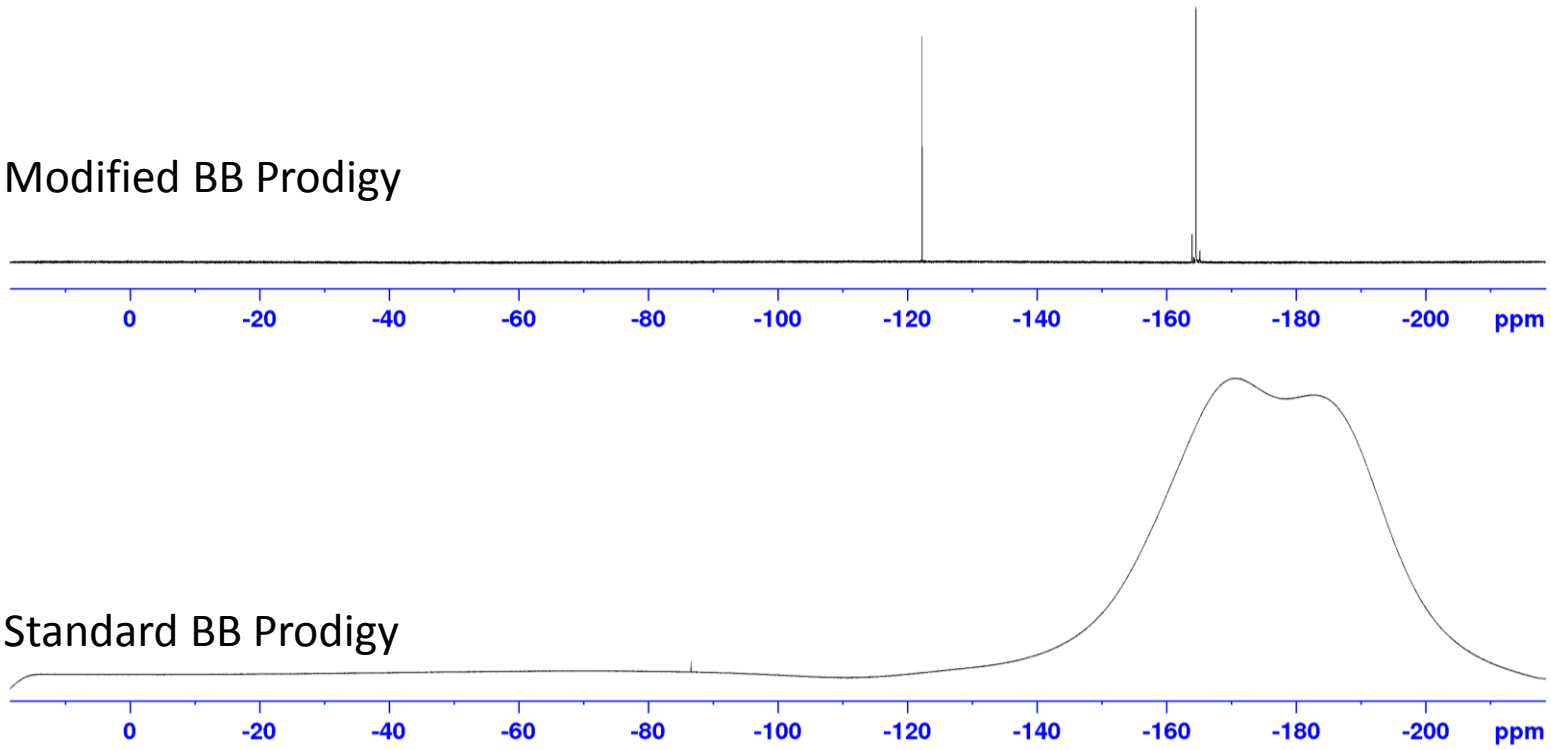
90:10 H_2O:D_2O
600 µM protein
330 µl, 5 mm Shigemi

NS=1
5 mins

(F_1-F_3 experiment using "logical" rf channel)

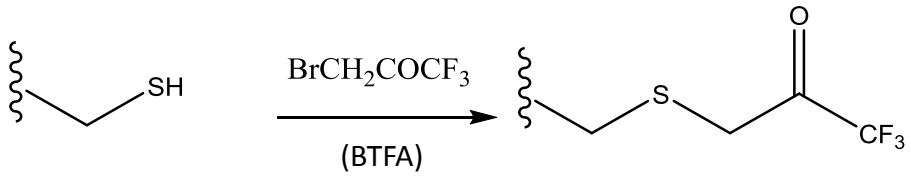
Applications

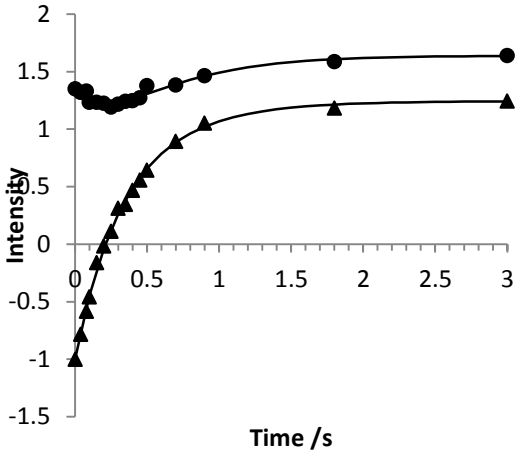
^{19}F NMR capability: probe background

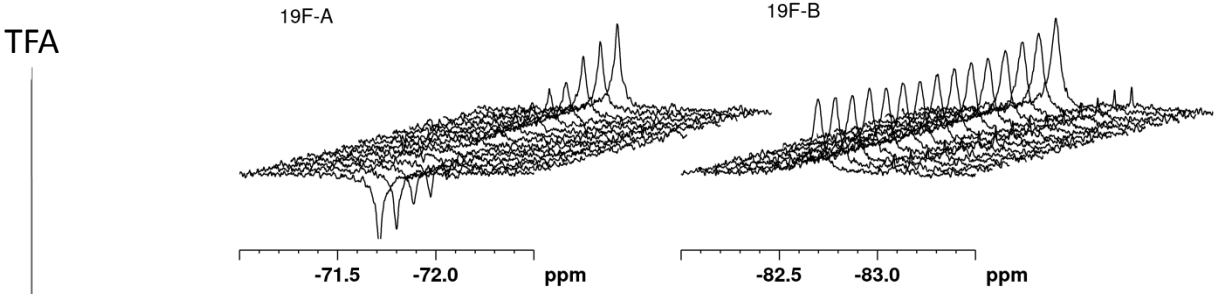


Applications

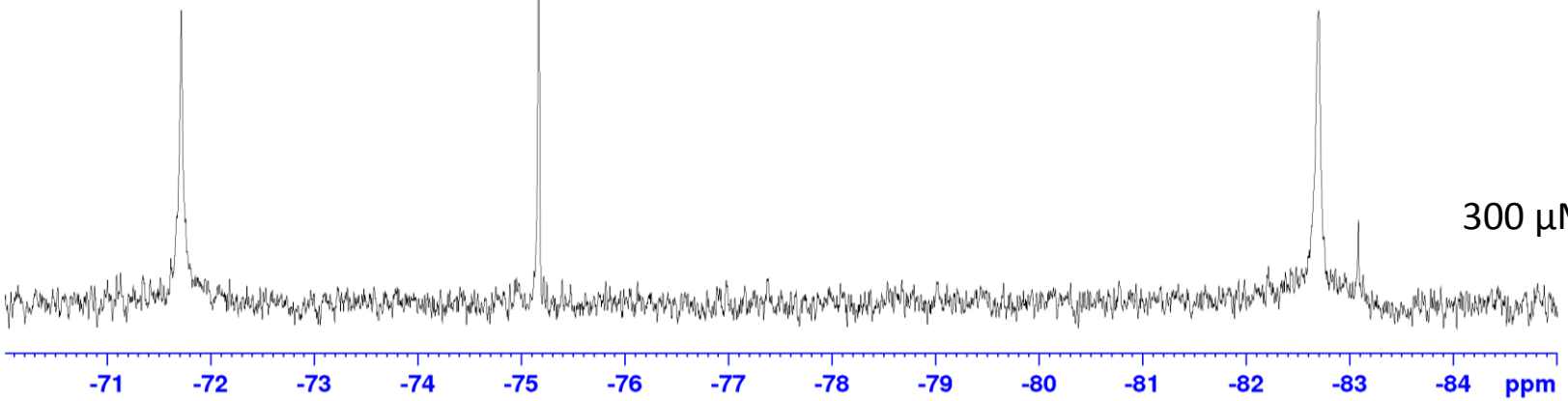
^{19}F labelled protein: Y-C mutant







300 µM protein

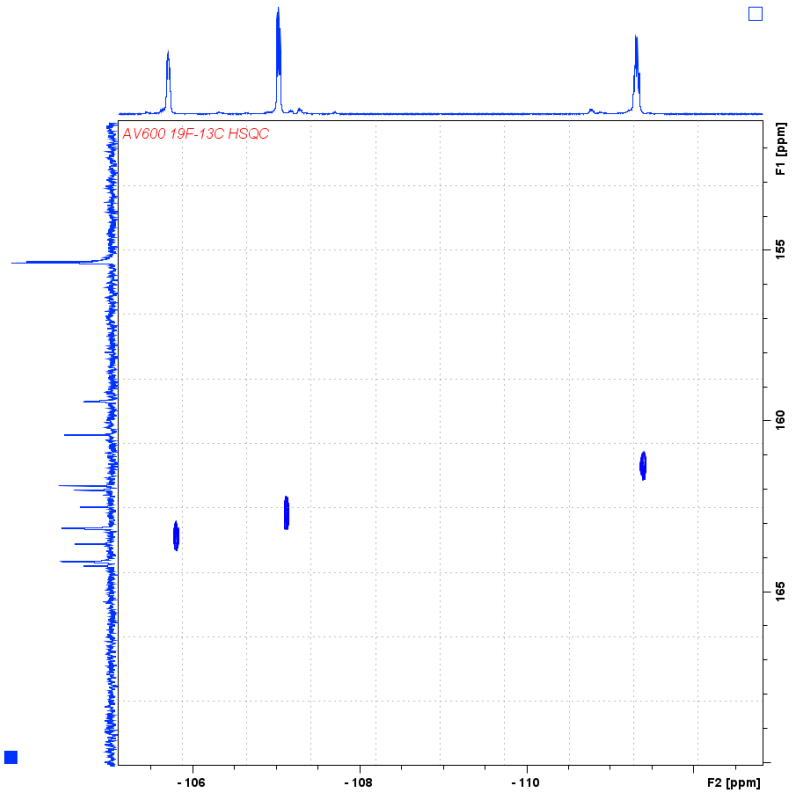


Applications

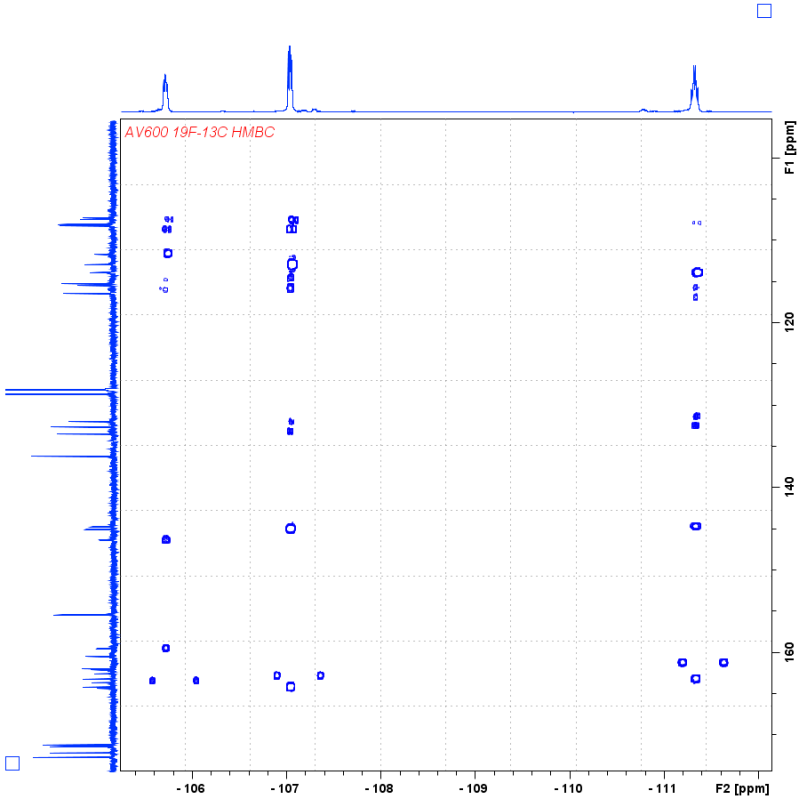
^{19}F-^{13}C NMR

fluorinated peptide

HSQC

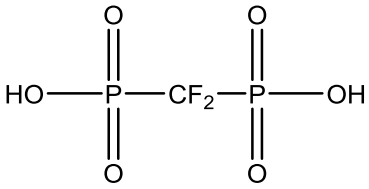


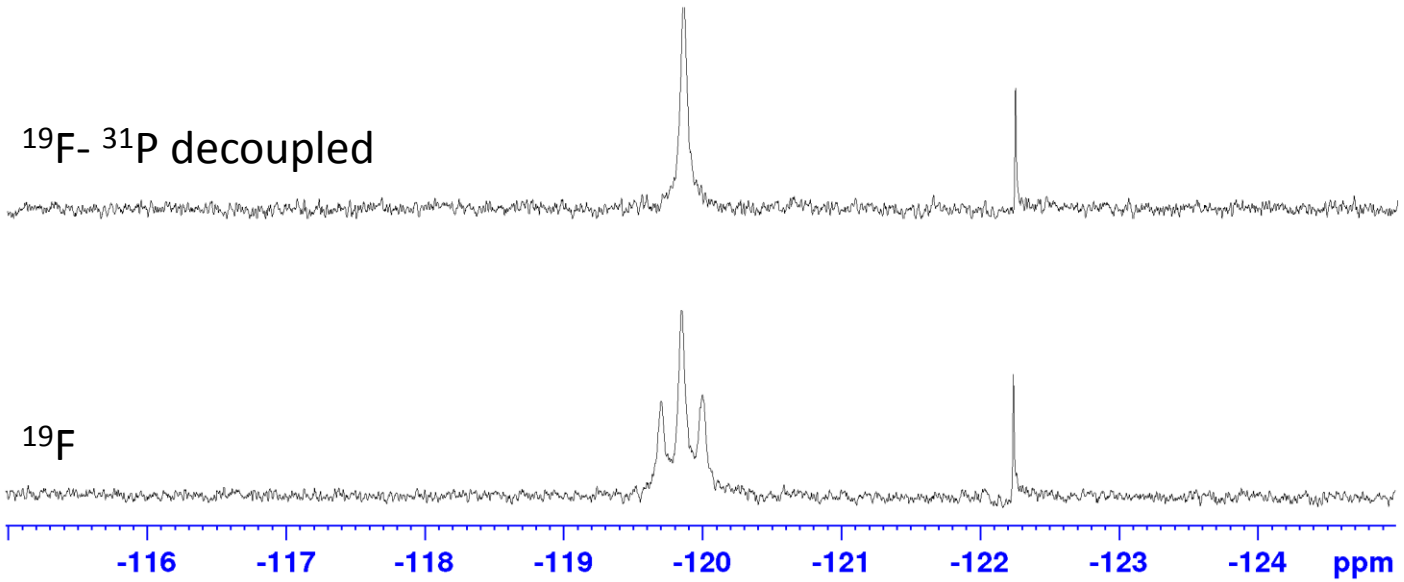
HMBC



Applications

^{19}F-^{31}P NMR



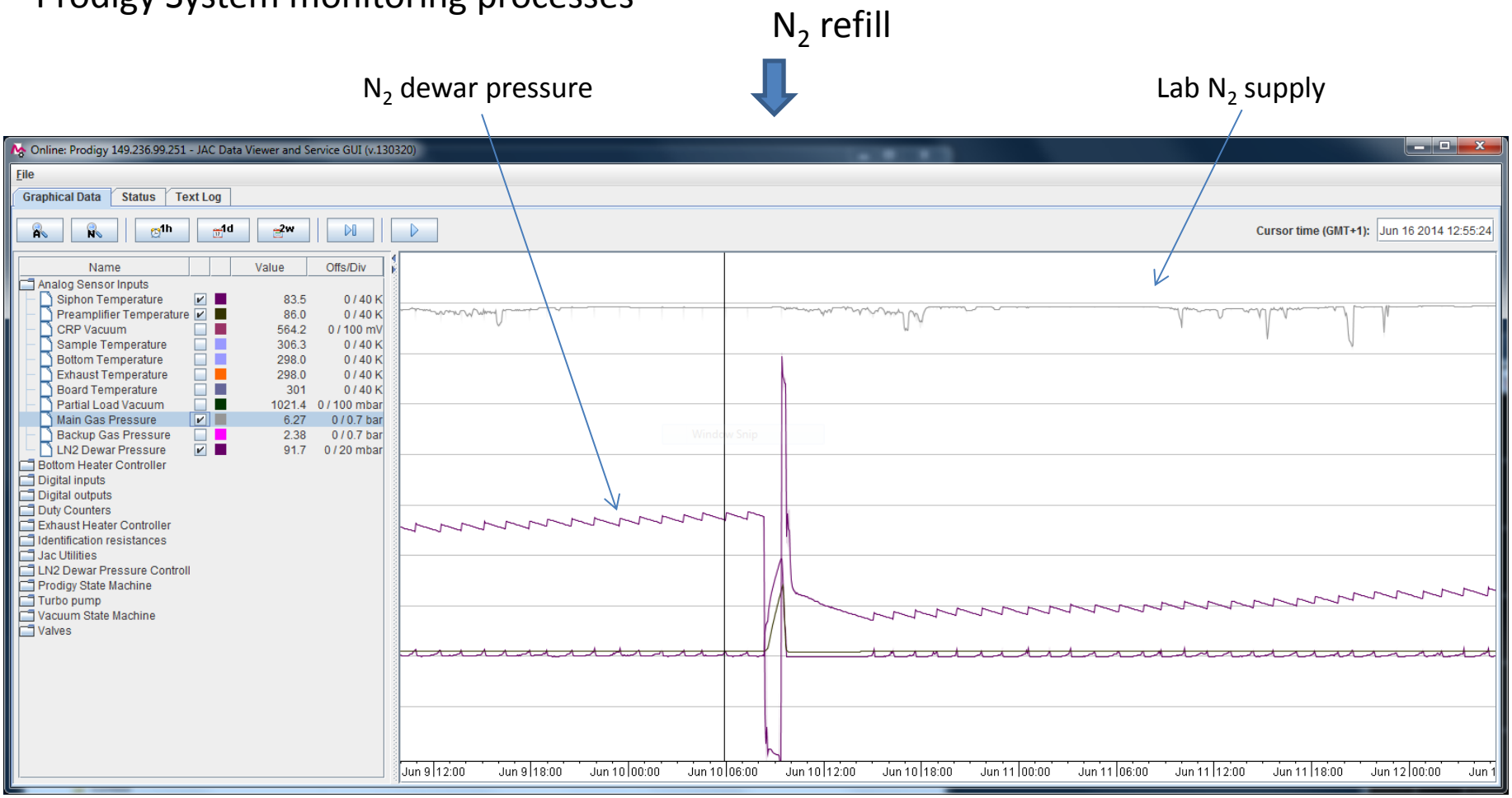


Operational Aspects

- Siting & installation requirements
  - Straightforward wrt He probes
  - Standard 16A power supplies
  - N_2 gas supply (Lab @ 6 bar) + N_2 backup cylinder
- Cryogen filling
  - Magnet and Prodigy dewar every week, simultaneously
  - Time required: ~ 1 hr
- N_2 supply gas reliability is critical
  - In-built monitoring protocols very good
  - *Probe warmed up twice due to supply pressure failure*
- Issues with hold time efficiency of N_2 transfer line (vacuum lifetime?)… Had to pump transfer line once….*being monitored…*

Operational Aspects

Prodigy System monitoring processes



Operational Aspects

N_2 Dewar hold times