Analyzing and Correcting Spectrometer Temperature Sensitivity

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Stability and reproducibility of the spectrometer are fundamental to the success of many modern NMR experiments. Variation in room temperature is a particularly important source of instability, in part because it can cause coherent artifacts in NMR spectra. Small changes in room temperature lead to corresponding changes in the phases, amplitudes, and frequencies of NMR signals. These can lead in turn to apparently random spectral artifacts such as t_1-noise in two-dimensional (2D) NMR and to the incomplete cancelation of signals in difference spectra, but also, importantly, to F_1 satellite signals in 2D spectra. These "parallel diagonals" arise from the use of air conditioning, which typically forces room temperature to oscillate within a fixed band. Work to identify, quantify, and suppress sources of temperature sensitivity in a modern 300-MHz spectrometer has led to a greater than 10-fold improvement in the signal-to-artifact ratio. © 2001 Academic Press

INTRODUCTION

The ability of the NMR spectrometer to reproduce faithfully a desired set of experimental conditions is of great importance in many modern NMR experiments. For example, shortcomings in reproducibility or stability can result in the appearance of artifacts in the final spectrum (*1–5*). Irreproducibility between successive transients leads to the familiar phenomenon of t_1-noise in two-dimensional (2D) spectra and to spurious signals in difference spectra. These artifacts arise from unwanted signal modulations from a variety of sources, including variations in radiofrequency (RF) pulse phase and amplitude, variations in field homogeneity, and variations in field/frequency ratio (*1*).

It has been shown (*2*) that much of the instability in a typical modern high field spectrometer is attributable to two systematic sources: variations in room temperature and modulation of the B_0 field by 50 (60) Hz mains (line) interference from the ac electricity supply. The same paper also showed how the latter could successfully be treated by adding a signal of appropriate amplitude and opposite phase to the Z_0 shim power supply, canceling the mains interference, and gave several suggestions

as to how temperature-driven instabilities could be treated. Apart from one paper (*4*) describing an elegant system of environmental monitoring using the spectrometer computer and advocating care in selecting the set point of air conditioning, and an earlier description of "air conditioning F_1 sidebands" (*3*), the effects of temperature instability on spectra have received little attention in the literature. This may be partly due to the difficulties in identifying and isolating the sources of temperature sensitivity, which are addressed here. This paper describes investigations carried out into the temperature sensitivity of a 300-MHz spectrometer and illustrates the improvement in spectral quality that can be obtained by careful regulation of the temperatures of sensitive spectrometer components. The spectrometer used, a Varian Inova 300, is representative of the current state of the art and considerably more stable than many earlier instruments, but nevertheless produces spectra which frequently show the effects of room temperature variation.

Sensitivity to temperature is widely distributed throughout an NMR spectrometer; as will be seen, it even extends to the cabling between units. Piecemeal attempts to reduce temperature sensitivity can easily lead to frustration; for example, reducing the magnitude of the negative temperature coefficient of phase of the preamplifier will actually lead to an increase in the overall temperature coefficient for the complete NMR experiment if this is positive. This paper describes a number of simple strategies that have been used to investigate the temperature sensitivity of a modern spectrometer. It will be shown below that identifying and attacking the root causes of temperature sensitivity of a spectrometer can make the problem much less intractable. All electronic components show some degree of sensitivity to temperature, and the inductors and capacitors of tuned circuits in particular can lead to this sensitivity translating into changes in RF phase. The degree of sensitivity and the ubiquity of these circuit elements in spectrometers are such that seeking components with very low temperature coefficients is not a practical proposition; rather, strategies are required for limiting the damage done by the temperature sensitivity of components. While some of the experimental methods described below (and the extensive use of cardboard!) are more appropriate to a proof of principle than to the manufacture of commercial instruments, it is hoped that the results described here will help inform the design of future spectrometers.

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EFFECTS OF TEMPERATURE VARIATION

It is well known that variation in room temperature is a significant source of instability in NMR spectrometers, causing changes in the phases, amplitudes, frequencies, and lineshapes of NMR signals. One of the most common classes of systematic artifacts, "parallel diagonal" signals seen in homonuclear correlation 2D spectra, normally arises from the use of air conditioning to maintain an acceptable room temperature. Most air conditioning units are simple two-state (on–off) devices, which typically cause room temperatures to oscillate within a narrow band of about 1°C. If the evolution period of a 2D experiment is incremented or decremented linearly, as is the case in virtually all of the 2D experiments performed to date, then F_1 sidebands will be generated at offsets of $(P/D)\mathrm{sw}_1$ Hz from the true F_1 frequency, where P is the period of oscillation, D is the duration of the experiment, and sw_1 is the F_1 spectral width. As the 2D data acquisition progresses, mapping out the true signal modulations as a function of t_1, so the external influences on the NMR signals add an extra modulation as a function of real time. The effect is that the real time modulations appear as modulations as a function of t_1, with their frequencies scaled up according to the ratio of the duration of the experiment to the range of evolution times explored.

It has recently been shown (5) that systematic F_1 artifacts in 2D spectra arising from temperature variations can be suppressed completely by acquiring the t_1 increments in random order. This method of data acquisition converts a coherent modulation of signal phase, frequency, etc., into an apparently random modulation, which, after Fourier transformation of the t_1 dimension, gives rise to slightly increased t_1-noise rather than coherent peaks. While this technique is a very simple way of suppressing potentially misleading artifact peaks from spectra, the increase in t_1-noise (albeit slight in most cases) may on occasions interfere with spectral interpretation. An altogether more satisfactory approach would be to render the spectrometer less sensitive to temperature disturbances in the first place, by establishing the exact source(s) of the temperature sensitivity and then reducing the sensitivity of the affected component(s).

AIR CONDITIONING

The obvious solution to the problem of spectrometer temperature sensitivity is to stabilize the temperature of the laboratory. Indeed, Braun *et al.* (4) have shown that the generation of coherent F_1 artifacts can be avoided by careful choice of the set point and regulation parameters for the air conditioning units. However, there are severe limitations on this strategy, which in essence reduces to disabling the air conditioning by choosing a set point close to the quiescent temperature that the room would adopt in the absence of an air conditioner. The simple air conditioning systems generally used for NMR laboratories are on/off devices, which start cooling if the temperature

rises above some upper limit T_U and stop if it falls below T_S. The hysteresis band $T_U - T_S$ is typically between 0.5 and 3°C.

The effect of such an air conditioner on the time course of temperature in a room depends on the quiescent temperature. If the latter lies above T_U, the air conditioner will cool the room air until the temperature at the sensor reaches T_S. At this point the unit will cease cooling, and room temperature will drift upward until T_U is reached; the cycle then repeats indefinitely. If, however, the quiescent temperature lies within the hysteresis band, the air conditioner will remain passive, and the room temperature will vary in an uncontrolled fashion until transgresses the limit T_U or T_S. Paradoxically, this may well prove far less damaging to the quality of results than the coherent oscillations generated when the quiescent temperature lies outside the hysteresis band, a point well made in Ref. (4). Some air conditioning units also have a heating capability, but again simple on/off control will lead to oscillation if the quiescent laboratory temperature lies outside the hysteresis band. More complex control systems which do not suffer from oscillation are common in devices such as the variable temperature units used to control the probe temperature in NMR experiments, but can be punitively expensive to install and run on a laboratory scale.

ASSESSING SPECTROMETER
TEMPERATURE SENSITIVITY

This paper describes an investigation of the temperature sensitivity of a typical modern 300-MHz spectrometer, a Varian Unity Inova 300. Compared with earlier spectrometers this is highly stable. Improvements in short-term stability, through the use of low-noise electronics, reductions in stray mains (line) magnetic fields, etc., have both made it easier to detect the effects of spectrometer temperature sensitivity and increased their relative importance. The three principal elements of the investigation described are the measurement of signal characteristics such as signal phase and amplitude on appropriate timescales and the monitoring and the manipulation of the temperatures of different components of the spectrometer.

A laboratory environment experiences temperature perturbations on a variety of timescales. Draughts, opening doors, sample manipulation with compressed air, etc., cause fluctuations lasting seconds or minutes, while air conditioning typically causes cycling on a 5- to 15-min scale. Diurnal temperature variation can also affect NMR experiments, and while seasonal variations are slow on the timescale of an NMR experiment they may affect shorter term temperature stability through changes in air conditioner regulation and the temperatures of incoming gases. Most of the critical components in an NMR spectrometer have significant thermal mass, so temperature fluctuations on timescales less than a second or two are smoothed out. Temperature variation within a single measurement of a free induction decay is therefore rarely a problem, so the effects of temperature variation may be followed by monitoring the phases,

amplitudes, frequencies, etc., of successive free induction decays. Such FID-to-FID comparisons have been used extensively in previous investigations of spectrometer stability (2).

A good deal can be learned from purely passive experiments, in which the spectrometer is run under normal operating conditions and signal behavior and the temperatures of specific components are monitored and correlated. This has the virtue that the instrument behaves exactly as it would in a "real" experiment, but because the signal dependence on temperature is multifactorial it can be difficult to isolate the effects of individual components. The alternative is to intervene actively, either stabilizing the temperatures of selected components or deliberately changing them. A simple example of the former would be passive control by the use of a heat exchanger in a large insulated bucket of water as a preconditioner for probe air: this does not regulate the probe temperature, but simply ensures that any temperature variation is very slow. In the experiments described below, this type of approach is used along with temperature jump methods and active regulation. The choice of how to intervene with a particular component is dictated in part by the information sought and partly by the cooling/heating power demands of that component. A unit which has substantial power dissipation will require a large flow of cooling air; to stabilize this with a passive heat exchanger would require an unrealistically large thermal mass (although it is entertaining to consider the relative costs of, say, a 3-kW-capacity air conditioner and a few meters of copper piping embedded in a 1-m cube of concrete).

Signal Measurements

The results described below were obtained using a number of different variants of a simple strategy; the basic template is described here, and the variations are described in Results below. Several hundred free induction decays are measured at regular intervals, providing information on variations in signal phase, amplitude, and frequency with a resolution of a few seconds over several hours. Time averaging allows time resolution to be traded for reduced susceptibility to other sources of signal irreproducibility on the second or shorter timescale, so here experiments typically average 16 transients over 30 s for each free induction decay. Experiments are carried out with a static sample; sample spinning can lead to rapid small fluctuations in signal phase and amplitude through Q modulation. A concentrated solution of chloroform in acetone-d_6, doped with chromium tris-acetylacetonate to shorten T_1, is used, in order to ensure good signal-to-noise ratio. The resulting free induction decays are given a strong exponential weighting, zero-filled to ensure good digitization, and, after Fourier transformation, a constant zero-order phase correction is applied to bring the successive spectra to approximate dispersion mode. Signal phases and/or amplitudes are then determined and plotted as a function of time and examined for evidence of instability; for a Lorentzian line, the deviation from pure dispersion phase $\Delta\theta$ is $(S_+ - S_-)/(S_+ + S_-)$ radians, and the signal amplitude is $(S_+ + S_-)$, where S_+ and S_- are the positive and negative excursions of the signal.

The same logic may be applied to measurements on synthetic signals, allowing the behavior of different parts of the spectrometer to be isolated by leaking continuous low-level RF signals into different stages, bypassing the probe and NMR sample. The decoupler channel of the spectrometer and the decoupler synthesizer are both potential sources of high-purity RF signals. For example, if the decoupler and observe channels of the spectrometer are derived from the same synthesizer output and a continuous signal is leaked directly from the decoupler into the receiver (via a suitable attenuator), the resultant signal at zero frequency is largely unaffected by any synthesizer instability and completely bypasses the probe. Figure 1 shows a simplified block diagram of the principal components of the Unity Inova 300 spectrometer, modified to illustrate the leaking of a signal from the decoupler channel into the preamplifier. The dc output from the receiver may be examined directly in the time domain (for example to reveal low-level mains [line] phase modulation) or weighted and Fourier transformed (with any dc correction disabled) in the same manner as successive free induction decays, to follow slow changes in phase or amplitude. This approach is used below both to examine the behavior of the complete receiver system, using the decoupler channel to generate the leakage signal, and to examine specific parts, for example, by generating a 10.5-MHz signal using the decoupler synthesizer and leaking this directly into the intermediate frequency stage of the receiver. Another useful approach is a difference experiment, in which a given unit is alternately included in, and excluded from, the signal path using a computer-controlled thermally insulated RF switch. The difference between the two signals is recorded as a function of time, isolating the effect of temperature on the chosen unit, and can then be correlated with the recorded temperature of the unit under investigation.

Temperature Monitoring

Locating the exact sources of temperature sensitivity within the spectrometer requires the monitoring of temperatures at many locations simultaneously. Semiconductor temperature sensors (type LM35, RS Components Ltd.) were fitted at various positions within the spectrometer console, probe, preamplifier and associated electronics, the compressed air supplies, and the laboratory. The sensors give an output of approximately $10 \, \text{mV} \, °\text{C}^{-1}$, which was amplified to a level suitable for digitization and low-pass filtered. Output voltages from the amplifiers were digitized using a 16-channel, 12-bit analogue-to-digital converter (ADC) board (Model PC27, Amplicon Liveline Ltd.) installed in a 386 PC. PASCAL software was used to control the sampling and storage of the digitized voltages, and experimental calibration data for the different sensors were used to convert these voltages to temperatures. Temperature and NMR data were combined for analysis purposes using a "C" program on the spectrometer Sun Sparc 4 workstation.
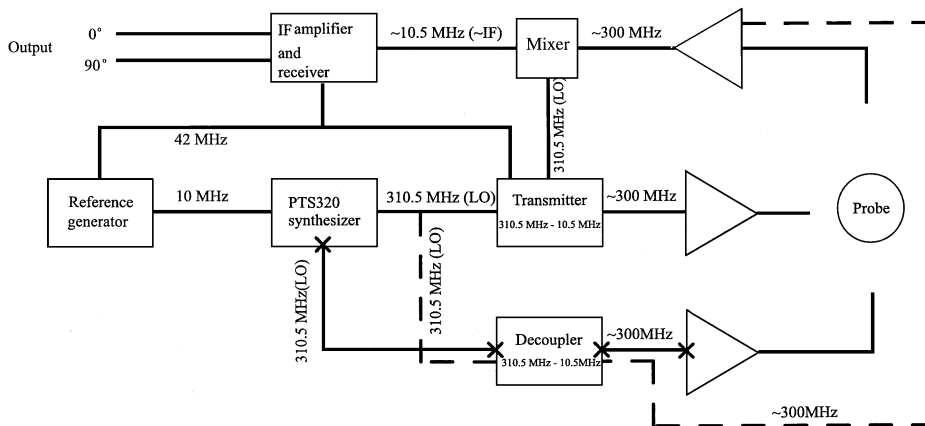
FIG. 1. Simplified block diagram of the architecture of the Varian Unity Inova 300 NMR spectrometer. The dashed lines indicate the signal routing used to leak the decoupler signal into the receiver chain, while the lines with crosses indicate the signal routing for normal decoupler operation.

Temperature Manipulation

It is possible to establish approximate temperature coefficients for many of the components of a spectrometer by selectively altering the temperature of a particular unit while examining the effect on signal over time. If the unit to be assessed is easily removed (e.g., the first preamplifier), perturbing its temperature is relatively straightforward. One crude but very effective method is to place the unit in an insulated container, monitor the signal to establish the quiescent behavior, and then add a hot brick. This gives a rapid rise in temperature, followed by very slow relaxation to room temperature. Selective perturbation of other units can be effected by altering the temperature of the incoming cooling air. In the Inova console it is possible to perturb each of the RF circuit boards in turn, by inserting insulating material such as cardboard between the boards and directing hot air past an individual board through the normal ventilation pathway. Depending on the power dissipation, and hence the cooling air requirements, of a given unit, the temperature of the air supply used may be controlled either actively or passively (see below).

Analysis of Experimental Results

The temperature-sensitive components of a spectrometer are diverse and often difficult of access. It is not always possible to monitor the temperature at the critical component, which in general will lag behind the temperature measured at the outside of the relevant unit. Consider a unit dissipating a constant power P watts through a thermal resistance k W $°C^{-1}$ to an external environment of uniform temperature T_e. If the unit has a heat capacity C J $°C^{-1}$ and is a much better thermal conductor internally than in its interface with the surroundings, it will enjoy a uniform temperature $T_u(0) = T_e + P/k$. If the temperature of the external environment now suddenly increases by ΔT, this step function will cause an exponentially damped growth of T_u toward the new temperature $T_e + \Delta T + P/k$,

$$T_u(t) = T_u(0) + \Delta T\{1 - \exp(-t/\tau)\}, \qquad [1]$$

where the time constant $\tau = C/k$. In general, the temperature $T_u(t)$ for arbitrary $T_e(t)$ will show exponential damping,

$$T_u(t) = T_e \otimes \exp(-t/\tau) + P/k, \qquad [2]$$

where the symbol $\otimes$ denotes convolution. To make the best use of experimental data correlating RF phase or amplitude with the temperature measured at a given unit it is therefore desirable to allow for the effects of temperature lag. Temperature coefficients may be estimated by finding the best fit between the RF phase or amplitude deviation as a function of time and the recorded temperature convoluted by an exponential of variable time constant. Conversely, in some cases the sensitive components may have less thermal mass than the sensors used; here it is necessary to find the best fit between the recorded temperature course and the phase or amplitude deviation convoluted with an exponential. Of course, where sensor and unit show the same thermal lag and where the random error on the temperature is small, simple linear regression is also possible. Examples of all three approaches are given below.

SUPPRESSING SPECTROMETER TEMPERATURE SENSITIVITY

Once the temperature sensitivities of individual units of the spectrometer have been determined, strategies may be devised to minimize the overall sensitivity of the complete instrument. The "gold standard" of ensuring that the spectrometer is maintained at a constant temperature is a popular goal, but ultimately unattainable. Any laboratory is subject to both endogenous (e.g., gated decoupling, variation in RF power) and exogenous (sample introduction, door opening) sources of temperature fluctuation. Improvements in sensitivity and in other aspects of spectrometer stability mean that temperature changes of less than a tenth of a degree have easily measurable effects, so a desirable target for temperature regulation would be about a

hundredth of a degree. This would necessitate stringent room insulation, sophisticated (e.g., proportional integral derivative, PID) control, rapid response, and high peak cooling/heating power and would be expensive in power consumption as well as capital cost.

A more practical goal is to minimize perturbation by careful siting of spectrometer components, air conditioning intakes, sensors and vents, etc., and then to control the temperatures of sensitive components. Here it may suffice to suppress coherent oscillations in temperature, for example by damping the rate of change: a 24-h multidimensional experiment in a laboratory with a temperature oscillating over 1°C every 10 min will show substantial artifacts, whereas one that experiences a slow drift of 1°C over the course of the experiment will show very few ill effects.

A strategy is needed for choosing how to control different spectrometer components. Some have little or no power dissipation (e.g., cable runs) and may simply be thermally insulated from their surroundings. Their sensitivity to external air temperature is governed by the ratio of the thermal conductance of the insulation to the thermal mass of its contents, so it can be worth adding extra material inside the insulation to damp further any fluctuations in temperature. Components that do dissipate significant power cannot safely be isolated by thermal insulation, because this could cause an unacceptable rise in temperature. Cooling air is required, so control is most easily established by controlling the temperature of the incoming air and ensuring that the component in question is only in thermal contact with its surroundings through this airflow. Units with low power dissipation require only modest air flow rates; here the most economic method of control is passive, restricting the rate of change of the temperature of the air by passing it through an insulated heat exchanger with a large thermal mass. Fluctuations in the temperature of the incoming air are damped in the outgoing air to an extent depending on the ratio of the air flow rate to the thermal mass of the heat exchanger.

Radiofrequency power amplifiers are prime examples of units which combine temperature sensitivity and high power dissipation. Here active temperature regulation of the incoming air is needed. This is more expensive than passive regulation, but can achieve similar stability and is relatively cheap because the heating capacity needed to ensure a constant input air temperature to a given unit or group of units is modest. There is some scope for regulation methods which do not use external heating or cooling, for example, using varying cooling air flow rates to maintain a constant temperature, but achieving good regulation by this method would be difficult.

RESULTS

Analysis of Temperature Sensitivity

The multifactorial nature of spectrometer temperature sensitivity, and the limitations of passive experiments as a tool
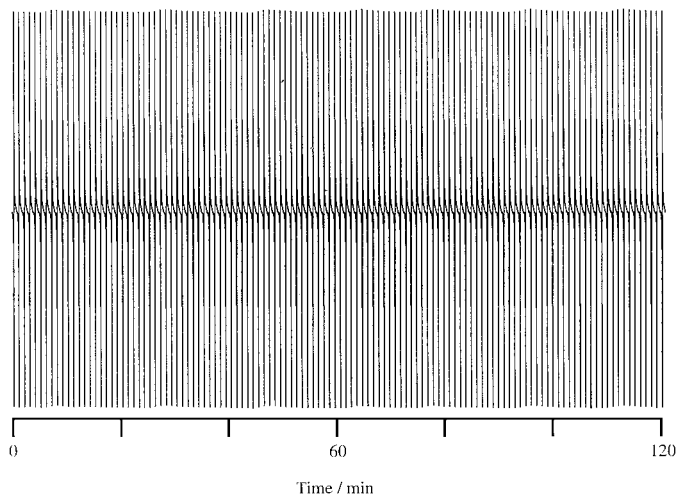

FIG. 2. Dispersion-mode 300-MHz proton spectra of a sample of doped chloroform from an experiment acquiring 120 free induction decays averaging 16 transients of 4096 complex points, recorded at intervals of 30 s. The spectra were processed with 5 Hz of Lorentzian line broadening and zero filling to 8192 complex points.

for investigating it, are illustrated by the results of an experiment measuring phase and amplitude deviations of the NMR signal while monitoring the temperature at different sites in the spectrometer. Figure 2 shows the results of a sequence of 120 identical experiments performed on a solution of chloroform in acetone-d_6 (ca. 10% v/v), doped with chromium tris-acetylacetonate to shorten T_1. Each spectrum was acquired using a 90° pulse of 13 μs and was the average of 16 transients of 4096 complex points, recorded with a spectral width of 1200 Hz and an interpulse delay of 1.88 s, giving a time resolution of 30 s per spectrum. Lorentzian line broadening of 5 Hz was used; this dominates the instrumental and natural linewidths, ensuring that the net lineshape is very close to Lorentzian (and also limiting the timescale over which phase changes during the acquisition of a single free induction decay influence the apparent signal phase).

Figure 3 shows the relative phases and amplitudes of the spectra of Fig. 2, together with temperatures recorded in the laboratory and close to or inside the spectrometer. The changes in phase and amplitude clearly correlate with the temperature variations, with an overall temperature coefficient of RF phase of roughly +0.5°/°C with respect to the temperature at the spectrometer console. However, the correlation is far from exact: the temperature at the magnet clearly also has an effect, and it is not possible to separate the contributions of the individual console units. The apparent temperature coefficient is itself quite irreproducible: repeating the experiment under slightly different conditions can give significantly different figures.

Figure 3 does allow some useful conclusions to be drawn. The relative temperature sensitivities of RF signal amplitude and phase differ, being of the order of −0.08% amplitude/°C and +0.5° (0.009 radians) phase/°C, respectively. Since the
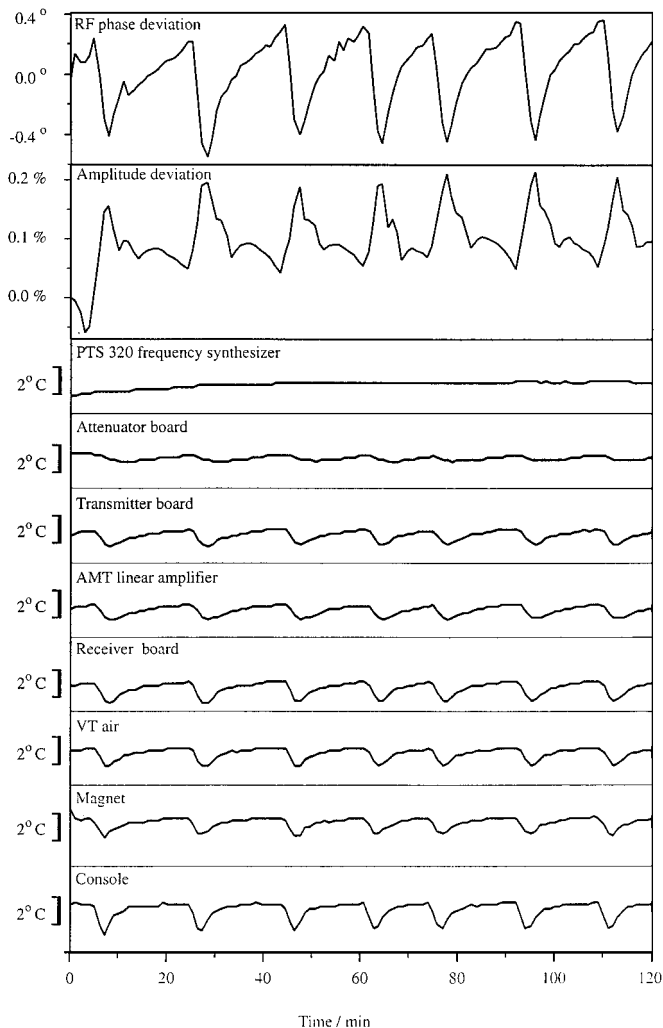
FIG. 3. (Top) RF phase and (second from top) amplitude deviations determined from the spectra of Fig. 2. The bottom portion of the figure shows temperatures recorded at various locations in the spectrometer. Approximate average temperatures for the units were as follows: inside the PTS frequency synthesizer 33°C; on the attenuator board 25°C; on the transmitter board 23°C; inside the AMT linear amplifier 23°C; on the receiver board 22°C; in the variable temperature air supply to the probe 21.5°C; close to the magnet 21°C; and in the main air inlet to the spectrometer console 21°C.

angle. Because this sine dependence makes the effect of pulse flip angle essentially immune to small variations in RF pulse amplitude, signal amplitude error is dominated by effects in the receiver chain from probe through preamplifier, receiver, and mixer to the ADC. Experiments with lower pulse flip angles show significantly worse amplitude stability, particularly where the use of very brief pulses emphasises the role of switching transients.

Figure 3 demonstrates the importance of separating the contributions of individual units to the overall variation of signal phase and amplitude. One way to narrow down the range of units involved is to use synthetic signals to bypass the probe, as indicated in Fig. 1. Figure 4a shows phases calculated from 600 identical experiments in which a synthetic signal from the decoupler channel was leaked into the receiver through a fixed attenuator, and Fig. 4b shows the temperature recorded near the spectrometer console. Each measurement used a single transient of 4096 complex points and a spectral window of 3000 Hz, with a time resolution of 6 s per sample. The spectra were obtained by Fourier transforming the time-domain data without dc
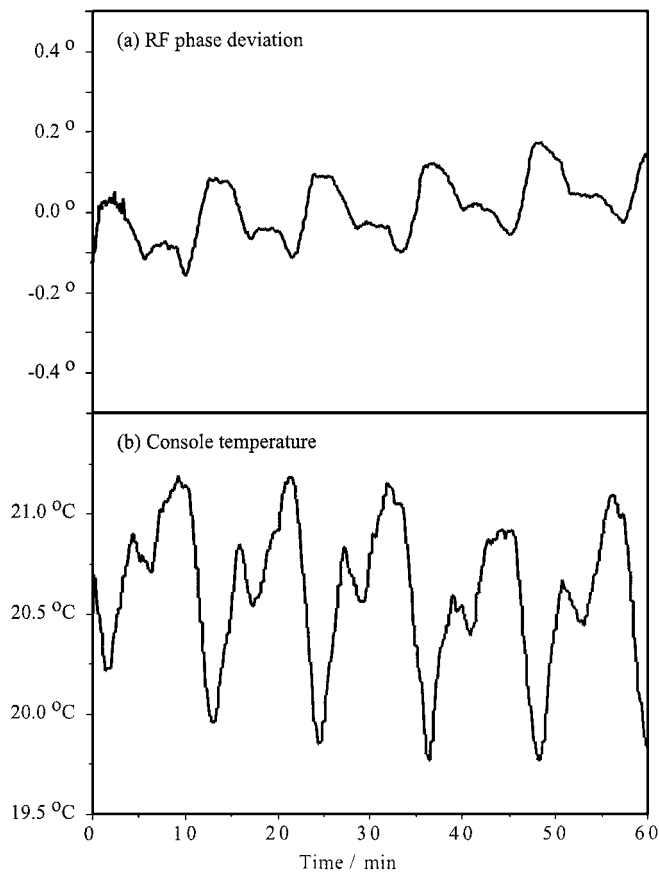

FIG. 4. (a) Deviation in the phase of a RF signal leaked from the decoupler into the receiver chain of the spectrometer, as described in the text and illustrated in Fig. 1; and (b) temperature recorded at the main air inlet into the spectrometer console. A total of 600 signal acquisitions of 4096 complex points with a spectral width of 3 kHz were made at 6-s intervals.

amplitude of the error signal introduced by taking the difference between two signals of unit amplitude differing in phase by a small angle $\Delta\theta$ radians is of order $\Delta\theta$, and the error amplitude introduced by taking the difference between two signals of identical phase and amplitudes 1 and $1-\alpha$ is just α, it can be seen that in this experiment the effect of temperature on phase is about 10 times (0.9/0.08) more important than that on temperature. For this reason the remainder of the results reported here concentrate on the RF phase instability.

Note, however, that the experiment of Figs. 2 and 3 used 90° pulses. This was deliberate: most NMR experiments give signals proportional to simple powers of the sine of the flip

correction, using 2 Hz of Lorentzian line broadening. As well as bypassing the most sensitive part of the spectrometer, the probe, the use of a synthetic signal reduces the random error on the phases measured, since the signal-to-noise ratio of the synthetic signal is much higher than that of the NMR signal.

In contrast with the data shown in Fig. 3 for the entire spectrometer, the results for the decoupler channel plus receiver chain show a temperature coefficient of approximately $-0.2°/°C$, with evidence for some damping by long thermal time constants. The overall temperature coefficient for the NMR signal phase of about $+0.5°/°C$ must thus contain a positive contribution of about $+0.7°/°C$ from the probe and its associated electronics. This helps explain the irreproducibility of the overall NMR phase temperature coefficient, since small changes in the flow rate of air through the probe will significantly affect its temperature sensitivity. Indeed, at (inconveniently) low probe air flow rates it is possible to come close to a zero net temperature coefficient.

Figure 5 shows the effect on the phase of the leaked signal of applying a brief selective perturbation of approximately $13°C$ to the air supply of the receiver board, together with the result of manually fitting the phase to the temperature recorded at the receiver board using the program Mathematica (6), and the difference between the two. The large selective perturbation effectively avoids interference from room temperature variation over the 30-min timescale of the experiment. The fitting yields a temperature coefficient of $+0.042°/°C$; no convolution of temperature or phase was required, showing that the thermal time constants of the sensor and sensitive components are similar. For comparison, Fig. 6 shows the result of linear regression of the RF phase against recorded temperature; the same temperature
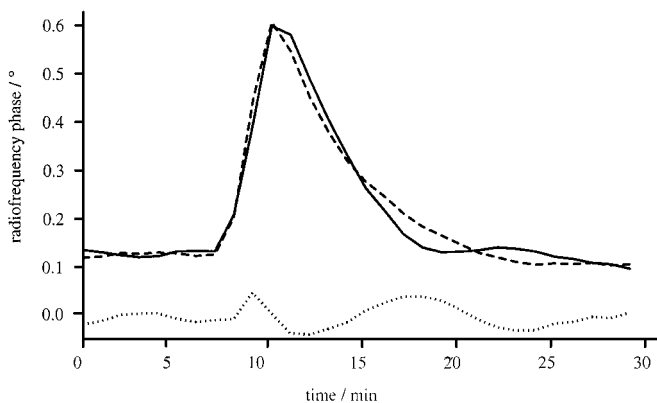

FIG. 6. Scatter plot of the experimental phase changes and temperatures recorded in the experiment of Fig. 5. Linear regression (solid line) gave a temperature coefficient of $+0.042°/°C$, in agreement with the conclusions of the fitting process used for Fig. 5.

coefficient of $0.042°/°C$ is obtained. Figures 5 and 6 both show evidence of small systematic deviations from a simple linear relationship; small timing discrepancies, differences in thermal time constants, and breakdown of the single time constant model may all play a part here, but have little effect on the observed temperature coefficient. Similar experiments were performed for other units, including the transmitter board, AMT power amplifier, and preamplifier.

In contrast to the receiver, the transmitter board shows evidence of significant thermal lag between the temperature at the sensor mounted on the board and that at the component(s) responsible for the temperature sensitivity. The phase of the leaked signal was monitored during a brief temperature jump of approximately $12°C$. In order to fit the phase deviations observed to the temperatures recorded, it was necessary to convolute the latter with an exponential of time constant 2 min. This gave the results shown in Fig. 7, yielding a temperature coefficient of $-0.15°/°C$. Attempts to fit without allowing for the thermal lag gave significantly higher rms errors and obvious systematic deviations and underestimated the temperature coefficient by 30%. Similar experiments to those used for Figs. 5–7 were carried out for other spectrometer components such as the preamplifier and AMT power amplifier.

Temperature monitoring inside the frequency synthesizer (PTS 320) shows only very slow change. Since the synthesizer provides the 310.5-MHz LO ("local oscillator") signal to both transmitter and receiver, the only significant contribution of the synthesizer to signal irreproducibility should be through phase or amplitude variation during a single free induction decay, and the effects of temperature variation should be negligible. Similar arguments apply to the 42-MHz reference generator board (see Fig. 3). No experimental evidence was found of temperature sensitivity on the part of either of these units contributing to the observed signal phase and amplitude variation.

FIG. 5. Time course (solid line) of the change in leaked signal RF phase caused by a brief step of approximately $13°C$ in the temperature of the receiver board of the spectrometer console, together with the time course of the temperature change measured at the surface of the board (dashed line) scaled by a temperature coefficient (determined by manual optimization) of $+0.042°/°C$, and (dotted line) the difference between the phase and the scaled temperature. Phase changes were measured using 120 measurements of 16 averages of 4096 complex points with a spectral width of 2 kHz over 30 s; only 30 min of data are shown. The phase course was fitted with the measured temperatures using the program Mathematica (6), without convolution of the temperature data.
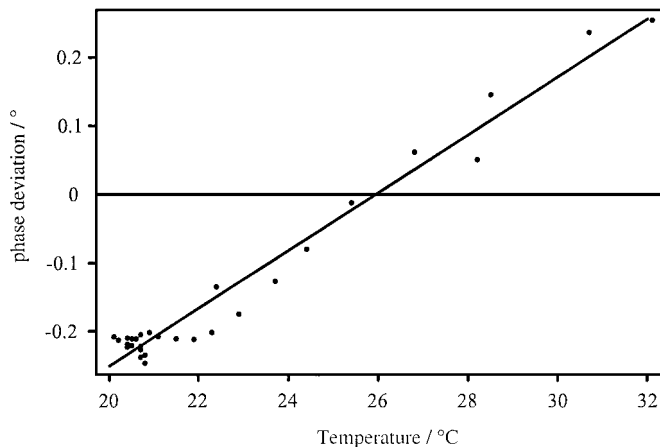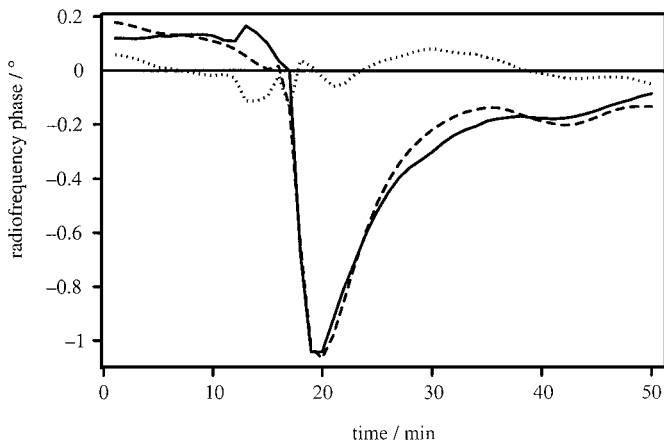
FIG. 7. Time course (solid line) of the change in leaked signal RF phase caused by a brief step of approximately 12°C in the temperature of the transmitter board of the spectrometer console, together with the time course of the temperature change measured at the surface of the board (dashed line) scaled by a temperature coefficient (determined by manual optimization) of $-0.15°/°C$ and convoluted by a decaying exponential of time constant 2 min, with (dotted line) the difference between the phase and the scaled temperature.

At first sight the effect of temperature on the coaxial cable used throughout the spectrometer might be thought unimportant. The linear thermal expansion coefficient of copper is sufficiently low (ca 1.7×10^{-5} °C^{-1}) to suggest that for cable lengths of the order of meters (a few wavelengths at 300 MHz), temperature dependence of phase should be small. For cable with a typical propagation velocity of $0.67 c$, linear expansion of the physical path length would generate a phase shift of approximately $0.004°$ °C^{-1} m^{-1}. However, this simple picture is complicated by thermal expansion of the cross-section of the cable and the temperature dependence of the dielectric medium used, and by their effects on characteristic impedance. Experimental temperature coefficients can in fact be quite significant.

The temperature sensitivity of coaxial cable was investigated by performing a difference experiment in which a synthetic signal was alternately leaked from the decoupler into the receiver directly, and through an additional 15 m of coaxial cable (type RG58C/U, RS Components Ltd.). The cable was warmed slightly above room temperature, fitted with a temperature sensor, and lagged with polythene bubble wrap insulation to ensure slow cooling. The difference in signal phase between the two routes was correlated at 1-min intervals for 55 min with the temperature measured on the surface of the cable. The slow cooling means that damping can be ignored, allowing a simple linear correlation to be sought between phase and temperature. Figure 8 shows the difference in phase between the two signal paths as a function of temperature at the cable surface, together with the result of linear regression. The observed temperature coefficient corresponds to $-0.06°$ °C^{-1} m^{-1}. This figure should only be regarded as indicative, since the effects of temperature-induced changes in the cable characteristics will depend on the source and load impedances attached to the cable, but clearly

shows that cable runs of the lengths found in typical spectrometer configurations can be a significant source of temperature sensitivity. Such effects are partly mitigated by the tendency to run related cables in parallel, so that, for example, the phase change experienced by the transmitter pulse as it travels from the console to the probe is similar to that encountered by the LO signal running from the console to the mixer.

As noted earlier, the probe is one of the most significant sources of temperature sensitivity. This is unsurprising: the probe observe coil is the highest Q LC circuit in the spectrometer and hence the most sensitive to thermal changes in component values. As is well known, careful control of the probe temperature is vital for instrument stability. As mentioned earlier, an effective and inexpensive way of minimizing short-term variations in probe temperature is to pass the probe air supplies through a passive heat exchanger consisting of a length of copper pipe in an insulated bucket of water. Packing the pipe with lead shot increases the efficiency of temperature equilibration, reducing temperature variations in the output air 100-fold and providing a degree of stability well beyond that normally offered by commercial NMR variable temperature controllers. Probes typically use two independent air supplies, one for sample temperature control and the other a supply of buffering air to protect the remainder of the probe contents from extremes of temperature. Where stability is critical it can therefore be desirable either to use two heat exchangers or to run both air supplies from a single exchanger.

Such heat exchangers also provide a way of selectively manipulating the temperatures of the two probe air supplies, simply by changing the temperature of the water. Figure 9 shows a
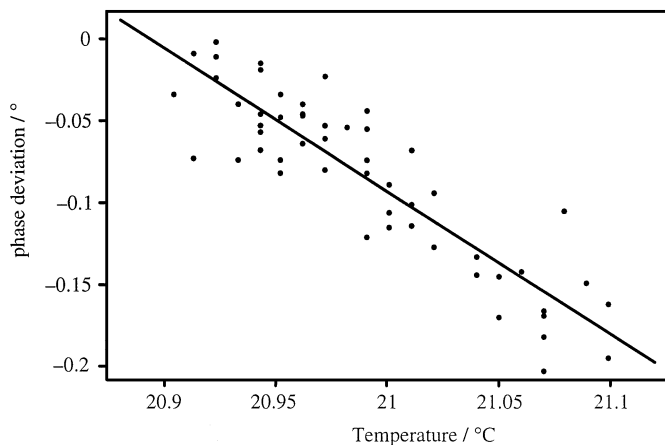

FIG. 8. Scatter plot of the experimental phase changes and temperatures recorded during a change of 0.2°C over 55 min in the temperature of a thermally insulated coil of 15 m of 50-Ω characteristic impedance coaxial cable; linear regression (solid line) gave a temperature coefficient of $-0.9°/°C$. Phases of the leaked signal were measured alternately via and bypassing the coaxial cable, using a double pole, double throw RF coaxial relay switched under spectrometer control, and the difference in the two phases plotted to isolate the contribution of the cable. Phases were measured using 60 pairs of 30-s measurements of 16 transients, each of 512 complex points with a spectral width of 500 Hz.
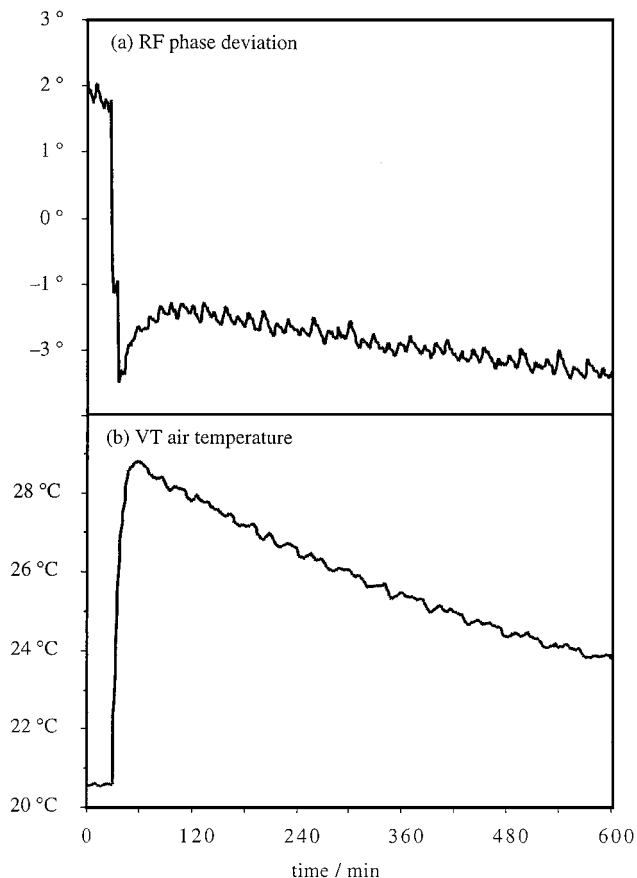
FIG. 9. (a) Change in the phase of the measured NMR signal of a doped chloroform resonance, and (b) temperature of the air supplied to the variable temperature inlet of the probe during a 10-h experiment in which the temperature of the water in the heat exchanger described in the text was rapidly increased by ca. 8°C. A total of 600 spectra, each the average of 32 free induction decays of 1024 complex points with a spectral width of 512 Hz, were measured.

probes often rely on friction fits for components such as coil mounting capillaries and insert tubes, so the phase jumps seen may reflect discontinuous movement of the RF coil. Another obvious possible source of stick–slip behavior is the tuning capacitors, but the fact that phase discontinuities such as those in Fig. 10 are associated with changes in the variable temperature air supply rather than the probe cooling air suggests that the capacitors may not be to blame. Results from similar experiments on the cooling air show continuous behavior with an approximate temperature coefficient of +0.9°/°C. These temperature coefficients for the probe are consistent with the overall spectrometer behavior seen in Figs. 2 to 4, given that in normal operation the cooling and variable temperature air supplies do not experience the full swing in room temperature. The results confirm that while the spectrometer console is a significant contributor to temperature sensitivity, the single most important element is, unsurprisingly, the probe.

The temperature coefficients found for representative sections of the spectrometer are summarized in Table 1. The figures are indicative only; no error estimates are given, because of the systematic bias introduced by the arbitrary choice of temperature monitoring points on the various units. In many cases the statistics of the fitting processes were very good, as evidenced, for example, by the data in Figs. 5 and 6, but a different estimated temperature coefficient with an equally low standard error would
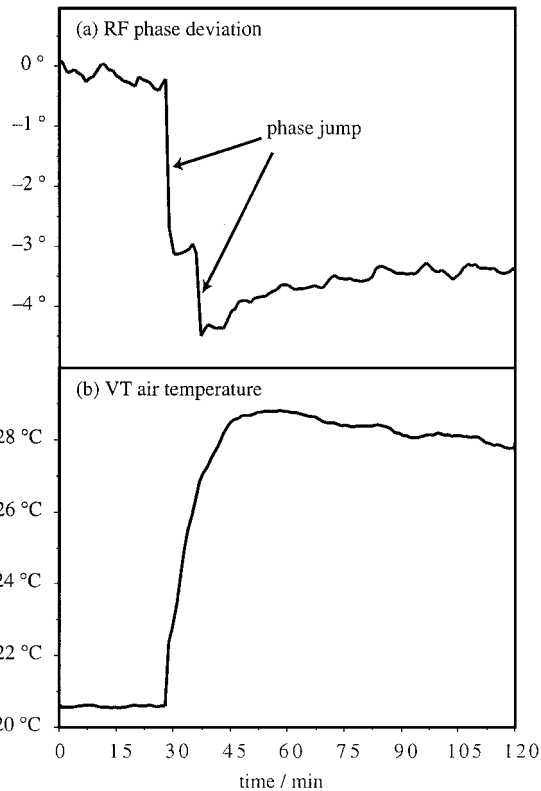
comparison between signal phase deviation and temperature for 1200 identical measurements over 10 h of the chloroform proton resonance, using temperatures recorded with a sensor in the airflow to the probe variable temperature air inlet. The water temperature was raised by approximately 8°C shortly after the start of the experiment by adding hot water to the heat exchanger and mixing rapidly. Leaving aside the dominant, early, part of the phase course, the latter part of the results shows a basic temperature coefficient of approximately +0.25°/°C. The earlier part, shown expanded in Fig. 10, highlights the existence of another, intermittent source of variation in NMR signal phase, which is responsible for much larger phase errors than the basic temperature coefficient.

Superimposed on the slow systematic change in phase with temperature are two steep negative steps in the signal phase. These are irreproducible, are only occasionally seen in experiments that do not involve rapid temperature change, and are probably the result of "stick–slip" differential expansion or contraction of probe components. Coil support systems in modern

FIG. 10. Expansion of the first 2 h of Fig. 9, showing the discontinuous change in phase caused by the temperature step.

TABLE 1
Estimated Temperature Coefficients of RF Phase for Selected Components of the Inova 300 Spectrometer

Unit	Temperature coefficient/°/°C
Probe (cooling air supply)	+0.9
Probe (variable temperature air supply)	+0.25
Preamplifier	+0.02
Mixer	0
Receiver	+0.04
Synthesizer	0
Reference generator	0
Attenuator	0
Transmitter	−0.15
Power amplifier	−0.1
Coaxial cable	$-0.06\,\mathrm{m}^{-1}$

have been obtained had the sensor been positioned differently. The significance of the results in Table 1 lies rather in the pattern they show of distributed sensitivity to temperature across the spectrometer.
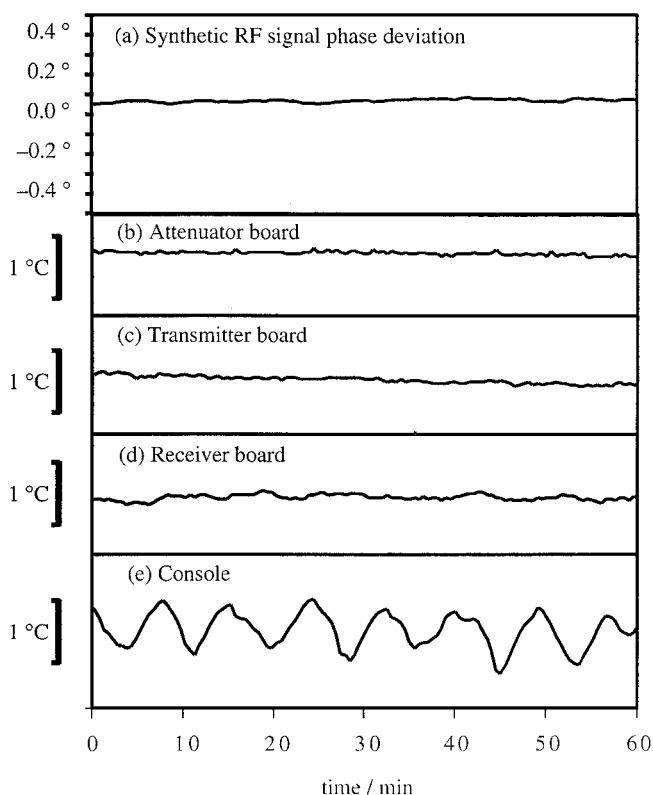

FIG. 11. (a) Variation in phase over a 60-min period of a signal leaked from the decoupler into the receiver channel, with (b) to (e) the temperatures measured on the attenuator, transmitter, and receiver boards and in the main air inlet to the spectrometer console. The temperatures of all sensitive components of the spectrometer except the probe (which was bypassed by the signal path) were stabilized as described in the text. A total of 120 phase measurements of 16 transients were made at 30-s intervals; each measurement used 512 complex points with a spectral width of 500 Hz.
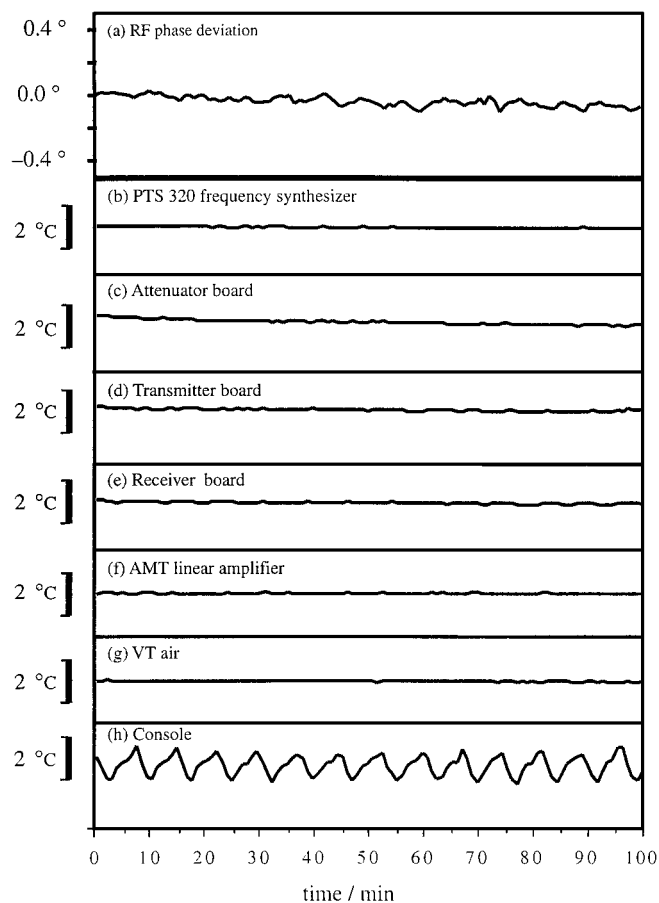

FIG. 12. (a) Variation in the phase of a doped chloroform NMR signal and (b) to (h) temperature changes measured at the frequency synthesizer, attenuator board, transmitter board, receiver board, power amplifier, probe variable temperature air inlet, and in the main console air supply. The temperatures of the sensitive components of the spectrometer were stabilized as for Fig. 11, and in addition the air supplies to the variable temperature and cooling air inlets of the probe were held at constant temperature using high thermal mass heat exchangers. The NMR signal phase was followed using 100 measurements of 32 transients in 60 s, each of 4096 complex points with a spectral width of 1200 Hz.

Suppression of Temperature Sensitivity

As the results above show, the overall temperature coefficient seen in Fig. 3 is the sum of temperature coefficients of differing signs from the key stages in the transmitter and receiver chains. With the exception of the probe, which is frequently supplied with stabilized air, attempting to regulate the temperature of any individual spectrometer component in isolation is unlikely to effect a significant improvement and may indeed degrade overall performance. Effective treatment of temperature sensitivity requires a concerted attack on all known sources of instability, although the approaches taken to individual units will differ.

The cooling demands of the spectrometer make direct temperature regulation of the entire console impractical, since typical console dissipation is several kilowatts. The desired stability can, however, be obtained by targeted intervention, using different
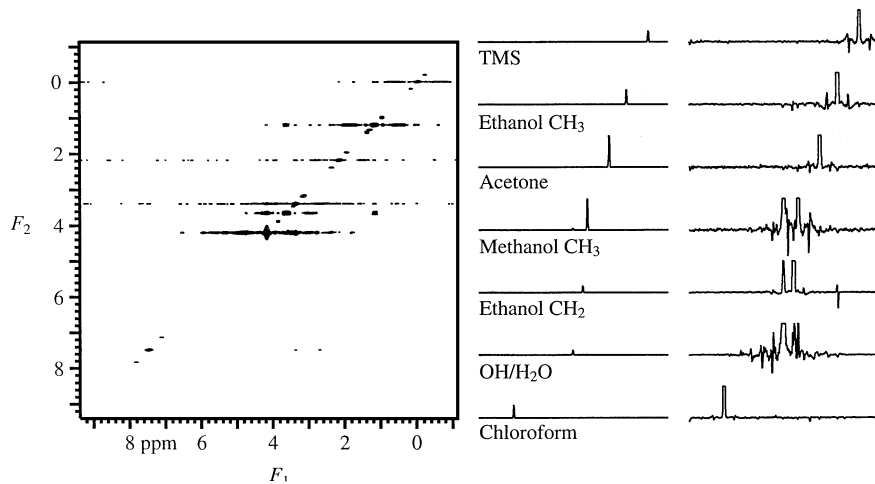
FIG. 13. Phase-sensitive NOESY spectrum of a mixture of ethanol, methanol, acetone, chloroform, and TMS in deuteriochloroform, acquired on a Varian Unity Inova 300 spectrometer. A total of 256 increments of four transients of 4 K data points were acquired over a period of 4 h. The data were processed using Gaussian weighting in each dimension and zero-filled once in both t_1 and t_2 before Fourier transformation. The standard instrument configuration was used, with neither probe nor console temperature regulated.

methods according to the power dissipation needs of different units. The air supplies to the probe were stabilized to within a few hundredths of a degree using the passive heat exchanger system described above. The RF boards of the console typically dissipate only a few watts and can be maintained at acceptable temperatures with only modest airflow. These boards were stabilized using air at about 100 L min^{-1} fed from a heat exchanger similar to that used for the probe air, steered past the boards using cardboard baffles.

The AMT RF amplifier, on the other hand, can dissipate almost a kilowatt and thus demands much more cooling air. These demands can be moderated significantly by regulating only the high-frequency (proton) amplifier temperature, since the power

supply (the largest single source of heat) does not require stabilization, and the low band amplifier is largely used for nuclei with sensitivity too poor for phase and amplitude errors of the magnitudes described here to be significant. The proton amplifier temperature was stabilized by constructing a thermostatted air supply, again fed to the relevant part using cardboard baffles. The air temperature was regulated at about 2°C above room temperature using a PID controller (Model TDH02 FFRR00, Thermospeed Ltd.), fed by a 10 mV/°C sensor of the type described earlier, to control an air heater (Model 308-215, Thermospeed Ltd.) designed for 120 V/750 W operation but operated at 12 V dc. This results in a narrow range of operating temperature, because of the limited heating power available, but this is
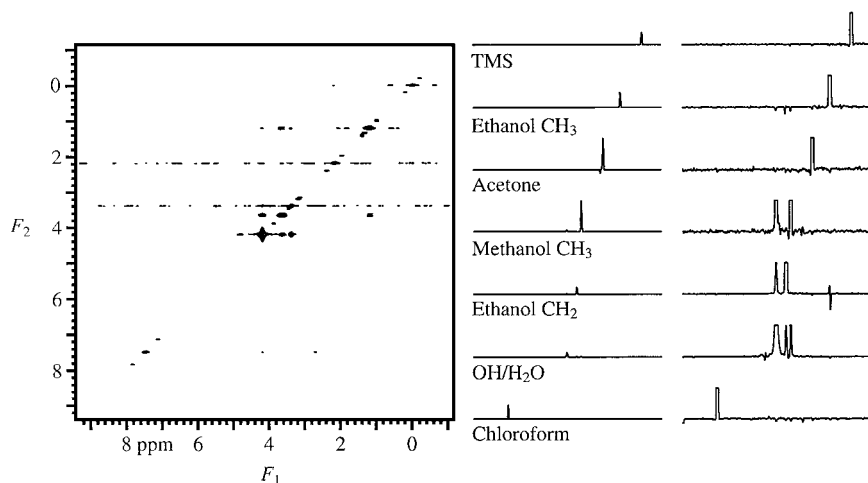

FIG. 14. Phase-sensitive NOESY spectrum of the same sample as that in Fig. 13, acquired with the temperature of the air supplies to the probe stabilized using the passive heat exchanger described in the text.
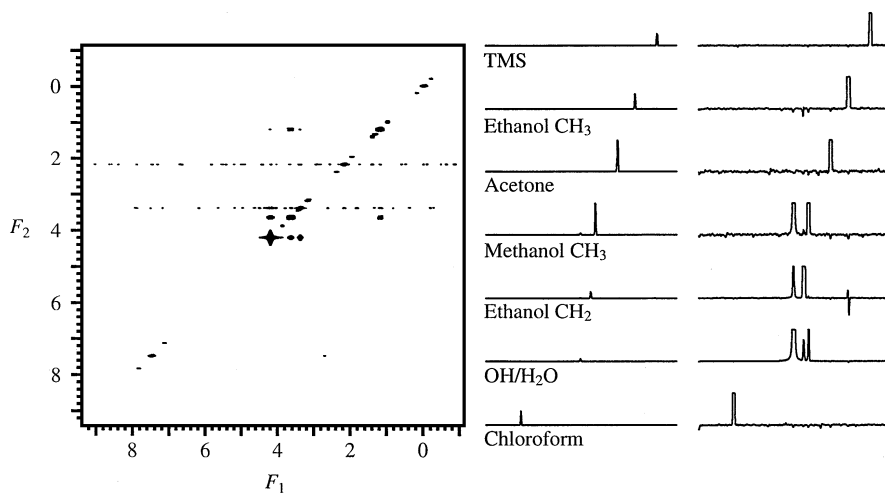
FIG. 15. Phase-sensitive NOESY spectrum of the same sample as that in Fig. 13, acquired with both probe air supply stabilized and the temperatures of specific units within the spectrometer console regulated as for Figs. 11 and 12.

not a problem in a laboratory kept within a narrow temperature range by air conditioning. The performance obtained was excellent, with temperature regulation approaching one hundredth of a degree. Contributions to temperature sensitivity originating from coaxial cables both inside and outside the spectrometer console were minimized by insulating them with polythene bubble wrap.

All critical components of the spectrometer were thus held at fixed temperatures using the standard laboratory compressed air supply, and with only a few tens of watts of added power dissipation. The principal disadvantages of the improvisatory methods used were hindered access to the instrument and a marked degradation in its appearance. Figure 11 shows the time course of RF phase deviation of a synthetic signal and temperature at selected points. The RF boards monitored all show greatly reduced temperature excursions, and the oscillations in leaked signal phase caused by the air conditioning have been reduced to about $0.01°$ peak-to-peak, an improvement of a factor of 20.

Experiments using synthetic signals are powerful diagnostic tools, but the critical test is the reproducibility of real NMR signals. Figure 12 shows the results of a series of measurements of the phase of a chloroform proton NMR signal similar to the series used for Figs. 2 and 3, but with the probe temperature stabilized using the passive heat exchanger described and the console electronics stabilized as for Fig. 11. Once again a very substantial improvement is obtained, the peak-to-peak NMR signal phase excursion being about $0.03°$, again about a 20-fold improvement.

The practical implications of the instrumental modifications described are illustrated by a series of NOESY spectra of a mixture of TMS, ethanol, acetone, methanol, and chloroform in deuteriochloroform. This sample was chosen for a number of reasons. The sharp singlet signals show few, if any, cross peaks, allowing unequivocal identification of artifactual signals, while the ethanol OH signal shows a strongly temperature-dependent chemical shift and hence is a sensitive indicator of probe temperature regulation. The spectrum of Fig. 13 was recorded with

the spectrometer in its normal configuration, with no regulation either of the console or of the probe temperature. The effects of room temperature oscillations caused by air conditioning are clearly visible as a series of satellite signals in F_1 either side of the $F_1 = F_2$ diagonal. A pure sinusoidal variation in room temperature would give rise to small index phase modulation and cause two satellite signals equally spaced either side of the diagonal; the more complex patterns seen reflect deviations from simple sinusoidal behavior.

Regulating the probe temperature reduces the artifacts substantially, as can be seen in Fig. 14. In particular, the averaged signal from the water and OH protons can now be seen to give rise to two cross peaks, attributable to NOEs between the ethanol OH and methylene resonances and the methanol OH and methyl, although weak sidebands remain visible. Regulating the entire spectrometer, as for Fig. 12, yields a further improvement. In Fig. 15 the effects of room temperature oscillation have finally been reduced to the threshold of visibility for this particular spectrometer and experiment.

CONCLUSIONS

The experiments described here serve to characterize the temperature sensitivity of a typical modern NMR spectrometer and demonstrate the practicality of adapting spectrometer designs to reduce vulnerability to temperature-induced instability. While the temperature coefficients reported here are specific to this instrument and test configuration, both the generic methods used and the general pattern of temperature sensitivity are likely to be applicable to most modern NMR instruments. In particular, the distribution of sources of temperature sensitivity throughout the spectrometer accounts for the relative failure of earlier attempts to compensate for phase and amplitude variations by applying temperature-controlled phase and amplitude corrections to the incoming NMR signal (7). The degree of improvement

in instrument stability obtained here with relatively modest (if unesthetic) means is gratifying and suggests that a more professional approach would pay dividends. It is perhaps useful to reflect that the best results reported here only succeed in reducing temperature-induced phase and amplitude modulation artifacts to the level of the t_1-noise, itself frequently a significant limiting factor in multidimensional NMR. Recent advances in the sensitivity of spectrometers, and the considerable further improvements to be expected from cooled probe coil techniques, will place a further premium on the correction of artifacts such as those described here.

There is a further class of artifacts caused by room temperature oscillations which has not been considered here. As well as phase and amplitude variation, and the temperature dependence of chemical shift noted in Fig. 12, some spectrometers show a further source of unwanted modulation in a temperature dependence of the instrumental linewidth. No significant effects of this sort were observed on the spectrometer under study, which typically showed only a few hundredths of a hertz linewidth change over intervals of an hour or more, but on at least one instrument linewidth variations with room temperature of more than 0.1 Hz/°C have been seen. These were specific to the particular magnet involved (subsequently replaced). They may have reflected a relatively poor adjustment of the Z^2 superconducting shim and/or a defect in the magnet suspension system within the magnet dewar vessel and were thought to arise from thermally induced movement of the magnet solenoid with respect to the probe. A second source of similar problems, not responsible in this particular case, is the presence of large ferrous metal objects, for example, girders, roof beams, or steel reinforcing rods, within the magnet fringing field.

ACKNOWLEDGMENTS

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