

Pre processing techniques as an important part in analysis of ¹H NMR data.

Franciszek Binczyk, *Politechnika Śląska*
(dr hab. inż. Joanna Polanska, Prof.nzw.Pol. Sl.)

Abstract

During last few years NMR spectroscopy becomes very popular technique in tumor diagnosis process. Thanks to spectroscopy we are able to determine metabolism of tested area and decide about kind of tumor and its range. Analyzed signal has a form of frequency domain spectrum. To determine chemical composition, area under peaks should be obtained. It appears to be rather simple procedure. Nothing could be further from the truth. Unfortunately raw signal (taken from spectrometer) contain many artifacts (e.g. noise) that should be removed during data pre processing. One of such a distortion is called phase error and follows from methodology that saves 3D precession signal in a form of two 2D projections. There exist a lot of methods to deal with the problem that differs in complicity and possibility to use. They may be divided into two separate groups. First group consist of manual methods that requires human decision. Second are methods based on assumed quality/stop criterion. Three most popular: Entropy minimization, eDispa and Automics are briefly described and explained. All mentioned are examined with usage of artificially generated data set and exemplary real medical data. Experiment leads to the conclusion that automatic methods of phase correction are important part of ¹H NMR data pre processing. However there is still chance to improve results by proposing own modifications based on the idea of parameters tuning.

1. Introduction.

NMR (abr. Nuclear Magnetic Resonance) is a bio-chemical technique used to determine chemical compounds of tested specimen. It is based on a phenomenon of nuclear resonance that may take place in a nucleons that poses magnetic spin (e.g. ¹H, ¹³C). Such a nucleons are placed in homogenous magnetic field that is used as a reference. In the next step electromagnetic pulse of defined strength and frequency is applied to the specimen. It causes forced change of nucleus spin.

After pulse removal, particles get back to its original spin in a process called precession. In a consequence, energy wave is emitted. Such a signal has its own harmonic frequency and the coil of the spectrometer detects it. Harmonic frequency of emitted wave are influenced by two factors:

- Electromagnetic neighborhood of nucleus (electron clouds)
- Chemical bounds between nucleus, under resonance, and other present in chemical structure of specimen.

Because of these two facts one is able to determine exact structure of chemical compound that contain nucleus that is under resonance, basing on a known dependencies between nucleons.

As a result of NMR test a so-called Free Induction Decay (FID) signal is obtained. It is a time domain signal containing in fact decaying precession wave emitted from nucleons. It is worth to notice that such a signal is 3 dimensional one, however only its two projections are collected. As a result of such a methodology, signal obtained from spectrometer is in form of complex number which real and imaginary part represents both projections. Obtained FID is in fact one signal that contains all frequency components originate from all nucleons under resonance.

NMR is widely used in biology, chemistry and medicine. One of its applications is called MRI (Magnetic Resonance Imaging) a very popular medical imaging technique. Such a method is in fact pure NMR spectroscopy which is selective to one chemical compound (e.g. water) and present its distribution across tested specimen in a form of gradient image.

¹H NMR (magnetic resonance of ¹H nucleons) spectroscopy is commonly used to diagnose tumors present in human body. One of possible application is determination of brain tumor metabolism. Exemplary spectrum taken on human brain is presented on Fig.1.

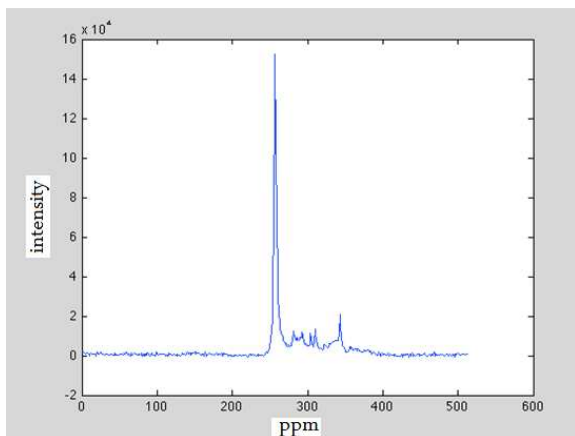


Figure 1. Exemplary spectrum obtained on human brain.

In order to do so, spectrum obtained from spectrometer must be transformed into form that may be analyzed. As it was mentioned raw data are time domain signal. Such a set of data must be transformed from time domain into frequency one (because harmonic frequencies should be determined). The simplest solution is to use one of Fourier Transform variation, called Fast Fourier Transform (FFT). Of course one may notice that all unwanted components such as noise, baseline and dominating water/reference signal are also present in transformed signal. Moreover, dividing 3D signal into projections is cause of so called phase errors. All mentioned artifacts should be removed from the spectrum before final steps that is identification of chemical components and their amounts in tested area.

2.Signal pre-processing.

As it was mentioned in previous paragraph raw spectrum contains distortions, which should be removed before signal analysis. Such a procedure may be divided into two parts:

- Signal normalization- containing FFT transform (already mentioned), noise filtration, baseline removal and phase correction.
- Subtraction of dominating reference signal from the spectrum

3.Phase error problem.

One of mentioned distortions present in signal obtained from spectrometer is Phase Error, which follows from the test methodology. (mentioned in previous paragraphs)

According to the literature and test procedures peaks that are visible in frequency spectrum should poses Lorentz line shape. Moreover it should be complex Lorentz line shape as analyzed signal is in form of a complex number. It is also assumed that spectrum obtained from real part of signal will be in form of so-called absorption line shape, and imaginary spectrum will be in form of so called dispersion

spectrum. Exemplary absorption and dispersion line shapes are presented on the Fig.2.

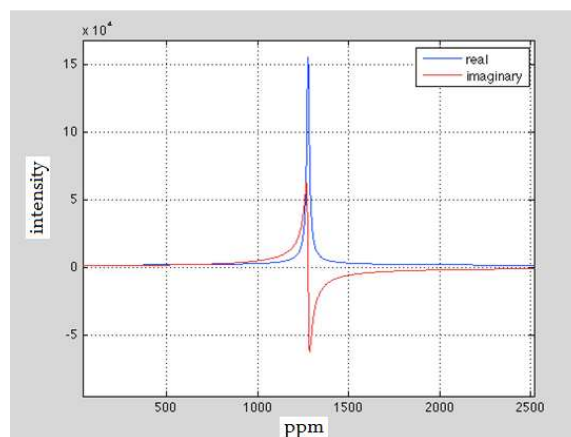


Figure 2. Exemplary absorption (red) and dispersion (blue) line shape of the peak(component) of the signal.

What is worth to notice both parts (real and imaginary) of the signal may be presented together on one plot (complex plane). As it was noticed correct line shapes of both parts of the signal result in an ellipse-form plot on such a complex plane. Such an ellipse is symmetrical according to X (real in complex plane) axis and has its origin in the center of coordinate system. Moreover according to the literature we may assume that phase error does not influence magnitude spectrum. Connecting both mentioned facts it is possible to derive quality criterion for methods under investigation. Such a criterion assumes that maximal magnitude of signal may be treated as the longer diameter of the ellipse shape. In addition magnitude is not changing with phase error thus we are interested only in difference between phase angle of maximal magnitude and 0 (according to the assumption that proper ellipse shape is symmetrical according to real axis of complex plane). Exemplary ellipse on complex plane is presented on Fig.3. Such a shape is also called Dispa (Dispersion vs. absorption) plot according to notation of real and complex plot of the spectrum.

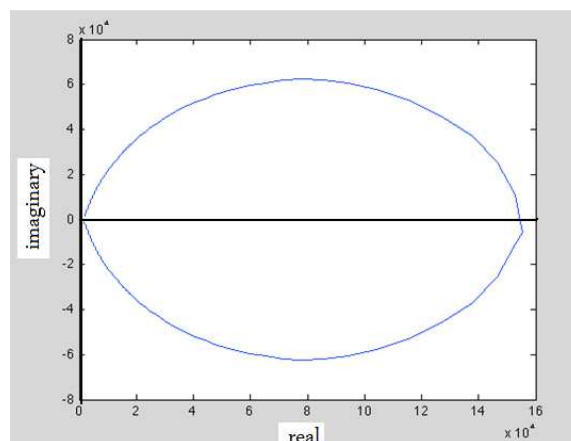


Figure 3. Exemplary Dispa plot obtained for proper line shape spectrum.

3.1 Entropy minimization method.

The first method that was under investigation was called „entropy minimization”. It is based on calculation of so called Shannon entropy of the given NMR spectrum. The problem of minimization is given by:

$$\text{Min } S = - \sum h_i \cdot \ln(h_i) + P \quad (1)$$

, Where S is Shannon entropy, h is probability distribution of the given spectrum as its normalized first derivative:

$$h_i = \frac{(R'_i)}{\sum(R'_i)} \quad (2)$$

And P is penalty function added to the function to ensure elimination of negative bands in final spectrum:

$$P = \gamma[a * R_i^2] \quad (3)$$

, Where

$$a = \begin{cases} 0 & \text{for } R_i \geq 0 \\ 1 & \text{for } R_i < 0 \end{cases} \quad (4)$$

According to the authors of the method parameter γ , which is named penalty factor, should be set to the proper value to eliminate level difference between values of entropy and penalty function.

3.2.eDispa method.

The second mentioned method is called eDispa and it was published in 2007 by Cobas and Sykora. Authors defined a so-called Q-factor functional of the form:

$$q = (|Y_k|^a \cdot R_k^b) \cdot \exp\left(\frac{-w \cdot (2k - N)}{N}\right) \quad (5)$$

, Where

Y_k -is a complex point of given spectrum after phase error correction.

$$Y_k = y_k \cdot \exp\left(-j \cdot \left(\frac{\varphi + 2\pi\tau(k - N)}{N}\right)\right) \quad (6)$$

y_k -is a complex point of spectrum before phase correction

N-length of given spectrum

φ, τ -Parameters of defined functional

a,b,w-parameters, set empirically by authors of the method (a=2,b=1,w=2).

It is also worth to notice that only y_k that fulfils condition (7) are taken into account.

$$|y_k| \geq c \quad (7)$$

, Where c was defined to be equal 0.1. However this parameter may be tuned to acquire better and more precise result.

Having defined functional it is important to properly define range of parameters φ, τ for which the algorithm would be run. As a next step for every value of τ q functional factor value is calculated. It is worth to notice that for single value of τ q factor should be calculated for every defined parameter φ .

Knowing these values, the maximal one should be chosen and set as part of new functional $Q(\tau)$. Moreover for every $Q(\tau)$, φ parameter for which it was obtained should be stored as another defined functional $\Phi(\tau)$. After performing above calculations for all defined values of coefficient τ functional $\eta(\tau)$ is defined which is normalized $Q(\tau)$ raised to the fourth power.

$$\eta(\tau) = 360 \left(\frac{Q(\tau) - \min Q(\tau)}{\max Q(\tau) - \min Q(\tau)} \right)^4 \quad (8)$$

Optimal values of parameters for maximal value of functional $\eta(\tau)$ - τ value, and corresponding $\Phi(\tau)$ - φ value. Corrected spectrum is then obtained according to equation (6).

3.3. Automics method.

The method was proposed by Wang et al. in 2009. Its main idea is to define phase errors at the beginning and end of frequency spectrum. In the first step four small regions should be defined. Two of them should lie in low frequency region and two in high frequency region. All our intervals should belong to the part of the spectrum, which is free of significant peaks (only baseline and noise). Authors do not strictly define length of the region. It is worth to notice that its length may be treated as additional parameter that after proper tuning may improve accuracy of the method. All defined regions should be summed up to get four complex numbers Z_1, Z_2, Z_3, Z_4 with real parts: R_1, R_2, R_3, R_4 and imaginary parts I_1, I_2, I_3, I_4 . The center coordinate of every region should be stored as well, and denoted with c_1, c_2, c_3, c_4 . The main assumption of the method is that magnitudes of Z_1 and Z_2 should be more or less equal as well as magnitudes of Z_3 and Z_4 .

$$\begin{aligned} R_1 \cos(\theta_0) + I_1 \sin(\theta_0) \\ = R_2 \cos(\theta_0) \\ + I_2 \sin(\theta_0) \end{aligned} \quad (9)$$

$$\begin{aligned} R_3 \cos(\theta_1) + I_3 \sin(\theta_1) \\ = R_3 \cos(\theta_1) \\ + I_3 \sin(\theta_1) \end{aligned} \quad (10)$$

The phase errors at the beginning and at the end of the spectrum are then calculated according to formulas:

$$\theta_0 = \text{atan}\left(\frac{R_1 - R_2}{I_1 - I_2}\right) \quad (11)$$

$$\theta_1 = \text{atan}\left(\frac{R_3 - R_4}{I_3 - I_4}\right) \quad (12)$$

As a last step first and zero order phase coefficient (phc_0 and phc_1) must be calculated, according to formula:

$$\theta_0 = \text{phc}_0 + \left(\frac{c_1 + c_2}{N}\right) \cdot \text{phc}_1 \quad (13)$$

$$\theta_1 = \text{phc}_0 + \left(\frac{c_3 + c_4}{N}\right) \cdot \text{phc}_1 \quad (14)$$

Using values obtained from equation 13 and 14 spectrum should be corrected.

4.Experiments.

To check quality and effectiveness of described phase correction methods, experiments

were made. It is quite difficult to obtain set of spectra that are similar (comparable number of peaks) but differ in phase error (observable trend: increase or decrease) and noise. That is why artificial set of data were created to determine the quality of method result according to varying phase error and additionally presence of noise, which was suspected to has influence. The artificial data set were prepared in a form of 3 dimensional table for which: first dimension corresponds to spectrum resolution (number of points), second dimension corresponds to varying phase error from $-\pi/2$ to $\pi/2$ and third dimension corresponds to varying noise of level from $\text{SNR} = 1$ up to 8%. The result for every mentioned phase correction technique was presented on the separate graph. In order to decrease number of plots level of noise is not varying but SNR is equal 4%.

5.Results

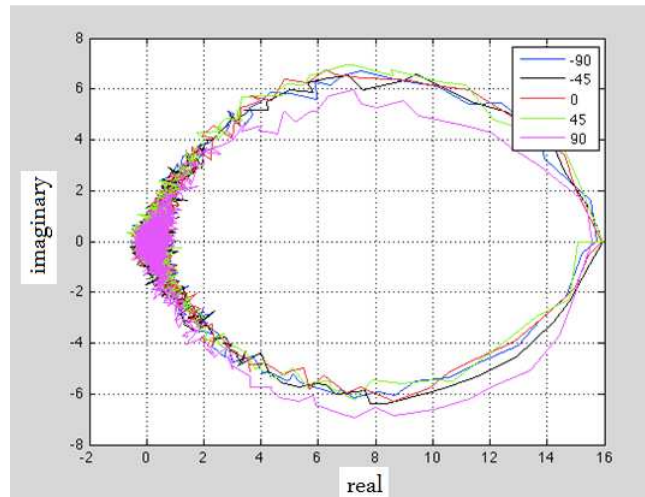


Fig. 4. Dispa plot of corrected spectra obtained for eDispa method, for different phase errors.

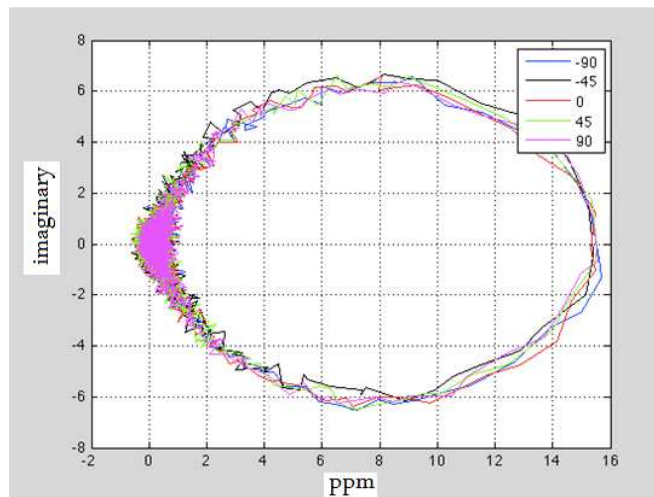


Fig. 5. Dispa plot of corrected spectra obtained for Automics method, for different phase errors.

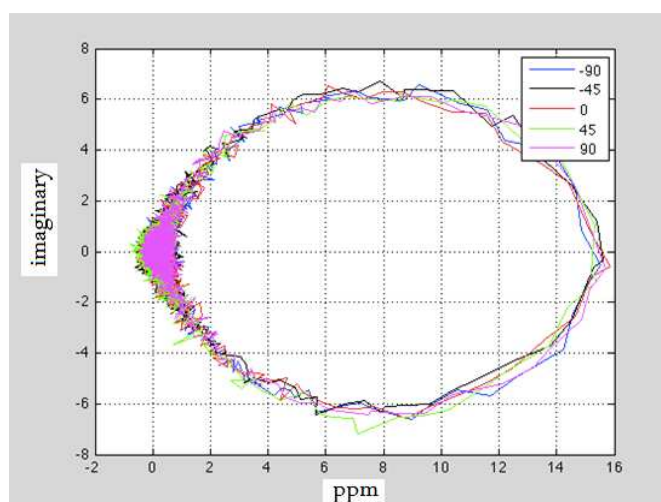


Fig. 6. Dispa plot of corrected spectra obtained for entropy minimization method, for different phase errors.

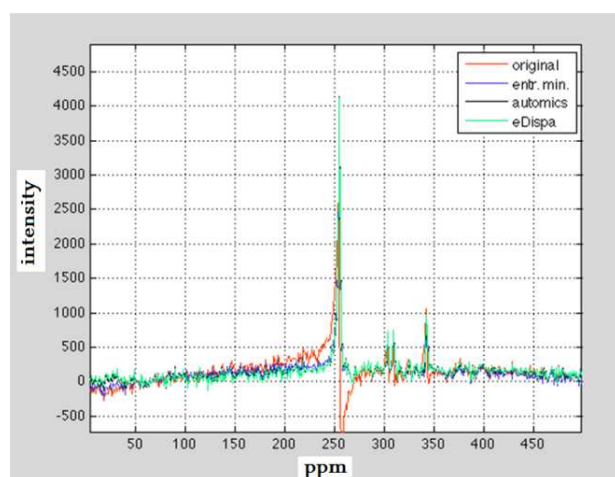


Figure 7. Exemplary comparison of mentioned algorithms-absorption spectrum.

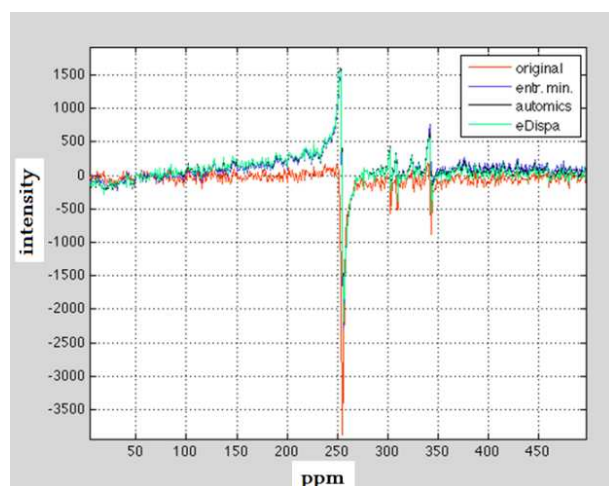


Figure 8. Exemplary comparison of mentioned algorithms-dispersion spectrum.

6. Summary.

As it was shown in experiment all mentioned and implemented methods deal with the given problem quite well. However there is visible that there exist some differences in their precision,

(according to assumed quality criterion of ellipse symmetry), other words there is still scope for improvements. It is worth to notice that all of mentioned methods have parameters that are not strictly defined by their authors but left to set by method users according to the data under experiment. This lead to the conclusion that such a parameters are able to be tuned to optimal value in order to obtain much better quality of final result-spectrum properly phased.

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Authors:



MSc. Franciszek
Binczyk
Politechnika Śląska w
Gliwicach, Automatic
Control Department
ul. Akademicka 16
44-100 Gliwice
tel. 792727518

email: franciszek.e.binczyk@gmail.com
