High-resolution at 3T for in vivo derivative NMR spectroscopy in medical diagnostics of ovarian tumor: exact quantification by shape estimations

Dževad Belkić^{1,2} · Karen Belkić^{1,2,3,4}

Received: 15 August 2021 / Accepted: 20 August 2021 / Published online: 8 September 2021
© The Author(s) 2021

Abstract

Time signals are measured experimentally throughout sciences, technologies and industries. Of particular interest here is the focus on time signals encoded by means of magnetic resonance spectroscopy (MRS). The great majority of generic time signals are equivalent to auto-correlation functions from quantum physics. Therefore, a quantum-mechanical theory of measurements of encoded MRS time signals is achievable by performing quantum-mechanical spectral analysis. When time signals are measured, such an analysis becomes an inverse problem (harmonic inversion) with the task of reconstruction of the fundamental frequencies and the corresponding amplitudes. These complex-valued nodal parameters are the building blocks of the associated resonances in the frequency spectrum. Customarily, the MRS literature reports on fitting some *ad hoc* mathematical expressions to a set of resonances in a Fourier spectrum to extract their positions, widths and heights. Instead, an alternative would be to diagonalize the so-called data matrix with the signal points as its elements and to extract the resonance parameters without varying any adjusting, free constants as these would be absent altogether. Such a data matrix (the Hankel matrix) is from the category of the evolution matrix in the Schrödinger picture of quantum mechanics. Therefore, the spectrum of this matrix, i.e. the eigenvalues and the corresponding amplitudes, as the Cauchy residues (that are the squared projections of the full wave functions of the system onto the initial state) are equivalent to the sought resonance parameters, just mentioned. The lineshape profile of the frequency-dependent quantum-mechanical spectral envelope is given by the Heaviside partial fraction sum. Each term (i.e. every partial fraction) in this summation represents a component lineshape to be assigned to a given molecule (metabolite) in the tissue scanned by MRS. This is far reaching, since such a procedure allows reconstruction of the most basic quantum-mechanical entities, e.g. the total wave function of the investigated system and its 'Hamiltonian' (a generator of the dynamics), directly from the encoded time signals. Since quantum mechanics operates with abstract objects, it can be applied to any system including living species. For example, time signals measured from the brain of a human being can be analyzed along

Extended author information available on the last page of the article

these lines, as has actually been done e.g. by our research. In this way, one can arrive at a quantum-mechanical description of the dynamics of vital organs of the patient by retrieving the interactions as the most important parts of various pathways of the tissue functions and metabolism. Of practical importance is that the outlined quantum-mechanical prediction of the frequency spectrum coincides with the Padé approximant, which is in signal processing alternatively called the fast Padé transform (FPT) for nonderivative estimations. Further, there is a novelty called the derivative fast Padé transform (dFPT). The FPT and dFPT passed the test of time with three fundamentally different time signals, synthesized (noise-free, noise-contaminated) as well as encoded from phantoms and from patients. Such systematics are necessary as they permit robust and reliable benchmarkings of the theory in a manner which can build confidence of the physician, while interpreting the patient's data and making the appropriate diagnosis. In the present study, we pursue further this road paved earlier by applying the FPT and dFPT (both as shape and parameter estimators) to time signals encoded by *in vivo* proton MRS from an ovarian tumor. A clinical 3T scanner is used for encoding at a short echo time (30 ms) at which most resonances have not reached yet their decay mode and, as such, could be detected to assist with diagnostics. We have two goals, mathematical and clinical. First, we want to find out whether particularly the nonparametric dFPT, as a shape estimator, can accurately quantify. Secondly, we want to determine whether this processor can provide reliable information for evaluating an ovarian tumor. From the obtained results, it follows that both goals have met with success. The nonparametric dFPT, from its onset as a shape estimator, transformed itself into a parameter estimator. Its quantification capabilities are confirmed by reproducing the components reconstructed by the parametric dFPT. Thereby, fully quantified information is provided to such a precise extent that a large number of sharp resonances (more than 160) appear as being well isolated and, thus, assignable to the known metabolites with no ambiguities. Importantly, some of these metabolites are recognized cancer biomarkers (e.g. choline, phosphocholine, lactate). Also, broader resonances assigned to macromolecules are quantifiable by a sequential estimation (after subtracting the formerly quantified sharp resonances and processing the residual spectrum by the nonparametric dFPT). This is essential too as the presence of macromolecules in nonderivative envelopes deceptively exaggerates the intensities of sharper resonances and, hence, can be misleading for diagnostics. The dFPT, as the quantification-equipped shape estimator, rules out such possibilities as wider resonances can be separately quantified. This, in turn, helps make adequate assessment of the true yield from sharp resonances assigned to metabolites of recognized diagnostic relevance.

Keywords NMR spectroscopy · Ovarian tumor · Derivative estimations · Fast Fourier transform · Fast Padé transform

1 Introduction

In nuclear magnetic resonance (NMR) spectroscopy, or magnetic resonance spectroscopy (MRS) as it is called in medical diagnostics, the main problems are low sensitivity, compared to all other spectroscopies. This stems from the fact that in e.g. ^1H MRS only a very small fraction of protons from a Boltzmann quasi-equilibrium population enables the transition between the two Zeeman energy levels to yield the magnetic resonance (MR) effect. This number is merely about 10 or 20 out of a million spins at room temperature at the static magnetic field strengths $B_0 = 1.5$ and 3T, respectively. Such an occurrence can give only some weak signals that are difficult to detect. This worried researchers since the time of the discovery of the MR phenomenon [1–7]. Eventually, powerful measuring devices (MR spectrometers, clinical scanners) have been constructed to bring this obstacle under control.

However, notwithstanding the steady progress in the hardware, there still remains the issue about the software for analysis and interpretation of the recorded data. It is here where theory comes into play with its signal processing to tackle the two main hurdles, strongly coupled together, low resolution and poor signal-to-noise ratio (SNR), especially for in vivo MRS at clinical scanners (1.5 and 3T).

How to unequivocally separate the physical or genuine from nonphysical or spurious (“noisy”) information? As always in science, asking the right question, as the critical part of research, might beneficially provide some useful insights into the potential solutions of the studied problem. The posed question, although being critical for further advances in signal processing, has been addressed most frequently in some phenomenological/empirical ways. Such approaches could have not offered any mechanism for robust disentangling physical from unphysical information in the encoded data. The measured data in NMR spectroscopy are time signals, or equivalently, free induction decay (FID) digitized curves.

However, of late, a veritable renaissance took place in analyses of encoded MRS data, be they from phantoms or from patients. It led to a breakthrough by establishing the powerful concept of signal-noise-separation (SNS) [8]. The mechanism governing SNS is the phenomenon of pole-zero cancellations through unequivocal identification of Froissart doublets [9, 10]. This strategy is uniquely germane to the Padé approximant (PA) [11–13]. In the present context, the PA is a frequency-dependent function in the form of a ratio of two polynomials, P/Q , which represents a total shape spectrum or envelope. More generally, the quotient P/Q can have different observables as the independent variables.

The Padé rational polynomial is the most frequently employed method within theory of approximations in applied mathematics. It is used for data analyses in vastly different branches ranging from physics, chemistry through signal processing in biology, medicine to engineering, technologies and industries [14–19]. It is alternatively called the fast Padé transform (FPT) in signal processing [20, 21]. This was done to emphasize the dual representation of the PA with its autonomous direct and inverse transformations from the time to the frequency domain

with no need at all to use the corresponding fast Fourier transform (FFT) nor the inverse fast Fourier transform (IFFT) [17], as has explicitly been shown in Ref. [19] in the footsteps of Prony [11].

The adjective fast in the FPT is justified because e.g. the diagonal form P_K/Q_K can be computed by means of the Euclid algorithm [22, 23] with the computational complexity of $K \log^2 K$ multiplications. To extract the expansion coefficients $\{p_r\}$ and $\{q_s\}$ ($0 \leq r, s \leq K$) of polynomials P_K and Q_K , respectively, one solves a system of linear equations set up by the definition of the Padé approximant, $(a_0 + a_1 u + \dots + a_{2K} u^{2K}) Q_K(u) = P_K(u)$, where the expansion coefficients $\{a_m\}$ are known. This is recognized as the Toeplitz system of linear equations for finding the unknown coefficients $\{p_r, q_s\}$. A Hankel matrix $\mathbf{H}_{K \times K}$ (often called 'data matrix' in signal processing) is a Toeplitz matrix $\mathbf{T}_{K \times K}$ with its columns reversed. Thus, bearing in mind this simple change in notation, solving a system of linear equations with Hankel matrix $\mathbf{H}_{K \times K}$ amounts to solving the equivalent system with Toeplitz matrix $\mathbf{T}_{K \times K}$.

The usefulness of the equivalence of the system of linear equations for $\{p_r, q_s\}$ ($0 \leq r, s \leq K$) in the PA with the Toeplitz system for $\mathbf{T}_{K \times K}$ is in saving both computational time ($K \log^2 K$) and space/memory allocation ($K \log K$). Here, unlike the FFT algorithms, K does not need to be of any particular form (e.g. a prime number or $K = 2^\ell$, where ℓ is a positive integer, etc.). Similar advantages also apply to the general PA in the form P_L/Q_K of unequal polynomial degrees L and K . Without the fast Euclid algorithm, the number of multiplications in solving the system of linear equations in the PA would be K^2 . For smaller values of K , the $K \log^2 K$ multiplications may not be advantageous at all over the K^2 multiplications. However, for larger K , the Euclid algorithm, with its $K \log^2 K$ multiplications, is much faster than any other computations with the K^2 multiplications.

In physics the PA is abundantly used in practically all the sub-disciplines such as solid state physics, plasma, atomic and nuclear physics and sub-nuclear physics (elementary particle physics, quantum field theories, ...). Although versatile applications of the PA are enormous in physics alone, here are only a few of the typical examples:

- *Acceleration of slowly converging series/sequences.* Such series are usually encountered in physical system exposed to external fields (e.g. Zeeman effect, Stark effect, etc.) [24–30]. The coefficients of these series are provided by the Rayleigh-Ritz perturbation theory for nondegenerate states.
- *Inducing convergence in diverging series/sequences.* Here, the Cauchy concept of analytical continuation is exploited. For instance, problems in particle physics dealing with strong interactions encounter diverging perturbation series due to a strong coupling used as the expansion parameter. The PA comes to the rescue by performing the so-called resummation of the originally divergent series. This amounts to forcing the given divergent series to converge by mapping it to a ratio of two polynomials P/Q that can yield the finite and, therefore, physical result.

This is remarkable, especially given that the perturbation expansions are the only practical/calculable method for strong interactions among elementary particles.

Here, one invests a huge effort to obtain higher-order perturbation terms only to find out in the end that the series is divergent. Fortunately, the effort is not lost because the PA salvages the matter (by inducing convergence into divergent series) and, thus, enables the theory of strongly interacting particles to work. Similarly, the PA resummed the divergent Brillouin–Wigner perturbation theory for degenerate states and even provided the bounds to the exact degenerate energies.

- *Performing exact quantification of all spectroscopic data based on time signals, encoded and/or synthesized.* The quantification problem consists of carrying out spectral analysis to reconstruct the dynamics of the system which produced the time signal as the response to external perturbation. It is an inverse problem called harmonic inversion since time signals are exponentially damped harmonics. Generally, solving an 'inverse problem' amounts to determining the unknown *causes* by relying upon the observed/measured *effects*. Virtually all of medicine is based on the concept of inverse problems. Moreover, all but trivial experimental measurements in laboratories and observations in the outer space are basically inverse problems.

With in vivo MRS, noninvasively and without ionizing radiation, it is possible to peer into the chemical content of the scanned tissue. The system dynamics are characterized by a fixed number, say K , of spectral parameters. These are the complex fundamental frequencies $\{\omega_k\}$ and the corresponding amplitudes $\{d_k\}$. Such nodal parameters are the eigen-characteristics of the molecule-specific harmonic attenuated oscillations in the tissue. Molecules that take part in various metabolic processes in tissue are called metabolites. Using in vivo MRS, the quantification problem would be solved if the fundamental set $\{\omega_k, d_k\}$ ($1 \leq k \leq K$) of all the genuine damped harmonic oscillations in the tissue could reliably be recovered for the encoded FIDs. This includes determination of the total number K of the metabolites that are physically present in the scanned tissue.

But how can one be sure that the reconstructions contain the entire information? Could it be that there are some information losses in data analyses? The answer can be given by quantum-mechanical signal processing. According to the completeness relation in quantum mechanics, anything that could possibly be extracted from the given system, using the pertinent measured data, is fully contained in the nonstationary $\Phi(t)$ and stationary Ψ_k total wave functions, satisfying the dynamical Schrödinger equations. However, these wave functions, describing the states of the system, are the solution of the direct problems, that are the time-dependent and time-independent Schrödinger equations, with the known dynamics generator, the 'Hamiltonian' of the system, Ω . The quantification problem is not a direct problem, as stated. Therefore, if we could somehow extend quantum mechanics to encompass also inverse problems, we would be in a position to guarantee that there would be no information losses in harmonic inversion either.

It is indeed possible to extend quantum-mechanical Schrödinger equations and the completeness relation to harmonic inversion without knowing the dynamic operator Ω of the system. This is done by expanding the state wave function Ψ_k in the Krylov nonorthogonal basis, which is comprised of the discretized nonstationary

Schrödinger states $\{\Phi_n\}$. The expansion coefficients are column vectors $\mathbf{A} = \{A_{n,k}\}$. In this basis, all the matrix elements of the quantum-mechanical time evolution operator $e^{-i\Omega t}$ are given in terms of the encoded set of time signal data points $\{c_n\}$ ($0 \leq n \leq N-1$) of length N .

Due to nonorthogonality of the expansion functions $\{\Phi_n\}$ ($1 \leq n \leq M$), the nonzero overlap matrix elements $(\Phi_m|\Phi_n)$ appear for $m \neq n$. Therefore, the generalized eigen-value problem of the evolution matrix $\mathbf{U}$ is solved yielding the exact solutions for the expansion coefficients $\{A_{n,k}\}$ and the fundamental frequencies $\{\omega_k\}$. From here, the corresponding exact amplitudes $\{d_k\}$ are deduced as the squared convolutions of $\{A_{n,k}\}$ and $\{c_n\}$. This is how quantum mechanics works for harmonic inversion. All the mentioned explicit expressions are given in e.g. Refs. [17, 31].

There is more to this quantum-mechanical spectral analysis, which solves exactly the quantification problem in NMR spectroscopy. Namely, the envelope in the frequency domain, predicted by quantum mechanics, is given by the Heaviside sum of exactly K partial fractions (component spectra). When summed up, this Heaviside partial fraction representation coincides with the paradiagonal Padé approximant, P_{K-1}/Q_K . This means that one can alternatively use the FPT to solve the quantification problem without ever dealing with more involved generalized eigen-value problem of the evolution matrix. Simply, rooting the denominator polynomial by solving the secular equation $Q_K = 0$, one would obtain all the fundamental frequencies $\{\omega_k\}$.

In fact, this latter nonlinear operation is replaced by its equivalent linear counterpart, which is the eigen-value problem of the Hessenberg matrix. This matrix is extremely sparse because its elements are the expansion coefficients $\{q_s\}$ of Q_K lying on the first row, unity on the main diagonal and zero elsewhere. Moreover, in the FPT, the amplitudes $\{d_k\}$ are computed easily from the analytical expression for the Cauchy residue of P_{K-1}/Q_K taken at the obtained eigen-roots $\{\omega_k\}$ of Q_K . Since P_{K-1}/Q_K is a meromorphic function (having poles as the only singularities), the roots of $P_{K-1} = 0$ and $Q_K = 0$ are the zeros and poles of the system's response function P_{K-1}/Q_K , respectively.

- *Unambiguously disentangling the physical signal from noise.* The mentioned SNS concept relies upon pole-zero coincidences (Froissart doublets) and the ensuing pole-zero cancellation. Noise from the encoded FIDs is shared by P_{K-1} and Q_K in the Padé envelope P_{K-1}/Q_K . The expansion coefficients of the numerator P_{K-1} and denominator polynomials Q_K inherit the noise from the encoded FID. This inheritance is equi-partitioned between the roots of P_{K-1} and Q_K that coincide (pole-zero coincidences). In other words, noise appears in pairs, poles and zeros of P_{K-1}/Q_K (Froissart doublets). They cancel out in the quotient P_{K-1}/Q_K (pole-zero cancellations). This is the signature of SNS. Noise is gone from the envelope. It is in such a way that the FPT implements its self-correction, the denoising Froissart filter (DFF). Noise is recognized as error and, as such, discarded from the reconstructed total shape spectrum, P_{K-1}/Q_K .

The SNS and DFF concepts, with the underlying pole-zero coincidence, are transparently illustrated by the Argand plot for the real and imaginary parts of the roots of each of the two polynomials, P_{K-1} and Q_K . This plot enables comparisons of

the distributions of poles and zeros of P_{K-1}/Q_K in the complex frequency plane. It clearly shows the two diametrically opposite patterns for the physical (genuine) and unphysical (spurious, noisy) frequencies. The noisy poles and zeros coincide in the Argand plot, whereas the physical poles and zeros appear as nonconfluent. In the end, we are left with genuine resonances alone. This is how the mechanism of signal-noise separation, SNS, is manifested in practice.

This mechanism is also self-explanatory from the mentioned Cauchy residue for the analytical formula for the amplitudes $\{d_k\}$. Each reconstructed amplitude d_k is proportional to the product of differences of all the roots of P_{K-1} and Q_K . Therein, all the noisy roots of P_{K-1} and Q_K are confluent, implying that the entire product is zero and so is d_k for any spurious frequency ω_k . Overall, this twofold signature proves the veracity and robustness of the concept of signal-noise separation, SNS, through the denoising Froissart filter, DFF: (i) coincidence of noisy poles with noisy zeros, and (ii) vanishing of noisy amplitudes.

- *Earlier studies applying the FPT to MRS time signals from the ovary*

Among our initial studies using the FPT were for noise-free and noise-corrupted time signals, associated with malignant and benign ovarian cyst fluids [19, 32–36], similar to in vitro MRS encoding from Ref. [37]. Despite a high magnetic field of a Bruker spectrometer (600 MHz $\approx$ 14.1 T) employed in Ref. [37], only 12 resonances were quantified by integration. Therein, for all the other numerous resonances, it was not possible to determine the integration boundaries for a numerical quadrature to approximately assess the peak areas in order to deduce concentrations of metabolites. This difficulty itself points to the need for a more adequate way of performing quantification.

Subsequently [19, 32–36], using the parameters of the 12 resonances from Ref. [37], we synthesized the corresponding time signals and subjected them to the FFT and FPT. For the noiseless FIDs, with only 64 time signal points, all the 12 input resonances were resolved by the FPT and the corresponding metabolite concentrations were exactly computed [19, 32, 33]. In sharp contradistinction, with 64 time signal points, the FFT produced only rudimentary envelopes. In fact, the FFT needed a huge set of some 32768 signal points for convergence of envelopes. With increasing levels of noise, the FPT was also able to resolve and correctly quantify all the 12 resonances for benign and cancerous ovarian cyst fluids.

In our follow-up study [38], comparisons were made between the resolution capabilities of the FPT and FFT applied to in vivo MRS time signals encoded from borderline serous cystic ovary. Encoding was made with a 3T clinical scanner at two echo times (TE) of 30 and 136 ms [39]. We employed the FIDs encoded at TE = 30 ms. The spectra averaging procedure [40] was successfully applied through the FPT to attenuate the noisy spikes. Therein, an unstable spike at about 3.4 ppm was the largest structure. Moreover, there were many other prominent unstable spikes intermingled at frequencies belonging to the spectral range of interest (SRI): 0.7–3.75 ppm. Using the 11 reconstructed complex envelopes ($K = 575, 580, \dots, 620, 625$), an average total shape spectrum was generated. In it, the unstable (noisy, spurious, unphysical) spikes disappeared altogether and only the stable (physical, genuine) peaks remained. The

largest physical peaks were nitrogen-acetyl neuraminic acid (acNeu) at 2.06 ppm and nitrogen-acetyl aspartate (NAA) at 2.03 ppm.

Many other genuine resonances could also be identified. Among the diagnostically most notable were a lactate (Lac) doublet and lipids (Lip) at about 1.3 ppm, glutamate (Glu) and glutamine (Gln) at around 2.45 ppm, a myo-inositol (m-Ins) triplet at around 2.6 ppm, choline (Cho) at 3.2 ppm, a small phosphocholine (PC) peak at 3.22 ppm, a glycerophosphocholine (GPC) peak at 3.23, etc. When Glu and Gln are not seen as separate peaks, they are usually labeled by Glx, i.e. $\text{Glx} = \text{Glu} + \text{Gln}$. Similarly, total choline (tCho) is used to denote the compound of 3 resonances, Cho, GPC and PC, i.e. $\text{tCho} = \text{Cho} + \text{GPC} + \text{PC}$. The real parts of the so-called 'Ersatz' component spectra were displayed [38], helping to visualize overlap of closely-lying or hidden resonances. In the parametric FPT, the peak positions, widths, heights and phases are reconstructed exactly for all the resonances.

When the phases $\{\phi_k\}$ of the amplitudes $\{d_k\} = \{|d_k|e^{i\phi_k}\}$ become available, each resonance can be phase-corrected through multiplication of the amplitudes $\{d_k\}$ by $\{e^{-i\phi_k}\}$ to cancel $\{e^{i\phi_k}\}$ in $\{|d_k|e^{i\phi_k}\}$. This gives the phase-corrected amplitudes $\{d_k\} \times \{e^{-i\phi_k}\} = \{|d_k|e^{i\phi_k}\} \times \{e^{-i\phi_k}\}$ that evidently become the magnitudes $\{|d_k|\}$, i.e. pure real numbers. Such a procedure is, of course, equivalent to setting $\phi_k = 0$ in every complex amplitude $d_k = |d_k|e^{i\phi_k}$. Viewed in either of these two ways, the net result is the same: removal of interference effects. This implies that the real parts of all the resonances are purely absorptive Lorentzians. For such 90 phase-corrected 'Ersatz' components, almost complete convergence to the level of data stochasticity was attained throughout the entire SRI (0.7–3.75 ppm) [38].

With spectra averaging and extrapolation used in constructing the components (with and without phase corrections), the spectral parameters fully converged [41]. It is from the components with complex amplitudes $\{d_k\}$ (no phase corrections, the so-named 'Usual' components), by fully taking into account all the interference effects, that metabolite concentrations are reliably computed. This parametric FPT serves as the gold standard for quantification in MRS. Subsequently, within the SNS concept, applying the FPT to in vivo MRS time signals encoded from the ovary, the role of spectral poles and zeros was examined, as the key to stability of the system to external perturbations [42].

After spectra averaging, we implemented time signal extrapolation. This is done by applying the IFFT to the stabilized envelope obtained by spectra averaging in the FPT. The result is the new time signal. Because the envelopes in the FPT can be computed at any number of frequencies, we can utilize a grid/mesh longer than the total signal length N of the originally encoded FID. Therefore, inversion of the given Padé envelope of length longer than N leads to an extrapolated time signal.

Such an extrapolation is physical (unlike the one followed by zero filling of the FID) since it uses the encoded time signal points contained in the Padé envelope, which is inverted. Also, from the mathematical stance, the Padé extrapolation is realistic as it employs the rational polynomials that produce no spectral artifacts (wiggles, Gibbs phenomena, ...). In the FFT, a zero-filled FID leads to a longer Fourier envelope. However, a zero-filled FID yields only a trigonometric interpolation in the frequency domain [43–46] and the ensuing elongated Fourier envelope is always distorted by the appearance of spectral sharp-edged wiggles.

The Padé rational polynomials within the FPT interpolate and extrapolate. Genuine extrapolation amounts to prediction. Hence, the Padé rational polynomial makes the FPT a prediction method, which can reliably generate the FID data points beyond the original total acquisition time $T = N\tau$ in the encoding (τ is the sampling or dwell time). The FFT is not a predictive model as the interpolating trigonometric functions after zero filling of the FID are artificial and with no relation whatsoever with the encoded time signal data points.

In the FPT, coupling of spectra averaging and FID extrapolation is a powerful platform for investigation of convergence of all the variables under study. Thus, in Refs. [41, 42], it was demonstrated that spectra averaging and Padé-based extrapolation of time signals were crucial for the reconstructed poles and zeros, as well as for the associated magnitudes and phases. This procedure was necessary to check the stability of the retrieved fundamental parameters and to accurately reconstruct all the physical component resonances.

Overall, the systematic implementation of spectra averaging and time signal extrapolation in the FPT was shown [41, 42] to be critical for solving the major drawback of all parametric methods. This drawback is the pronounced instability of reconstructions to changes in the model order K . Physically, K is the true number of metabolites. Therefore, this key parameter must also be exactly reconstructed alongside the complex fundamental frequencies and amplitudes. In the MRS literature, the abundant fitting techniques consider K as a guessing quantity. This invariably leads to finding the nonexistent metabolites in the scanned tissue and/or failing to detect some of the true metabolites. Both deficiencies of all the fitting recipes [47–49] are anathema to medical diagnostics.

2 Theory

The theory of the diagonal FPT to be used in the present work is summarized in our most recent study [31] and here only the main outlines need to be given. In Ref. [31], the $\text{FPT}^{(-)}$ version was employed, where the minus superscript refers to the harmonic variable with the minus sign in the exponent, $z^{-1} = e^{-i\omega\tau}$. Presently, the same Padé variant will be used throughout and, therefore, the acronym $\text{FPT}^{(-)}$ will be simplified as FPT. As is well-known, the total shape spectrum or envelope in the diagonal version of this method is given by the ratio of two polynomials, P_K and Q_K :

$$\text{FPT} \equiv \frac{P_K(z^{-1})}{Q_K(z^{-1})}, \quad z^{-1} = e^{-i\omega\tau}. \quad (2.1)$$

Here, angular frequency ω is generally complex. The corresponding linear frequency ν is $\nu = \omega/(2\pi)$. As mentioned, τ is the constant sampling rate of the encoded time signal $\{c_n\}$ ($0 \leq n \leq N-1$) of full length N , corresponding to the total acquisition time $T = N\tau$. Further, n is the time signal number and digitized time t is counted as $n\tau$.

Explicitly, the numerator P_K and denominator Q_K polynomials are defined in terms of their expansion coefficients $\{p_r\}$ and $\{q_s\}$, respectively:

$$P_K(z^{-1}) = \sum_{r=0}^K p_r z^{-r}, \quad Q_K(z^{-1}) = \sum_{s=0}^K q_s z^{-s}. \quad (2.2)$$

The pair $\{p_r, q_s\}$ is extracted uniquely from the MacLaurin polynomial $S(\omega)$ whose expansion coefficients are the time signal points:

$$S(\omega) = \sum_{n=0}^{N-1} c_n z^{-n} \quad (\text{the finite } z - \text{transform}). \quad (2.3)$$

As usual in MRS, the time signal c_n itself is quantum-mechanically represented by a linear combination of complex damped exponentials (a geometric progression):

$$c_n = \sum_{k=1}^K d_k z_k^n, \quad z_k^{-1} = e^{-i\omega_k \tau}, \quad (2.4)$$

where K is the number of resonances assigned to the corresponding metabolites. The fundamental harmonics z_k^{-1} are the roots of the characteristic equation for the denominator polynomial, $Q_K(z_k^{-1}) = 0$. The complex fundamental frequencies $\{\omega_k\}$ are deduced from z_k^{-1} via $\omega_k = (i/\tau) \ln(z_k^{-1})$. The corresponding complex fundamental amplitudes $\{d_k\}$ are obtained by taking the Cauchy residue of the envelope $P_K(z^{-1})/Q_K(z^{-1})$ at $z^{-1} = z_k^{-1}$. The result is the analytical formula $d_k = P_K(z_k^{-1})/Q'_K(z_k^{-1})$, where $Q'_K(z^{-1})$ is the first derivative of $Q_K(z^{-1})$ with respect to the independent variable z^{-1} , i.e. $Q'_K(z^{-1}) = (d/dz^{-1})Q_K(z^{-1})$.

After reconstructing the characteristic frequencies and amplitudes $\{\omega_k, d_k\}$, the FPT as a parameter estimator provides the K component spectra $\text{FPT}_k = (P_K/Q_K)_k$. The assembly of such K components is denoted by $\text{FPT}_{\text{Comp}} = \{\text{FPT}_k\}$:

$$\text{FPT}_{\text{Comp}} = \{\text{FPT}_k\} \equiv \left\{ \left(\frac{P_K}{Q_K} \right)_k \right\}, \quad 1 \leq k \leq K, \quad (2.5)$$

where

$$\left(\frac{P_K}{Q_K} \right)_k = \frac{d_k z^{-1}}{z^{-1} - z_k^{-1}}. \quad (2.6)$$

The sum of the K components FPT_k gives the envelope in the form of the Heaviside partial fraction representation of the FPT used as a parameter estimator:

$$\text{FPT}_{\text{Tot}} = \frac{p_0}{q_0} + \sum_{k=1}^K \frac{d_k z^{-1}}{z^{-1} - z_k^{-1}}. \quad (2.7)$$

In the FPT as a shape estimator, only the envelope is reconstructed (no components). In the computations, the magnitude mode alone will presently be used. The notation $|\text{FPT}|_{\text{Tot}}$ will later be reserved for the nonparametric FPT only when compared

to the parametrically reconstructed $|\text{FPT}|_{\text{Comp}}$. However, when either nonderivative or derivative envelopes are compared with each other, the subscript 'Tot' becomes superfluous and, as such, will be omitted. i.e we shall write $|\text{FPT}|$.

Derivative estimations proceed by applying the derivative operator of the fixed m th order $D_m \equiv (d/d\nu)^m$ to the spectra from the parametric and nonparametric FPT [50–55]. This produces the derivative fast Padé transform (dFPT), for parameter and shape estimations. The explicit expressions for the derivative spectra are available from Refs. [54, 55] as either the analytical expressions (parametric dFPT) or the recursive formulae (nonparametric dFPT). Both representations are computationally attractive as they can be readily programmed into their expedient algorithms capable of generating literally hundreds of derivative spectra within only a few minutes.

Besides the FPT and dFPT, we shall also use the standard fast Fourier transform, FFT, and the derivative fast Fourier transform (dFFT). The working formulae of the FFT and dFFT are well-known. For completeness, the expression for the dFFT is also given in Refs. [54, 55]. In the present work, instructive comparisons will be made between the two pairs of the processors {FFT, dFFT} and {FPT, dFPT}. The Padé derivative spectra will also be presented in the magnitude mode. The envelopes will be labeled as $|D_m \text{FPT}|$ or $|D_m \text{FPT}|_{\text{Tot}}$, whereas the components will be denoted by $|D_m \text{FPT}|_k$ and $|D_m \text{FPT}|_{\text{Comp}}$. Similarly, the Fourier nonderivative and derivative envelopes will be referred to as $|\text{FFT}|$ and $|D_m \text{FFT}|$, respectively.

3 Results and discussion

3.1 Time signals encoded at a 3T clinical scanner

The time signals or FIDs to be processed in this work have been kindly provided to us by our colleagues from the Department of Obstetrics/Gynecology and Laboratory of Pediatrics/Neurology, University Medical Centre Nijmegen, the Netherlands. These time signals were encoded at a 3T Siemens clinical scanner from a 56 year-old patient. Proton MRS was applied with single-voxel point-resolved spectroscopy sequence (PRESS). The size of the voxel of interest (VOI) was $3 \text{ cm} \times 3 \text{ cm} \times 3 \text{ cm}$ with the location in the inferior cystic part of the tumor. Subsequent to in vivo MRS encoding, the ovarian tumor was surgically removed and histopathologic diagnosis was a borderline serous cystic ovarian tumor.

The standard procedure called WET (water suppression through enhanced T1 effects) was used to partially suppress the giant water resonance. The acquisition parameters were: the Larmor frequency $\nu_L = 127.732 \text{ MHz}$ for $B_0 = 3\text{T}$, the bandwidth $\text{BW} = 1200 \text{ Hz}$, the full length of each of the FIDs, $N = 1024$, the sampling time $\tau = 1/\text{BW} \approx 0.833 \text{ ms}$, the repetition time $\text{TR} = 2000 \text{ ms}$, the number of excitations $\text{NEX} = 64$ and the echo times $T = 30$ and 136 ms . Only the FID for $\text{TE} = 30 \text{ ms}$ will be used here. In order to improve SNR, these 64 encoded FIDs were averaged. More details on this problem are given in Ref. [39], where the Fourier spectral envelopes have been presented.

3.2 Plan of illustration of the input and output data: the concept of shape and parameter estimations

The averaged FID based on the just mentioned encodings from Ref. [39] is presently processed by means of the Fourier and Padé analyses, using the nonderivative and derivative estimations. No phase correction is applied to the FID. The reason is that all the spectra will be plotted in the phase-insensitive magnitude mode. In the case of Fourier processing, it is customary to use zero-filling of encoded time signals. Presently, we zero-fill the encoded FID once to extend the original length to 2048. In other words, the second half of this elongated time signal includes 1024 data points of zero amplitude. This zero-filled FID is used for both Fourier and Padé processings.

Altogether 8 figures will be reported. Of these, one figure is on the FFT and dFFT only (Fig. 1), whereas five figures (Figs. 2, 5–8) are on the FPT and dFPT alone. Moreover, two figures (Figs. 3, 4) compare the Fourier and Padé nonderivative and derivative envelopes. The finer details of the FPT and dFPT analyses are divided in two parts, each dealing with comparisons between the shape and parameter estimations (both nonderivative, derivative). One part refers to comparisons between the parametric and nonparametric envelopes (Figs. 5, 6). The other part compares the nonparametric envelopes with the parametrically reconstructed components (Figs. 7, 8).

This systematics of presentation of the results is necessary in order that the reader gains a fuller insight into the relative performance of the two analyzed processors through their customary (nonderivative FFT vs FPT) and new (derivative dFFT vs dFPT) representations. Regarding derivative estimations, Figs. 3–8 will testify that it is amply sufficient to consider the first three derivative spectra in the dFFT and dFPT. Moreover, the purpose of comparisons between the Padé shape and parameter estimations (both derivative) is to cross-validate the former on the qualitative and quantitative levels.

In general, envelopes (Fourier, Padé, ...) computed with FIDs encoded by *in vivo* MRS from patients, are densely packed with many overlapped resonances. This occurs in most (if not all) encodings, even those performed at longer TEs (e.g. 136, 272 ms), despite the occurrence that many short-lasting resonance profiles could be unidentifiable as they may have decayed to the level of nearly zero-valued background baseline. The spectral density is usually much higher at shorter values of TE (e.g. 30 ms) and this exacerbates the problem of spectral crowding. However, the benefit of data acquisition at shorter TEs is clinically important since the encoded FIDs are metabolically the most abundant. This occurs since a short TE may still be long enough to allow broader resonances (shorter lifetimes) to evade decays. It is for this reason that we presently opt to use the FIDs encoded in Ref. [39] at TE = 30 ms rather than at TE = 136 ms.

3.3 Time signal waveforms and Fourier magnitude envelopes (nonderivative, derivative)

Figure 1 shows the averaged time signal with the originally encoded 1024 FID data points and the Fourier total shape spectra or envelopes (nonderivative FFT, derivative dFFT). The first row is on the FID, whereas the Fourier envelopes are on the second-to-the-fifth rows. The intensities of the real $\text{Re}(\text{FID})$ and imaginary $\text{Im}(\text{FID})$ parts of the complex time signal from panels (a) and (b) are of the same strength as they are bounded in the intervals $(-0.05, 0.25)$ au and $(-0.25, 0.05)$ au, respectively. Both FID curves are seen to heavily oscillate around their abscissae at the signal numbers $n \geq 200$. This secures that all the resonances decayed even much before the end of the total duration $T = N\tau$ of the FID.

The waveforms of $\text{Re}(\text{FID})$ and $\text{Im}(\text{FID})$ are ondulatory because they reflect the dynamical oscillation of molecules in the examined system, which is here an in vivo encoded borderline serous cystic ovarian tumor. These oscillations are of a harmonic functional form, $\sin(2\pi n\tau f_k)$ and $\cos(2\pi n\tau f_k)$, or equivalently, $e^{2\pi i f_k n\tau}$. Here, $f_k = \text{Re}(\nu_k)$, where ν_k is the linear complex nodal frequency of the given k oscillation. This particular mode of oscillation is dictated by the nature of the external perturbation of the sample. The external perturbations in all NMR encodings are a static as well as a gradient magnetic field and radio-frequency pulses. All these three perturbations are from the spectrum of electromagnetic field, which itself contains the sinusoidal and cosinusoidal oscillatory modes.

If the simple oscillatory complex harmonics $e^{2\pi i f_k n\tau}$ were the only factor governing propagation of the FID at all times $n\tau \in [0, T]$, neither $\text{Re}(\text{FID})$ nor $\text{Im}(\text{FID})$ would ever decay to zero at the end of encodings as opposed to the situation seen on panels (a) and (b) of Fig. 1. Time evolution of all naturally occurring phenomena is limited. This means that after a certain lifetime, all transient phenomena shall die out. This effect is taken into account by allowing the unattenuated exponentials $e^{2\pi i f_k n\tau}$ to decay. To accommodate for such an effect, a damping frequency factor $\Gamma_k > 0$ is introduced (the inverse of which is the k th resonance lifetime) through an exponential probability $e^{-\Gamma_k n\tau}$.

The latter term multiplies the simple harmonic oscillatory mode $e^{2\pi i f_k n\tau}$ to induce decay of the FID to zero by way of the attenuated exponential $e^{2\pi i f_k n\tau - \Gamma_k n\tau}$. Now, the free induction decay curve or FID will indeed decay with the passage of time $t = n\tau$ because the general k th transient in the time signal is exponentially attenuated, as described by the corresponding damped complex harmonic $e^{i\omega_k \tau}$, where $\omega_k = 2\pi \nu_k$ and $\Gamma_k = \text{Im}(\omega_k)$. The sum of these K harmonics $e^{i\omega_k \tau}$, each multiplied by the corresponding complex amplitude d_k , will constitute the FID as written in Eq. (2.4). Therein, the physical meaning of both $\text{Re}(\text{FID})$ and $\text{Im}(\text{FID})$ is conveyed by their decays to zero as the time $n\tau$ approaches the end of the acquisition period $T = N\tau$. This is what is seen on panels (a) and (b) in Fig. 1.

The remaining four rows in Fig. 1 give the information in the forms of Fourier envelopes. The nonderivative envelope $|\text{FFT}|$ is on panel (c). This is followed by the derivative envelopes $|\text{D}_m \text{FFT}|$ of the increasing derivative order m : the first ($m = 1$, d), the second ($m = 2$, e) and the third ($m = 3$, f). Spectra on panels (c–f) refer to a wider frequency interval covering chemical shifts 0.35–4.65 ppm. This

Fig. 1 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30 \text{ ms}$ [39]. Abscissae in time signal numbers n and ordinates in arbitrary units (au). The real (a) and imaginary (b) parts of the complex averaged FID are on the top row. The Fourier envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the second-to-the-fifth row. Nonderivative FFT (c). Derivative dFFT (d–f) of increasing order m : (d, $m = 1$), (e, $m = 2$) and (f, $m = 3$). Spectral range of interest, SRI: 0.35–4.65 ppm, including the water resonance. For details, see the main text (color online)

band includes the location of the resonance frequency of the residual water peak. Presently, the water resonance is placed at 4.5 ppm to cohere with our previous study [38], which used the same encoded FIDs from Ref. [39]. It is observed on panel (c) that, despite a partial suppression of the originally giant water peak in the encoding process, the residual H_2O resonance is still very intense. More precisely, the residual water peak is still larger by a factor of 10 compared to the tallest resonance located near 2.06 ppm, assigned to nitrogen-acetyl neuraminic acid, acNeu.

Because of a large dynamic range of the spectral intensities on the ordinate for $|\text{FFT}|$ (c), due to dominance of the residual water peak, the resonances for other metabolites appear merely as a part of a noisy and bumpy background baseline. As such, the nonderivative envelope $|\text{FFT}|$ (c) is metabolically uninformative. Namely, downfield (below 4.5 ppm), in the remaining portion of the spectrum, within solely a few compound structures, one could try to pinpoint some crude nonquantifiable resonances, usually associated with a handful of metabolites (e.g. Lac near 1.33 ppm, NAA/acNeu within 2.0–2.07 ppm, etc.). The residual water peak on panel (c) is tall and broad in the nonderivative envelope $|\text{FFT}|$. The two lifted, shouldered bumps on each side of the water peak indicate the existence of a hidden structure in $|\text{FFT}|$ (c).

We now pass onto the Fourier derivative spectra using the dFFT (d–f). The first derivative Fourier envelope $|\text{D}_1\text{FFT}|$ is on panel (d). This spectrum shows that the first derivative operator D_1 succeeded in considerably suppressing the residual water peak height from about 15 au (c) to around 4 au (d). Moreover, the water resonance is fragmented in Fig. 1d. Its previously obscured left shoulder in $|\text{FFT}|$ (c), now becomes a separate narrower sub-peak in $|\text{D}_1\text{FFT}|$ (d).

Further, the reduced dynamic range on the ordinate axis in $|\text{D}_1\text{FFT}|$ (d) allows the emergence of a large number of structures at chemical shifts 0.35–4.4 ppm. However, these latter structures are anything but some recognizable peaks as they all resemble some random white noise, appearing like spikes of almost uniformly similar heights. In other words, $|\text{D}_1\text{FFT}|$ (d) does not depart notably further from $|\text{FFT}|$ (c) at the resonance frequency 0.35–4.4 ppm, where the main metabolic content is expected to reside. This occurs despite a large reduction of the residual water peak when passing from $|\text{FFT}|$ (c) to $|\text{D}_1\text{FFT}|$ (d).

Next to visit within the dFFT is the second derivative envelope $|\text{D}_2\text{FFT}|$ (e). Herein, there is a yet another reduction of the dynamic range on the ordinate between the residual water peak structures and the rest of the spectrum. This is the case because the second derivative operator D_2 further suppresses the remaining structures around the water peak (4.5 ppm), which is now even more fragmented. As a result of diminished disparity between the water peak and the rest of the spectrum (0.35–4.4 ppm), the former noise-like peaks in $|\text{D}_1\text{FFT}|$ (d) now appear more prominently in $|\text{D}_2\text{FFT}|$ (e). However, this comes in $|\text{D}_2\text{FFT}|$ (e) at the expense of smoothing out some of the finer peak sub-structures in $|\text{D}_1\text{FFT}|$ (d). The implication is that the D_2 operator leads to linewidth broadening in the dFFT. Therefore, the remaining, principal part of the spectrum with the main metabolites (0.35–4.4 ppm), does not offer any diagnostically useful information.

Finally, the third derivative envelope $|\text{D}_3\text{FFT}|$ is displayed on panel (f) of Fig. 1. It is seen that resolution in $|\text{D}_3\text{FFT}|$ (f) is worse than in $|\text{D}_2\text{FFT}|$ (e). This is most evident when comparing especially the peaks with some sub-structures. For instance,

either a partial or complete filling in the dips in between most of the adjacent peaks is observed to occur in $|D_3\text{FFT}|$ (f) relative to $|D_2\text{FFT}|$ (e). This means that the D_3 operator further aggravates the situation with respect to D_2 by yielding more linewidth broadenings.

Overall, if on panels (d–f), we choose some narrow frequency bands and scroll down from one row to another to revisit the spectral content within the selected chemical shifts, a systematic resolution degradation would be observed when passing from the first $|D_1\text{FFT}|$ (d) through the second $|D_2\text{FFT}|$ (e) to the third $|D_3\text{FFT}|$ (f) derivatives in the dFFT. This is a clear breakdown of the derivative fast Fourier transform, dFFT. The reason is all too well documented by now [50–55]. Namely, the dFFT is predominantly sampling noise with the increasing derivative order m in $|D_m\text{FFT}|$. The culprit is the multiplier $(n\tau)^m$ of each of the encoded time signal points c_n . The offending power function $(n\tau)^m$ in the product $(n\tau)^m c_n$, which is processed by the dFFT, puts more weight on the latter encoded (and, hence, noisy) time signal data points $\{c_n\}$ ($0 \leq n \leq N-1$). The factor $(n\tau)^m$, or more precisely $(-2\pi i n\tau)^m$, multiplying c_n comes from the application of the m th order derivative operator $D_m = (d/d\nu)^m$ to the frequency-dependent part $e^{-2\pi i \nu \tau}$ in the Fourier transformation [50–55].

3.4 Padé magnitude envelopes (nonderivative, derivative)

Figure 2 is of the same type as Fig. 1. However, Fig. 2 skips showing the same FID. Moreover, Fig. 2 deals with Padé nonderivative and derivative envelopes $|D_m\text{FPT}|$ ($0 \leq m \leq 3$), where $|D_0\text{FPT}| \equiv |\text{FPT}|$ due to $D_0 = 0$. The nonderivative envelope $|\text{FPT}|$ is on panel (a), below which are derivative spectra: $|D_1\text{FPT}|$ (b), $|D_2\text{FPT}|$ (c) and $|D_3\text{FPT}|$ (d). To aid discussion, some of the metabolite acronyms are indicated close to chemical shifts at which the corresponding resonances are expected to appear (not that they really do on panel a). Of course, this is self-evident for the residual water peak H_2O around 4.5 ppm, but hardly so for any of the other eight indicated metabolites: Lac quartet (q) near 4.1 ppm, Cho and PC compound near 3.2 ppm, Cr and PCr compound near 3.0 ppm, NAA and acNeu compound around 2.0 ppm as well as Lac doublet (d) close to 1.3 ppm. In particular, close attention should be paid to the recognized cancer biomarkers (Lac, Cho, PC).

Evidently, there is not much to say about $|\text{FPT}|$ (Fig. 2a) because this nonderivative Padé envelope is almost entirely similar to its Fourier counterpart $|\text{FFT}|$ (Fig. 1c). In other words, $|\text{FPT}|$ (Fig. 2a) lacks any useful metabolic information, similarly to its companion $|\text{FFT}|$ (Fig. 1c). As in Fig. 1c, one of the reason for not recognizing anything in Fig. 2a at chemical shifts 0.35–4.4 ppm is the presence of the dominant residual water peak which expands the dynamic range on the ordinate. This enlargement of the ordinate diminishes the chance of appearance of the considerably smaller peaks within the band 0.35–4.4 ppm of resonance frequencies.

The application of the derivative operator D_m to the Padé nonderivative envelope P_K/Q_K in the FPT leads to the derivative spectra $D_m P_K/Q_K$, abbreviated as $D_m\text{FPT}$. The results of such applications for $1 \leq m \leq 3$ are given on panels (b–d) of Fig. 2 where this time, however, the D_m operator appears to be a game changer.

Fig. 2 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30$ ms [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the nonparametric Padé-based signal processing. The resulting Padé envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Nonderivative FPT (a). Derivative dFPT (b–d) of increasing order m : (b, $m = 1$), (c, $m = 2$) and (d, $m = 3$). Spectral range of interest, SRI: 0.35–4.65 ppm, including the water resonance. For details, see the main text (color online)

Already the first derivative $|D_1\text{FPT}|$ (b) shows a dramatic improvement (albeit merely qualitative) relative to $|\text{FPT}|$ (a). Specifically, on panel (b), a large number of peaks pop out in a relatively reasonable delineated manner by leaving behind the background baseline. This occurs in spite of the presence of many overlapping resonances that prevent any attempt at quantification by way of e.g. integration. It is seen, in particular, that the resonances for cancer biomarkers (Lac, Cho, PC) begin to show up. For example, both Lac(d) and Lac(q) near 1.33 and 4.1 ppm clearly reveal their spectral multiplicity due to J-splittings.

On the other hand, Cho and PC appear as a total choline (tCho) around 3.20–3.22 ppm. Resonances assigned to Cr, PCr and NAA also start to uncover the underlying compositions. For instance, Cr and PCr near 3.0 ppm are partially split apart and so are NAA and acNeu around 2.0 ppm. Resonance fragmenting and linewidth narrowing of the residual water peak can partially be credited for a clearer emergence of the metabolite peaks away from 4.5 ppm, the location of H_2O . By comparison, we saw that a similar resonance fragmenting of the H_2O peak in $|D_1\text{FFT}|$ (Fig. 1c) was not accompanied by any improvement of the rest of the envelope (0.35–4.4 ppm).

The trend in $|D_1\text{FPT}|$ (b) of peak fragmenting and linewidth narrowing of the H_2O resonance is drastically enhanced on panel (c) for the second derivative $|D_2\text{FPT}|$. This happened to such a notable extent that the residual water peak simply ceases to dominate the spectrum in $|D_2\text{FPT}|$ (c). Moreover, the multiple components of the compound H_2O peak in $|D_1\text{FPT}|$ (b) are now highly resolved and located close to the background baseline in $|D_2\text{FPT}|$ (c). Such a striking localization of the formerly wide residual water peak in $|D_1\text{FPT}|$ (b) is able to sharply cut off the long tail of the H_2O resonance. The result is a low-lying background baseline away from the H_2O peak, i.e. at frequencies smaller than 4.5 ppm.

This allows a much better display of all the other individual resonances at chemical shifts 0.35–4.4 ppm. For example, both Lac(d) and Lac(q) around 1.33 and 4.1 ppm, respectively, are very well resolved close to the background baseline. Moreover, tCho starts showing its splitting to Cho and PC at 3.20 and 3.22 ppm, respectively. In comparison with $|D_1\text{FPT}|$ (b), it is seen in $|D_2\text{FPT}|$ (c) that the dip between Cr and PCr near 3.0 ppm has descended further down toward the background baseline. Also, the NAA+acNeu compound has undergone a further splitting such that NAA emerges as a single peak, whereas acNeu appears as a triplet. Nevertheless, despite much improvement in $|D_2\text{FPT}|$ (c) compared to $|D_1\text{FPT}|$ (b), the second derivative operator D_2 has left behind a number of overlapped resonances.

This situation requires the application of the third-order derivative operator D_3 to the complex envelope P_K/Q_K . By reference to the elementary chain rule of derivatives, the result $D_3P_K/Q_K \equiv D_3\text{FPT}$ is, of course, the same as that obtained by subjecting the second-derivative envelope $D_2\text{FPT}$ to the D_1 operator, $D_1(D_2\text{FPT}) = D_3\text{FPT}$. Such an elementary chain rule in derivatives is handy here as it can give an instructive guidance when passing from a lower- to a higher-order derivative spectra $D_m\text{FPT}$. This is best illustrated by comparing $|D_2\text{FPT}|$ (c) and $|D_3\text{FPT}|$ (d) in Fig. 2. Therein, as per the just stated chain rule, we see that it suffices to differentiate the complex spectrum $D_2\text{FPT}$ (whose magnitude is on panel c) only once more, by way of D_1 , to arrive at a substantially improved complex envelope $D_3\text{FPT}$, as clear from its magnitude mode on panel (d).

In Fig. 2d, throughout the entire frequency band 0.35–4.65 ppm, some 165 peaks appear as isolated resonances. All these numerous peaks are fully resolved down to the chemical shift axis. They are, therefore, amenable to easy quantification by a numerical quadrature since the integration limits can unequivocally be set up. Moreover, the background baseline on panel (d) is practically immersed into the chemical shift axis. Such a most favorable situation in $|D_3FPT|$ (d) is enabled partially by the dramatically diminished residual water compound peak. In fact, the H_2O compound structure appears now as a well-resolved multiplet. This means that the dFPT treats the water compound peak just like every other composite resonance by splitting it into its components. Such four components of the residual H_2O compound peak are clearly seen in a very narrow band 4.50–4.51 ppm.

Of course, the water resonance *per se* is of no diagnostic relevance for MRS. This, however, does not mean that the residual water peak is allowed to be treated casually. Conventionally, throughout the MRS literature, the residual water peak is arbitrarily diminished by subtracting some model components obtained either by fitting the Fourier envelopes with a few Lorentzians or Gaussians (or both) or by using the Hankel–Lanczos singular value decomposition (HLSVD) [56]. Both such *ad hoc* recipes would invariably alter the surrounding physical resonances.

To appreciate this possibility, it suffices to look e.g. at the two well-resolved peaks around 3.98 ppm, i.e. in the immediate vicinity of the water peak location 4.50 ppm in $|D_3FPT|$ (d). By the mentioned subtraction procedures, these latter two peaks could be deformed to such an extent that they would not be quantifiable, as opposed to the corresponding absolutely clear situation in $|D_3FPT|$ (d). Moreover, the same subtraction recipes would alter the other neighboring resonances around the residual water peak. No such deficiencies are present in the dFPT as patently evidenced in $|D_3FPT|$ (d).

The irony is that the water content in the scanned tissue is basically all that matters for magnetic resonance imaging (MRI). By contrast, the water content in the same tissue is a nuisance for MRS. This burden, however, ought to be handled cautiously, as explained. The moral of the story with the residual water peak particularly for in vivo MRS is that only when this remnant spectral structure (a leftover after water suppression by way of encoding) is treated properly as in Fig. 2, can we expect that the rest of the spectrum is clinically reliable for an adequate interpretation. Artificial handling of the residual water peak can change the other metabolite concentrations by an unknown amount. This could possibly lead to misclassification in metabolic profiling and, hence, to misdiagnoses.

Among the numerous isolated resonances in $|D_3FPT|$ (d), a few overlapped low-lying peaks are still present. However, they too are resolved in $|D_4FPT|$ (not shown to avoid clutter). Crucially, it is seen in $|D_3FPT|$ (d) that the resonances assigned to the recognized cancer biomarkers (Lac, Cho, PC) are eminently well resolved. For example, Lac(d) and Lac(q) near 1.33 and 4.1 ppm, respectively, exhibit their super-resolution in $|D_3FPT|$ (d). And so do Cho and PC around 3.20–3.22 ppm. Likewise, Cr and PCr as well as NAA and acNeu are also perfectly split apart. This conclusion holds true for all the 165 resolved quantifiable resonances in $|D_3FPT|$ (d).

Such a result is all the more remarkable given that the Padé reconstruction outcomes are due to the dFPT applied to the FID encoded at a clinical scanner of only

$B_0 = 3\text{ T}$ [39]. By comparison, as mentioned earlier, the Fourier spectral envelopes in the FFT for the FIDs encoded at a Bruker spectrometer of 600 MHz ($B_0 \approx 14.1\text{ T}$) [37] are much less resolved with only 12 quantifiable peaks for which the integration limits could be defined.

We repeatedly mentioned the exceptional diagnostic relevance of the recognized cancer biomarkers (Lac, Cho, PC). Figure 2d can give us a hint about the actual meaning of such a relevance. Therein, the envelope $|D_3\text{FPT}|$ (d) shows that the Cho and PC resonances are small and of the relatively comparable peak heights (that are themselves proportional to the metabolite concentrations). On the other hand, the Lac(d) resonance is much more intense than those of Cho and PC. The overall lactate concentration becomes augmented when the peak areas of Lac(q) at 4.1 ppm is added to that of Lac(d) at 1.33 ppm. What does this mean clinically for making a differential diagnosis among the three typical cases, borderline, benign and malignant ovarian lesions?

This question is very important because usually in cancerous ovarian tumor, the concentration levels of Lac and tCho are both expected to be elevated relative to normal ovary. However, according to $|D_3\text{FPT}|$ (Fig. 2d), while the Lac level is indeed elevated, the level of tCho appears to be quite low. This may be why the analyzed spectral data in Fig. 2d can be categorized as those associated with the borderline ovarian tumor in accord with the histopathologic findings [39]. Thus, the spectral analysis by the dFPT seems to be consistent with the gold standard of cancer diagnostics, histopathology.

3.5 Fourier versus Padé (nonderivative, derivative)

Our presentation and discussion associated with Figs. 1 and 2 covered the relevant aspects of the nonderivative and derivative branches of these two processors, both used as shape estimators. Nevertheless, it would be instructive to line up the pertinent Fourier and Padé panels directly on the same figure. Such a layout would give a more insightful angle of comparison of the selected frequency bands as they would be closely lying nearby, one above the other. This is deemed to be a better option for comparisons than placing Figs. 1 and 2 side-by-side. However, we would end up by having too many panels piled up on top of each other if we are to amalgamate Figs. 1 and 2 into a single graph.

Therefore, to avoid clutter in combining $|D_m\text{FFT}|$ and $|D_m\text{FPT}|$, we opt to give two mergers by way of Figs. 3 ($m = 0, 1$) and 4 ($m = 2, 3$). Moreover, since Figs. 1 and 2 give practically all the needed information about the residual water content, it would suffice to juxtapose Fourier and Padé envelopes in a narrower frequency window 0.35–4.25 ppm, which excludes the H_2O peak and its close surrounding.

Figure 3 shows that the nonderivative Fourier $|\text{FFT}|$ (a) and Padé $|\text{FPT}|$ (b) envelopes are not fundamentally different. Quite the contrary, they basically have highly similar features throughout the spectrum 0.35–4.25 ppm. In fact, the same remark holds true also for the wider window 0.35–4.65 in Figs. 1 and 2 with the water peak included. Nevertheless, a closer look would reveal that $|\text{FFT}|$ (a) has sharp wiggles

Fig. 3 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30$ ms [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the Fourier and nonparametric Padé nonderivative and derivative signal processings. The resulting Fourier and Padé envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Nonderivative envelopes: FFT (a), FPT (b). First derivative envelopes: dFFT (c), dFPT (d). Spectral range of interest, SRI: 0.35–4.25 ppm, outside the water resonance. For details, see the main text (color online)

on almost every spectral structure, as opposed to $|FPT|(b)$. These wiggles are artifacts in the Fourier envelope due to zero-filling of the FID. Although the same zero-filled FID is used by Padé processing, the envelope $|FPT|(b)$ is free from such wiggles. If these artifacts exist in a hidden way they are recognized in the FPT as spurious information and, as such, smoothed out or canceled.

A much more insightful comparison is provided by juxtaposing the first derivative envelopes, $|D_1FFT|(c)$ and $|D_1FPT|(d)$. This time on the level of derivative processing, there is a substantial difference between the dFFT (Fourier) and dFPT (Padé). Generally, resonances in $|D_1FFT|(c)$ are considerably wider than their counterparts in $|D_1FPT|(d)$. This linewidth broadenings in $|D_1FFT|(c)$ exacerbates the overlap problem in the dFFT. Another point of interest is to note that the background baseline in $|D_1FFT|(c)$ is more elevated above the chemical shift axis than in $|D_1FPT|(d)$. This is explained in part by revisiting Figs. 1 and 2. Therein, the residual water peak from Fig. 1d has a higher and farther extending tail than in the corresponding H_2O resonance from Fig. 2b.

Figure 4 compares $|D_mFFT|(c)$ (Fourier) and $|D_mFPT|(d)$ (Padé) for the derivatives of the second ($m = 2$: a, b) and the third ($m = 3$: c, d) orders. This figure amply illustrates the usefulness of placing the Fourier and Padé envelopes above each other on the same graph. It suffices to look at the levels of the background baselines to clearly see at once the marked differences. These baselines are highly elevated in $|D_{2,3}FFT|(a,c)$ and either very low in $|D_2FPT|(b)$ or embedded into the chemical shift axis in $|D_3FPT|(d)$. Linewidth broadening, previously noted when passing from the nonderivative $|FFT|(Fig. 3a)$ to the first derivative $|D_1FFT|(Fig. 3c)$ envelopes, is observed to further worsen in $|D_2FFT|(Fig. 4a)$ and $|D_3FFT|(Fig. 4c)$. This is all the more telling when the respective comparisons are made with the associated Padé counterparts $|D_2FPT|(Fig. 4b)$ and $|D_3FPT|(Fig. 4d)$.

In particular, for the narrower frequency window 0.35–4.25 ppm of Fig. 4, some 150 peaks are fully resolved in $|D_3FPT|(d)$ all the way down to the chemical shift axis. By comparison, in $|D_3FFT|(c)$, no peak is resolved down to the chemical shift axis. Moreover, even merely a few peaks that seem to be single, near-symmetric resonances (albeit a bit wider) are, in fact, misleading. This is evidenced by the dFPT for which such so-called 'singlets' appear as having their fully-resolved components in $|D_3FPT|(d)$.

In both cases $m = 2$ (a,b) and $m = 3$ (c,d) of Fig. 4, the contrast is so sharp between the dFFT and dFPT that one is under the impression that some two different problems were examined by Fourier and Padé processings. In reality, however, these two processors analyze the very same FID under identical conditions. Yet the outcomes are diametrically opposite between e.g. $|D_3FFT|(c)$ and $|D_3FPT|(d)$ that only the latter is seen to meet with success in matching the expectation of derivative estimations: simultaneous improvement of frequency resolution and signal-to-noise ratio, SNR. One need not go far to find the reason for this occurrence. The dFFT profoundly alters the encoded FID, but the dFPT does not.

The detrimental change of the FID in the dFFT is due to the derivative operator D_m itself whose action on the FFT envelope generates the apodized time signal $(-2\pi i n \tau)^m c_n$. It is the latter alteration of the originally encoded time signal $\{c_n\}$ ($1 \leq n \leq N - 1$) that simultaneously causes linewidth broadening and worsened

Fig. 4 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $TE = 30$ ms [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the Fourier and nonparametric Padé derivative signal processings. The resulting Fourier and Padé envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Second derivative envelopes: dFFT (a), dFPT (b). Third derivative envelopes: dFFT (c), dFPT (d). Spectral range of interest, SRI: 0.35–4.25 ppm, outside the water resonance. For details, see the main text (color online)

SNR. This happens because for positive integers m (derivative order), the power function $(-2\pi i n \tau)^m$ accentuates the noisy tail of the encoded FID.

In contradistinction, by processing the intact input data set $\{c_n\}$ ($1 \leq n \leq N - 1$), the dFPT introduces no apodization whatsoever nor any other modification of the encoded FID. Rather, the application of the D_m operator on the nonderivative envelope P_K/Q_K is an algebraic procedure with the exact analytic expression. In other words, the offending dFFT-owned power function $(-2\pi i n \tau)^m$ is now completely absent and, thus, its devastating effect on Padé spectra is nonexistent altogether in the dFPT. This is an amazing twist. It shows the power and diversity of algebraic signal processing in the realm of the all-encompassing Padé methodologies.

3.6 Padé nonderivative and derivative envelopes: shape versus parameter estimations

A nonderivative lineshape $|FPT|$ (Fig. 3b) from the nonparametric FPT is qualitative as, for a dense spectrum, it generally shows only the profile, i.e. the form of a dependence of the envelope as a function of the resonance frequency (chemical shift). That is why such processors are categorized as 'shape estimators'. Likewise, the corresponding nonderivative envelope from the parametric FPT is also qualitative. In other words, all the envelopes are qualitative irrespective of the variant of the nonderivative FPT by which they are reconstructed. The same holds true for any other shape estimator applied to encoded FIDs.

However, the important issue is to have some suitable, robust and reliable procedures for controlling the reconstruction output results. The first step in this direction is to compare the envelopes from shape and parameter nonderivative estimations by the FPT. This is done in Figs. 5 and 6. Similarly to Figs. 3 and 4, to avoid clutter, we subdivide the comparisons into two groups. One group (Fig. 5) contains the nonderivative and the first derivative envelopes. The other group (Fig. 6) is on the second and the third derivative envelopes.

The top two rows of Fig. 5 show the nonderivative envelopes $|FPT|$ (a: parametric, b: nonparametric). The bottom two rows display the first derivative envelopes $|D_1FPT|$ (c: parametric, d: nonparametric). Throughout the four rows, even a cursory look at the nonderivative spectra $|FPT|$ (a, b) would reveal a complete coincidence between the parametric and nonparametric estimations, respectively, counting even the tiniest spectral structures. This is checked by merging the lineshape from panel (a, parametric) to panel (b, nonparametric) in which case only a single, joint curve becomes visible.

The lineshape coincidence seen on panels (a) and (b) reassures the correctness of two different computational algorithms in the parameter and shape estimations by the nonderivative FPT. This is important from the numerical standpoint. However, despite this perfect accord, neither $|FPT|$ envelope, the parametric (a) nor the nonparametric (b), is of diagnostic relevance due to lack of useful metabolomic information.

Fig. 5 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30$ ms [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the Padé parametric and nonparametric (nondervative, derivative) signal processings. The resulting Padé envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Nondervative envelopes, FPT: parametric (a) and nonparametric (b). First derivative envelopes, dFPT: parametric (c) and nonparametric (d). Spectral range of interest, SRI: 0.35–4.25 ppm, outside the water resonance. For details, see the main text (color online)

For this reason, we are redirected to the first derivative envelopes $|D_1\text{FPT}|$ (c: parametric, d: nonparametric). Therein, perfect agreement is observed between the parametric (c) and nonparametric (d) envelopes of the first-order derivative ($m = 1$). Again, not a smallest spectral detail (be it a single or overlapped resonances) could be noted to differ by going from panel (c) to panel (d). For $|D_1\text{FPT}|$, this is self-evident by a visual inspection and was also verified to uphold when the red curve (c: parametric) is drawn on top of the blue curve (d: nonparametric).

The full coherence between panels (c) and (d) in the case of $|D_1\text{FPT}|$ is a confirmation of the correctness of the two distinct numerical programs for the parametric and nonparametric dFPT. Yet, as we know from Fig. 2, the first derivative envelope $|D_1\text{FPT}|$ itself is inconclusive, no matter how it is reconstructed, parametrically or nonparametrically. The reason is in the presence of the overlapped resonances and in the appearance of a still noticeable background baseline, despite vastly improved resolution and SNR when passing from the nonderivative $|\text{FPT}|$ (a,b) to the first derivative $|D_1\text{FPT}|$ (c,d) envelopes. Such a situation is a clear rationale for our resorting to the higher-order derivative estimations (specifically $m = 2$ and 3) by the parametric and nonparametric dFPT.

This is the topic of Fig. 6, which compares the parametric and nonparametric dFPT on the level of the second and third derivative envelopes. Herein, the envelopes $|D_m\text{FPT}|$ in the parametric version are on panels (a: $m = 2$, c: $m = 3$), whereas those in the nonparametric variant reside on panels (b: $m = 2$, d: $m = 3$). Nothing short of perfect accord can be seen by comparing either the envelopes $|D_2\text{FPT}|$ (a: parametric, b: nonparametric) on the one hand or the envelopes $|D_3\text{FPT}|$ (c: parametric, d: nonparametric) on the other hand. This refers to even the minuscule parts of the overall envelope lineshapes (be they on panels a, b or c, d) throughout the spectral region of interest, SRI: 0.35–4.25 ppm. Such an observation is valid both visually or through plotting the red and blue curves for $|D_2\text{FPT}|$ (a, b) on the same panel as well as for $|D_3\text{FPT}|$ (c, d) on another common panel.

This time too, a maximal coincidence of the nonparametric and parametric spectra $|D_m\text{FPT}|$ in Fig. 6 for the given derivative order, be it $m = 2$ (a, b) or $m = 3$ (c, d), builds confidence in both computer programs, one performing shape and the other parameter estimations of envelopes. Figure 6 stops at the third derivative order via $|D_3\text{FPT}|$ (c, d), but we explicitly checked for the presently studied problem that the nonparametric and parametric dFPT yield the coincident reconstructions of the envelopes $|D_m\text{FPT}|$ for much higher derivative orders ($5 \leq m \leq 50$). This finding corroborates the like observations in our earlier study on synthesized FIDs associated with breast cancer [52]. Therein, for a wide range of derivative orders ($1 \leq m \leq 50$), exact agreement has been recorded between the nonparametric and parametric dFPT for the envelopes $|D_m\text{FPT}|$.

On top of these important numerical cross-validations between the nonparametric and parametric dFPT for the derivative spectral envelopes, Fig. 6 offers firm reassurance about the diagnostic soundness, particularly of $|D_3\text{FPT}|$ (c, d). Such a reassurance is reflected in the quantification possibilities of some 150 isolated resonances in a relatively small frequency band 0.35–4.25 ppm. This is clinically significant given that many among these resonances in $|D_3\text{FPT}|$ (c, d) can unequivocally be assigned to the known metabolites [37]. It should be noted that often, resonances

Fig. 6 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30$ ms [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the derivative Padé parametric and nonparametric signal processings. The resulting Padé envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Second derivative envelopes, dFPT: parametric (a) and nonparametric (b). Third derivative envelopes: parametric (c) and nonparametric (d). Spectral range of interest, SRI: 0.35–4.25 ppm, outside the water resonance. For details, see the main text (color online)

(found in spectral envelopes) that could not be assigned to the known metabolites, have been denoted in the literature by letter “U” (unknown), followed by a number, e.g. U3 (1.56 ppm) or U14 (4.20 ppm), etc. [37].

The optimal extent of confluence between the nonparametric and parametric Padé estimations (be they nonderivative or derivative) is essential for an intrinsic benchmarking on the level of envelopes. This confluence, in fact, cross-validates the two Padé variants (nonparametric, parametric) against each other within the same signal processing strategy. Nevertheless, our primary focus is on the occurrence that the nonparametric dFPT emerged from these checkings of envelopes as the estimator of confirmed validity. The reason is that it is precisely this variant of the dFPT that is presently aimed to be subjected to a string of benchmarkings. Within such a goal, the parametric dFPT plays the role of a reference method, the gold standard in the realm of Padé-based estimations.

3.7 Nonderivative and derivative Padé: envelopes (shape estimations) versus components (parameter estimations)

However, the mentioned string of benchmarkings is still incomplete. This is the case because either envelope, nonparametric or parametric, could possibly miss some of the weakest resonances of diagnostic relevance without noticeable changes of the lineshape profiles. To rule out such a chance, yet another step of checking the nonparametric envelopes from the dFPT is necessary. This concluding chain in the internal cross-validation within the dFPT is demonstrated in Figs. 7 and 8. Therein, comparisons are made between the envelopes (shape estimations) and components (parameter estimations) using the FPT (nonderivative) and dFPT (derivative).

The finest verification step begins by the observation that in the nonderivative parametric FPT, an envelope itself does not explicitly reveal its internal structure. Its structure, however, is implicitly present. This occurs since a nonderivative parametric envelope is generated only after all the components have been reconstructed. The implication is that we can graph these components separately to visualize the lineshape profiles of all the individual resonances to be assigned to different metabolites. On the other hand, every such component lineshape (in the nonderivative parametric FPT) is built from the retrieved resonance parameters (peak position, width, height, phase). The given metabolite concentration is extracted from these fundamental parameters (specifically from the peak area). Thus, as is well-known, the nonderivative parametric FPT provides quantitative information (components) alongside the qualitative information (envelopes).

As with Figs. 5 and 6, to have more easy-to-follow presentations in Figs. 7 and 8, we again separate the two pairs of spectra. The nonderivative and the first derivative spectra are in Fig. 7. The second and the third derivative spectra are allocated to Fig. 8. The common window in Figs. 7 and 8 covers the frequencies ranging from 0.35 to 4.25 ppm. As noted earlier with Figs. 3–6, this latter frequency interval is outside the band around the residual water resonance.

As seen earlier in Fig. 2, the residual water peak around 4.5 ppm is wide implying that its tail extends far, lifting the background baseline on which all the remaining

Fig. 7 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30\text{ ms}$ [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the Padé parametric and nonparametric (nonderivative, derivative) signal processings. The resulting Padé components and envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Nonderivative FPT: parametric (a, components) and nonparametric (b, envelope). First derivative, dFPT: parametric (c, components) and nonparametric (d, envelope). Spectral range of interest, SRI: 0.35–4.25 ppm, outside the water resonance. For details, see the main text (color online)

metabolite resonances are superimposed. The nonderivative parametric FPT finds that the residual water peak has several components. None of them is of interest to MRS and, moreover, they would lead to clutter when graphed alongside the other resonances. For this reason, we shall not plot any of the tails of the individual line-shape profiles $|D_m \text{FPT}|_k$ (within the assembly of all the components $|D_m \text{FPT}|_{\text{Comp}}$) whose resonance frequencies are located outside the shown window 0.35–4.25 ppm.

The parametric FPT and dFPT for the reconstructed nonderivative components $|\text{FPT}|_{\text{Comp}}$ and their first derivatives $|D_1 \text{FPT}|_{\text{Comp}}$ are on panels (a) and (c) of Fig. 7, respectively. These components are respectively compared, in the same figure, with the nonparametric FPT and dFPT for the nonderivative envelope $|\text{FPT}|_{\text{Tot}}$ (b) and the pertinent derivative envelope of the first-order, $|D_1 \text{FPT}|_{\text{Tot}}$ (d). There is a huge number of components in $|\text{FPT}|_{\text{Comp}}$ (a), small, large, thin and wide alike. Herein, it is immediately apparent that the centers (resonance frequencies) of all the shown peaks are within the range 0.35–4.25 ppm. In other words, as described, the tails of the residual water peak components (as well as those due to all the other resonances centered at frequencies greater than 4.25 ppm), are not plotted in $|\text{FPT}|_{\text{Comp}}$ (a).

Even without such tails, panel (a) in Fig. 7 is of very high spectral density. On top of the myriad of sharp resonances, there are more than 15 broad peaks. Four of them are extremely wide and centered near 2.0, 2.4, 3.25 and 4.1 ppm. The other quite wide resonances are centered close to 0.4, 0.9, 1.02, 1.15, 1.3, 1.4, 1.65, 2.9, 3.01, 3.45, 3.65, 3.95 and 4.125 ppm. The tallest wide peaks have 0.4 and 1.3 ppm as their resonance frequencies. The broadest peak is centered near 2.4 ppm. These wide peaks contribute heavily to the background baseline when using them to construct the nonderivative envelope in the parametric FPT (Fig. 5a) alongside all the other components seen in Fig. 7a. It is observed in $|\text{FPT}|_{\text{Comp}}$ (Fig. 7a) that the exceedingly high spectral density inevitably yields tight overlaps almost at every tiny segment of the entire band 0.35–4.25 ppm. High spectral density is expected at a relatively short TE = 30 ms (used for the FID under study) at which most resonances have not decayed to zero.

It is handy to have the nonderivative nonparametric envelope $|\text{FPT}|_{\text{Tot}}$ (b) beneath $|\text{FPT}|_{\text{Comp}}$ (a). Such a panel configuration makes plainly in evidence why, how and where the most notable build-ups occur in the lifted background baseline in the total shape spectrum $|\text{FPT}|_{\text{Tot}}$ (b). The most pronounced hills in $|\text{FPT}|_{\text{Tot}}$ (b) occur around 0.4, 1.3, 1.65, 2.0, 2.5, 2.9, 3.2, 3.4 and 3.55–4.25 ppm in accordance with the locations of some of the mentioned broad peaks in $|\text{FPT}|_{\text{Comp}}$ (a).

In particular, above 3.55 ppm, the background baseline in $|\text{FPT}|_{\text{Tot}}$ (b) is the most intense and, in fact, it surpasses the level of the strongest metabolite peak heights, NAA+acNeu (2.0–2.07 ppm). Such a hugely elevated background baseline in $|\text{FPT}|_{\text{Tot}}$ (b) is partially caused by the interference of the wide resonances above 3.0 ppm seen in $|\text{FPT}|_{\text{Comp}}$ (a). The other participants in these interferences are the tails of the resonances centered above 4.25 ppm (including the water peak). These latter contributors to the background baseline in $|\text{FPT}|_{\text{Tot}}$ (b) are absent from $|\text{FPT}|_{\text{Comp}}$ (a) for the reason of avoiding clutter, as discussed.

The fact that there are so many components in $|\text{FPT}|_{\text{Comp}}$ (a) comes as no surprise for the employed model order $K = 550$ in $(P_K/Q_K)_k$ ($1 \leq k \leq K$). In other words, the nonderivative parametric FPT finds 550 resonances in the entire Nyquist range,

Fig. 8 In vivo ^1H MRS for a borderline serous cystic ovarian tumor. The time signals were encoded at a 3T clinical scanner using a short echo time, $\text{TE} = 30\text{ ms}$ [39]. The encoded averaged FID from panels (a) and (b) of Fig. 1 is subjected to the derivative Padé parametric and nonparametric signal processings. The resulting Padé components and envelopes (intensities on the ordinates in au and abscissae as chemical shifts in parts per million, ppm) are on the first-to-the-fourth row. Second derivative dFPT: parametric (a, components) and nonparametric (b, envelopes). Third derivative dFPT: parametric (c, components) and nonparametric (d, envelope). Spectral range of interest, SRI: 0.35–4.25 ppm, outside the water resonance. For details, see the main text (color online)

a part of which appears in $|\text{FPT}|_{\text{Comp}}$ (a). Among the reconstructed 550 resonances, there are stable and unstable resonances. Stable resonances are resilient to various perturbations (varying K , truncating the total signal length N , etc.). They are, therefore, classified as physical or genuine. The other resonances that are hyper-sensitive to the slightest changes in e.g. the model order K or in the truncation level of N are categorized as unphysical or spurious. The latter resonances are filtered out from the spectra by pole-zero cancellations [10].

Spurious resonances haphazardly wobble around in the complex plane of the harmonic variable z similarly to random noise. Yet, they are orderly chaotic. Their resonance frequencies pile up in an orderly fashion, one as the zero of P_K and the other as the root of Q_K in the Padé spectrum P_K/Q_K . This order in chaos is further refined by the occurrence that the spurious zeros and poles of the spectrum P_K/Q_K coincide. As a result of such pole-zero coincidences, the spectrum P_K/Q_K undergoes pole-zero cancellation. This is the essence of the denoising Froissart filter, DFF [10]. As a result of such a feature of self-correcting, the cleaned spectrum P_K/Q_K contains only physical resonances. The unphysical resonances are all but gone by pole-zero cancellation, which is the basis of a very useful concept (unique to the FPT) called signal-noise separation, SNS [8].

There is a huge event when only the first derivative operator $D_1 = d/dv$ is applied to the k th nonderivative complex component FPT_k , as reflected in the assembly FPT_{Comp} of all the components ($1 \leq k \leq K$) in Fig. 7c. The jump from $|\text{FPT}|_{\text{Comp}}$ (a) to $|D_1\text{FPT}|_{\text{Comp}}$ (c) could not be more dramatic, especially when it comes to broad resonances. The mentioned four widest resonances centered at 2.0, 2.4, 3.25 and 4.1 ppm in $|\text{FPT}|_{\text{Comp}}$ (a) disappeared altogether from $|D_1\text{FPT}|_{\text{Comp}}$ (c). Moreover, the next in the line of wide resonances, the two strongest peak in $|\text{FPT}|_{\text{Comp}}$ (a) centered at 0.4 and 1.3 ppm are hugely reduced in $|D_1\text{FPT}|_{\text{Comp}}$ (c). Further, the remaining wider resonances in $|\text{FPT}|_{\text{Comp}}$ (a) at 0.9, 1.02, 1.15, 1.4, 1.65, 2.9, 3.01, 3.45, 3.65, 3.95 and 4.125 ppm are also significantly diminished in $|D_1\text{FPT}|_{\text{Comp}}$ (c).

As a net result, $|D_1\text{FPT}|_{\text{Comp}}$ (c) is much less congested than $|\text{FPT}|_{\text{Comp}}$ (a). Of course, a veritable forest of sharp resonances survived the application of the D_1 operator. While the spectra $|\text{FPT}|_{\text{Comp}}$ (a) and $|\text{FPT}|_{\text{Tot}}$ (b) are vastly discrepant in every regard, it now becomes possible to trace a good number of the components from $|D_1\text{FPT}|_{\text{Comp}}$ (c) in the envelope $|D_1\text{FPT}|_{\text{Tot}}$ (d). Of course, this is only an approximate tracing since most resonances in $|D_1\text{FPT}|_{\text{Tot}}$ (d) are still tightly overlapped. Some of them are glued together in $|D_1\text{FPT}|_{\text{Tot}}$ (d) as opposed to the corresponding isolated adjacent resonances in $|D_1\text{FPT}|_{\text{Comp}}$ (c). Despite these substantial differences between panels (c) and (d), the progress made in $|D_1\text{FPT}|_{\text{Tot}}$ (d) relative to $|\text{FPT}|_{\text{Tot}}$ (b) is immense. We can now use the word 'progress' with certainty because a reasonably good outcome from comparison between panels (c) and (d) is a positive sign that indeed the passage from $|\text{FPT}|_{\text{Tot}}$ (b) to $|D_1\text{FPT}|_{\text{Tot}}$ (d) has the directionality of an improvement in Fig. 7.

To check the further status of this directionality, we visit Fig. 8 whose top and bottom two rows are for the second and the third derivatives, respectively. Herein, in the first two rows, comparisons are made between $|D_2\text{FPT}|_{\text{Comp}}$ (a) and $|D_2\text{FPT}|_{\text{Tot}}$ (b). The third and the fourth row juxtapose $|D_3\text{FPT}|_{\text{Comp}}$ (c) and $|D_3\text{FPT}|_{\text{Tot}}$ (d). It is seen that all the remainders of the wider resonances in $|D_1\text{FPT}|_{\text{Comp}}$ (Fig. 7c) vanished

from $|D_2FPT|_{Comp}$ (Fig. 8a). This led to sparser component spectra in $|D_2FPT|_{Comp}$ (Fig. 8a). Moreover, all the thin and sharp resonances from $|D_1FPT|_{Comp}$ (Fig. 7c) became thinner and sharper in $|D_2FPT|_{Comp}$ (Fig. 8a).

As to the spectrum $|D_2FPT|_{Tot}$ (b), substantive further progress is made judging upon the increased resemblance of this envelope with the corresponding component lineshapes from $|D_2FPT|_{Comp}$ (a). Many overlapped resonances in $|D_1FPT|_{Tot}$ (Fig. 7d) are split apart in $|D_2FPT|_{Tot}$ (Fig. 8b). This permits a clearer delineation of many adjacent peaks in $|D_2FPT|_{Tot}$ (Fig. 8b) that, in turn, can be better paired with the associated component resonances in $|D_2FPT|_{Comp}$ (Fig. 8a). However, there are still quite a few overlapped resonances in $|D_2FPT|_{Tot}$ (Fig. 8b) that preclude making a fuller one-to-one mapping on the second derivative level between the single envelope and the multiple component spectra. Simply stated, it is just not yet possible to identify one curve on panel (b: envelope) with a multitude of curves in panel (a: components) throughout the range 0.35–4.25 ppm.

This is why a further comparison is needed. To this end, the third derivatives are plotted on panels (c: components) and (d: envelope) in Fig. 8. Therein, the former multitude of curves for the component spectra now collapses basically into a single curve for $|D_3FPT|_{Comp}$ (c). This occurs because of a further line narrowing in $|D_3FPT|_{Comp}$ (c). Moreover, by reference to the component gold standard $|D_3FPT|_{Comp}$ (c), it is seen that a gigantic progress is made also in the envelope $|D_3FPT|_{Tot}$ (d). The outcome of this progress is coincidence between $|D_3FPT|_{Comp}$ (c) and $|D_3FPT|_{Tot}$ (d).

In other words, on the level of the third derivatives, a pure shape estimation by the nonparametric dFPT is able to predict all the component spectra from the parametric dFPT. It is then safe to conclude that the third derivative nonparametric dFPT successfully passed the most stringent benchmarking test: exact reproduction of the third derivative components reconstructed by the parametric dFPT. This confirms that it is possible to carry out quantification (finding the peak parameters) by exclusive reliance upon shape estimation in the dFPT. Such a task is accomplished without ever solving the quantification problem *per se* (i.e. no rooting of the characteristic polynomial Q_K , nor finding solutions of eigen-value problems, etc.).

3.8 Implications of the present findings: focus on ovarian tumor diagnostics

Ovarian cancer is one of the few malignancies for which the 5-year survival rate has remained low and unimproved over the last several decades [57]. Among all the gynecologic malignancies, ovarian cancer has the highest mortality rate, with the vast majority of women still diagnosed at the late stages for which the survival rates are below 30% [58–60]. Strikingly, if found early, ovarian cancer carries an excellent prognosis [61]. It is precisely because of the lack of accurate early detection methods that most ovarian cancers are diagnosed at advanced stages.

- *Ineffectiveness of standard Fourier-based in vivo MRS for ovarian cancer diagnostics*

The possibilities offered by MRS for ovarian cancer diagnostics have been appreciated for well over two decades [62]. However, using the FFT alongside fitting, this potential for in vivo MRS to provide effective early ovarian cancer detection, has remained unrealized. Relatively few studies have been published reporting results from in vivo MRS of the ovary.

As a small, moving organ, it is technically difficult to encode MRS time signals in vivo from the ovary, with problems of resolution and poor SNR being major hindrances [63]. The published in vivo MRS investigations aimed at distinguishing malignant from benign ovarian lesions are summarized in Ref. [38], which contains our meta-analysis of the results of 14 papers reported through 2015 on in vivo MRS. Surprisingly, as of mid-2021, only two more papers have been subsequently published [64, 65].

In all these studies using in vivo MRS, the time signals were encoded at clinical (1.5 or 3T) scanners, and were processed by the FFT. In certain instances, the FFT-reconstructed envelopes were post-processed by fitting in attempts to extract some of metabolite concentrations. Most of the studies used a TE of 130 ms or longer. This was dictated by the desire to have sparser envelope spectra that were presumably easier to fit with fewer Lorentzian or Gaussians (or both). Overall, however, only a few peaks were identified, with the metabolic information mainly qualitative (presence or absence of the peak). The statistically significant findings from our meta-analysis were that only two resonances, choline and lactate, were more often observed in the cancerous lesions. However, now including these two more recent studies [64, 65], choline was not detected in over 20% of the ovarian malignancies.

Our overall conclusion from the meta-analysis was that in vivo MRS with the FFT alongside fitting did not adequately distinguish malignant versus non-cancerous ovarian lesions [38]. The most recent studies [64, 65] include a larger number of borderline ovarian lesions, and provide some further insights for this important intermediate group. Taken together, regarding time signals encoded by in vivo MRS, more advanced processing by the derivative FPT becomes all the more necessary. The in vitro data, further justify this pursuit.

- *In vitro MRS offers greater insights into the metabolic features of ovarian cancer*

In vitro MRS with spectrometers of 500–600 MHz (11.7–14.1T) provides more possibilities for identifying cancerous ovarian lesions compared to in vivo MRS. Improved resolution is due to stronger static magnetic fields together with the methods of analytical chemistry on the excised specimens. From analyses using encoded in vitro MRS data, in one or more of the following Refs. [37, 62, 66–70], numerous resonances have been reported as being in higher concentrations in cancerous ovarian lesions compared to borderline or benign lesions.

Starting from the lowest chemical shift, these resonances include: Lip methylene [0.9, 1.3 ppm], leucine (Leu) [0.90–0.96, 1.71, 3.74 ppm], isoleucine (Iso) [1.02 ppm], valine (Val) [1.04 ppm], 3-hydroxybutyrate (3-HB) [1.23, 2.5–2.6 ppm], threonine (Thr) [1.33 ppm], Lac [1.41 ppm], fatty acid [1.47, 2.80 ppm], alanine (Ala) [1.51 ppm], lysine (Lys) [1.67–1.78, 2.80 ppm], polyamines [1.7 ppm], NAA [2.0–2.1 ppm], methionine (Met) [2.13 ppm], Gln [2.42–2.52 ppm], Cr [2.97 ppm],

Cho [3.17–3.20 ppm], taurine (Tau) [3.34 ppm], hypoxanthine [8.2 ppm]. On the other hand, higher concentrations of glucose (Glc) [5.22 ppm] and citrate (Cit) [2.56, 2.67 ppm] are reportedly characteristic of benign lesions. Elevations in 3-HB were considered to reflect rapid cellular metabolism in cancerous ovarian cysts, while increased levels of branched chain amino acids Iso, Leu, and Val were viewed as protein breakdown products [62].

The Warburg effect, i.e. increased glycolysis is considered to explain the increased levels of Ala and Lac, as well as the depletion of Glc in cancerous ovarian tissue [67]. In vitro analysis [39] indicated that the resonance at 2.03 ppm contained NAA and nitrogen-acetyl groups from glycoproteins and/or glycolipids. Applying Gas Chromatography-Mass Spectrometry (GC/MS) [70], NAA was subsequently found in high concentrations in serous cystadenocarcinomas, while serum NAA concentrations were generally low in the examined patients. Therefore, it appeared that NAA had been produced within the tumors. Other histological types of cystadenocarcinomas did not display high concentrations of NAA.

A subsequent publication [71] employing GC/MS together with liquid chromatography tandem mass spectrometry corroborates the significantly elevated NAA in primary epithelial ovarian cancer compared to normal ovary. In addition, nitrogen-acetyl-aspartyl-glutamate acid (NAAG), which is the next step in the pathway from NAA, catalyzed by NAA synthetase, was also found to be significantly elevated in ovarian cancer. In another usage of GC/MS, a comprehensive metabolic profiling of some 101 high-grade serous epithelial ovarian cancers compared to 15 normal ovaries, concluded that NAA was among the most notable changes, with 28-fold elevation compared to the normal ovaries [72].

It should be noted that only in Ref. [66], ^1H MRS with high-resolution magic angle spinning (HRMAS) was used for ovarian lesions. This method enables the most thorough analysis of the composition of intact specimens without requiring tissue extraction. In Ref. [66], NAA was found mainly in serous carcinomas. Among four patients with poor survival, higher levels of Val, Leu and Lys were detected. Moreover, in Ref. [67], Lys was also found to be of a significantly higher level in 10 malignant ovarian cyst fluid samples compared to 5 borderline and 8 benign samples analyzed by in vitro MRS. It was in Ref. [67], as well, that the Cit levels were recorded to be of a significantly higher level in benign samples. The depletion of Cit in cancerous ovarian cyst fluid was attributed to its utilization in fatty acid synthesis needed for rapid turnover of tumor cells.

- *MRS visible cancer biomarkers in ovarian carcinoma cell lines*

Further insights are provided by studies of ovarian cancer cell lines. Choline metabolism has been a particular focus, viewed as a relatively new metabolic hallmark of cancer [73]. This pathway is considered to reflect interrelations between oncogenic signaling and cellular metabolism. Starting from Cho, the first step in the Kennedy pathway, catalyzed by the enzyme ChoK-a, generates PC, which is an MRS-visible metabolite. Epithelial ovarian cancer cells, but not normal ovarian cells, reportedly have increased activity of ChoK-a and thereby yielded up to 8-fold higher levels of PC compared to normal or immortalized ovarian epithelial cells [73–75].

Notably, PC is a recognized biomarker for a number of cancers [76], possibly mediated by a loss of the tumor suppressor p53 function [77]. In a study of ovarian epithelial cells that were resistant to multiple chemotherapeutic agents, significant changes of several MRS-visible metabolites were reported [78]. These included increased levels of glutamate Glu, as well as glycerophosphocholine, GPC, and decreased glycine Gly, myo-inositol m-Ins, creatine Cr and leucine Leu. Glycerophosphocholine is of particular note, as a later step in the circular choline pathway, catalyzed by the enzyme lyso-phospholipase A1 from its precursor 1-Acyl-GPC. The enzyme glycerophosphodiester phosphodiesterase catalyzes the process by which GPC is converted to Cho. In a recent review [79], it has been suggested that GPC may also be a cancer biomarker. The authors emphasize the need for further studies using “innovative” MRS methodologies.

- *Clinical considerations*

Our studies [38, 80] were aimed at the clinical audience to convey the advantages of applying the FPT to MRS time signals encoded in vivo from the ovary. Therein, the term ‘FPT-MRS’ was coined to denote Padé-optimized in vivo MRS. We stressed the potential added diagnostic value provided by FPT-MRS for early ovarian cancer detection.

There were several chemical shift regions for which these insights were the most clinically relevant. Around 1.3 ppm, the component spectra helped elucidate the overlap among Lac, Lip, Thr and other resonances. Thereby, it may be possible to clarify whether or not the presence of Lip at 1.3 ppm can help identify cancerous lesions. In our meta-analysis [38], Lip at 1.3 ppm was more often reported in malignant ovary. However, this was not a statistically significant finding.

On the other hand, as noted, Lac at 1.3 ppm was found in our meta-analysis [38] to be significantly associated with malignant ovary, although the data regarding Lac were scant. Padé-based quantification using a short TE could help identify and quantify Lip, Lac as well as other overlapping resonances assigned to metabolites around 1.3 ppm. Uncertainties around 2.0 ppm have also arisen for MR spectra from the ovary. Through the FPT, the two resonances between 2.0 and 2.1 ppm corresponding to NAA and acNeu were clearly disentangled [38], and this may help determine the actual importance of NAA versus acNeu for distinguishing cancerous from borderline and benign ovarian lesions.

Presently, through the nonparametric dFPT, it was found that Cho and PC at 3.20 and 3.22 ppm, respectively, were clearly identified and quantified. This is the first time that PC has been identified and quantified by shape estimations alone applied to in vivo MRS time signals encoded from the ovary. Since PC is identified as a biomarker of malignant transformations [75, 76, 79], it now becomes feasible by shape estimations with the dFPT to noninvasively and clearly assess these components of the Kennedy pathway for ovarian cancer diagnostics.

In Ref. [80], we discussed the difficulties regarding MRS from the ovary. Firstly, encoding high-quality MRS time signals from this small, moving organ still persists as a major challenge. Secondly, the inevitably noisy encoded time signals have been heretofore almost exclusively analyzed and interpreted utilizing the FFT and

eventual post-processing by way of fitting. These difficulties have undoubtedly hampered efforts to explore *in vivo* MRS for early ovarian cancer detection. Overall, the MRS community has tended to avoid this problem area. As stated, since 2015 there have been only two further *in vivo* MRS studies of ovarian cancer [64, 65]. We emphasized that the results applying FPT-MRS to the ovary strongly indicate that 'this situation can and should change' [80] (p. 524).

There is now good reason to further employ dFPT-MRS to identify ovarian cancer and help distinguish it with greater certainty from benign ovarian lesions. The goal of effective *in vivo* MRS-based screening for ovarian cancer, the potential for which has been highlighted for over two decades [62], could at last become a reality through dFPT-MRS. Thereby, the long sought hope of radiologists and other diagnosticians within oncology would be realized. That goal is to visualize the entire clinical MRS information by seeing clearly disentangled overlapping peaks and inspecting the displayed concentrations of all diagnostically-relevant metabolites, including potential and recognized cancer biomarkers.

4 Conclusion

Derivative signal processing has several attractive and important aspects of data analyses and interpretation for *in vivo* magnetic resonance spectroscopy, MRS. The benefit becomes most pronounced when coupled with the nonparametric derivative fast Padé transform, dFPT. This processor, from the onset of the analysis of spectra, is only a shape estimator. Such estimation is qualitative because it provides only the functional form (lineshape profile) of the spectral intensities versus resonance frequencies (chemical shifts). However, quickly, within already low-orders (3 or 4) of derivatives, the shape estimation by the dFPT becomes quantitative via a jump from a nonparametric to a parametric signal processor. Crucially, it provides the key numerical values of the peak positions, widths, heights, areas and, hence, abundance of the resonating nuclei. With this at hand, concentrations of metabolites also become available, as the most relevant quantities for medical diagnostics by MRS.

This is achieved by a systematic linewidth narrowing and the background flattening. As a result, both resolution and signal-to-noise ratio, SNR, are improved with respect to the standard nonderivative fast Padé transform, FPT. This is in sharp contrast with the derivative fast Fourier transform, dFFT, which yields linewidth broadening and lowered SNR. Yet worse, the dFFT is inferior to the usual, nonderivative fast Fourier transform, FFT. Such a failure is due to multiplication of the processed time signal by a time-power function (the time variable raised to a power, which is the derivative order). This modification, induced by the derivative operator itself, accentuates the noisy part of the encoded time signal. Consequently, SNR is deteriorated and resolution of spectral lines is lowered in the dFFT. No such nor any other modification of the time signal exists in the dFPT since in this processor the derivative operator acts on the analytical expressions for the spectral lineshape profiles in the frequency domain.

The performance and benchmarking of the nonparametric dFPT are tested by comparisons with the corresponding parametric estimations in the dFFT. Already at

the level of the third derivative, the envelope in the nonparametric dFPT (shape estimator) becomes coincident with the components in the parametric dFPT (parameter estimator). This establishes the nonparametric dFPT as a processor capable of solving the quantification problem by shape estimations alone. The present illustrations provide the complete proof of this statement.

The specific theme under study here is *in vivo* MRS for ovarian tumor. The associated time signals were encoded at a 3T clinical scanner using a short echo time of 30 ms. This permits detection of many resonances that have short relaxation times. The nonparametric dFPT exactly reconstructs some 150 isolated resonances in a small frequency range 0.35–4.25 ppm. As such, they are all readily quantifiable. For example, the peak area of each of these resonances (and, thus, metabolite concentration) can be accurately determined by e.g. a standard numerical quadrature since the integration limits are unambiguous. This is all the more remarkable given that often the use of *in vitro* MRS at e.g. 14.1T (600 MHz) spectrometers yields spectral envelopes with elevated bumpy background baselines and many unresolved peaks whose integration boundaries are undefinable [81].

Total shape spectra or envelopes for *in vivo* MRS reconstructed by any standard nonderivative signal processor contain a vast amount of information through the simultaneous appearance of a large number of resonances that need to be assigned to the known metabolites in the tissue. This is exacerbated by the presence of broad resonances (due to macromolecules and the residual water content) that create a high and rolling background baseline, which obscures the actual concentration levels of the diagnostically relevant metabolites. Under such circumstances, it becomes practically impossible to make a visual inspection and interpretation of envelopes, a common phenomenon for *in vivo* MRS. It is precisely here, within this bottleneck of *in vivo* MRS, that the derivative shape estimation of spectra by the dFPT comes to the rescue by effectively lowering the background baseline *en route* and resolving all the overlapped resonances. This achievement puts the ubiquitous spectral crowding problem for *in vivo* MRS to rest for good.

This novel approach to data analyses and interpretation by means of the dFPT-based quantification-equipped shape estimations translates into a very practical tool of utmost usefulness for the physician. The reason is in the dual and simultaneous representation of all the reconstructed MRS data right on the screen. The displayed numerous isolated lineshape profiles and their peak areas help make the efficient and proper assignments to the metabolites physically present in the tissue scanned by *in vivo* MRS. It is deemed that this strategy would be of notable significance both to the patient and to the overall management of MRS diagnostic modalities in hospitals.

Acknowledgements The authors thank Radiumhemmet Research Funds, King Gustaf the Fifth Jubilee Fund at the Karolinska University Hospital, the Stockholm County Council (FoUU) and the Marsha Rivkin Center for Ovarian Cancer Research (Seattle, USA). We would like to thank our colleagues, Professors Marinette van der Graaf, Leon Massuger, Ron Wevers, Eva Kolwijck, Udo Engelke, Arend

Heerschap, Henk Blom, M'Hamed Hadfoune, and Wim A. Buurman from Radboud University for kindly making the in vivo encoded FIDs available to us.

Funding Open access funding provided by Karolinska Institute.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

1. I.I. Rabi, J.R. Zacharias, S. Millman, P. Kusch, A new method of measuring nuclear magnetic moment. *Phys. Rev.* **53**, 318 (1938)
2. I.I. Rabi, S. Millman, P. Kusch, J.R. Zacharias, The molecular beam resonance method for measuring nuclear magnetic moments : the magnetic moments of $^3\text{Li}^6$, $^3\text{Li}^7$ and $^9\text{F}^{19}$. *Phys. Rev.* **55**, 526–535 (1939)
3. F. Bloch, W.W. Hansen, M. Packard, Nuclear induction. *Phys. Rev.* **69**, 127 (1946)
4. F. Bloch, Nuclear induction. *Phys. Rev.* **70**, 460–474 (1946)
5. F. Bloch, W.W. Hansen, M. Packard, The nuclear induction experiment. *Phys. Rev.* **70**, 474–485 (1946)
6. E.M. Purcell, H.C. Torrey, R.V. Pound, Resonance absorption by nuclear moments in a solid. *Phys. Rev.* **69**, 37–38 (1946)
7. E.M. Purcell, N. Bloembergen, R.V. Pound, Resonance absorption by nuclear magnetic moments in a single crystal of CaF_2 . *Phys. Rev.* **69**, 988 (1946)
8. Dž. Belkić, K. Belkić, The general concept of signal-noise separation (SNS): mathematical aspects and implementation in magnetic resonance spectroscopy. *J. Math. Chem.* **45**, 563–597 (2009)
9. M. Froissart, Approximation de Padé: Application à la physique des particules élémentaires, in *Prépublications de la Recherche Coopérative sur Programme n° 25*, L 8470, vol. 9, ed. by J. Carmona, M. Froissart, D.W. Robinson, D. Ruelle (Centre National de la Recherche Scientifique (CNRS), Strasbourg, France, 1969), pp. 1–13
10. Dž. Belkić, Exact signal-noise separation by Froissart doublets in fast Padé transform for magnetic resonance spectroscopy. *Adv. Quantum Chem.* **56**, 95–179 (2009)
11. R. de Prony, Essai expérimental et analytique: sur les lois de la dilatabilité de fluides élastique et sur celles de la force expansive de la vapeur de l'eau et de la vapeur de l'alcool, à différentes températures. *J. de l'École Polytechn. Paris* **1**, 24–76 (1795)
12. H. Padé, Sur la représentation approchée d'une fonction par des fractions rationnelles (Ph.D. Thesis), *Ann. Fac. Sci. de l'École Norm. Sup.* **9**, Suppl. 1–93 (1892)
13. L. Weiss, R.N. McDonough, Prony's method, Z-transforms and Padé approximations. *SIAM Rev.* **5**, 145–149 (1963)
14. J.C. Wheeler, R.G. Gordon, *The Padé Approximant in Theoretical Physics* (Academic Press, New York, 1970)
15. G.A. Baker, *Essentials of the Padé Approximants* (Academic Press, New York, 1975)

16. A. Sidi, *Practical Extrapolation Methods: Theory and Applications* (Cambridge University Press, Cambridge, 2003)
17. Dž. Belkić, *Quantum-Mechanical Signal Processing and Spectral Analysis* (Taylor & Francis Publishing, London, 2005)
18. K. Belkić, *Molecular Imaging through Magnetic Resonance for Clinical Oncology* (Cambridge International Science Publishing, Cambridge, 2004)
19. Dž. Belkić, K. Belkić, *Signal Processing in Magnetic Resonance Spectroscopy with Biomedical Applications* (Taylor & Francis Publishing, London, 2010)
20. Dž. Belkić, Fast Padé Transform (FPT) for magnetic resonance imaging and computerized tomography. *Nucl. Instrum. Methods Phys. Res. A* **471**, 165–169 (2001)
21. Dž. Belkić, Non-Fourier based reconstruction techniques. *Magn. Res. Mater. Phys. Biol. Med. (MAGMA)* **15** (Suppl. 1), 36–37 (2002)
22. R.J. McEliece, J.B. Shearer, A property of Euclid's algorithm and an application to Padé approximation. *SIAM J. Appl. Math.* **34**, 611–615 (1978)
23. R.P. Brent, F.G. Gustavson, D.Y.Y. Yun, Fast solution of Toeplitz systems of equations and computation of Padé approximant. *J. Algorithms* **1**, 259–295 (1980)
24. D. Shanks, An analogy between transients and mathematical sequences and some nonlinear sequence-to-sequence transforms suggested by it. Part I (NOLM 9994, Naval Ordnance Laboratory Memorandum, Project # NOL-4-Re9d-21-2, White Oak, MD, July 26, 1949), 42 pp (1949)
25. D. Shanks, Nonlinear transformation of divergent and slowly convergent sequences. *J. Math. and Phys.* **34**, 1–42 (1955)
26. P. Wynn, On a device for computing the $e_m(S_n)$ transformation. *Math. Tabl. Aids. Comp. (MTAC)* **10**, 91–96 (1956)
27. P. Wynn, On a procrustean technique for the numerical transformation of slowly convergent sequences and series. *Proc. Camb. Philos. Soc.* **52**, 663–671 (1956)
28. Dž. Belkić, New hybrid non-linear transformations of divergent perturbation series for quadratic Zeeman effects. *J. Phys. A: Math. Gen.* **22**, 3003–3010 (1989)
29. E.J. Weniger, Nonlinear sequence transformations for the acceleration of convergence and the summation of series. *Comput. Phys. Rep.* **10**, 189–371 (1989)
30. Dž. Belkić, Critical validity assessment of theoretical models: charge exchange at intermediate and high energies. *Nucl. Instrum. Methods Phys. Res. B* **154**, 220–246 (1999)
31. Dž. Belkić, K. Belkić, In vivo derivative NMR spectroscopy for simultaneous improvements of resolution and signal-to-noise-ratio: Case study, Glioma. *J. Math. Chem.* **59**, 46pp (2021). <https://doi.org/10.1007/s10910-021-01280-0>
32. K. Belkić, Resolution performance of the fast Padé transform: potential advantages for magnetic resonance spectroscopy in ovarian cancer diagnostics. *Nucl. Instrum. Methods Phys. Res. A.* **580**, 874–880 (2007)
33. Dž. Belkić, K. Belkić, Mathematical modeling applied to an NMR problem in ovarian cancer detection. *J. Math. Chem.* **43**, 395–425 (2008)
34. Dž. Belkić, K. Belkić, Magnetic resonance spectroscopy with high-resolution and exact quantification in the presence of noise for improving ovarian cancer detection. *J. Math. Chem.* **50**, 2558–2576 (2012)
35. Dž. Belkić, K. Belkić, Resolution enhancement as a key step towards clinical implementation of Padé-optimized magnetic resonance spectroscopy for diagnostic oncology. *J. Math. Chem.* **51**, 2608–2637 (2013)
36. Dž. Belkić, K. Belkić, How the fast Padé transform handles noise for MRS data from the ovary: importance for ovarian cancer diagnostics. *J. Math. Chem.* **54**, 149–185 (2016)
37. E.A. Boss, S.H. Moolenaar, L.F. Massuger, H. Boonstra, U.F. Engelke, J.G. de Jong, R.A. Wevers, High-resolution proton nuclear magnetic resonance spectroscopy of ovarian cyst fluid. *NMR Biomed.* **13**, 297–305 (2000)
38. Dž. Belkić, K. Belkić, In vivo magnetic resonance spectroscopy for ovarian cancer diagnostics: quantification by the fast Padé transform. *J. Math. Chem.* **55**, 349–405 (2017)
39. E. Kolwijck, U.F. Engelke, M. van der Graaf, A. Heerschap, J. Henk, H.J. Blom, M. Hadfoune, W.A. Buurman, L.F. Massuger, R.A. Wevers, N-acetyl resonances in in vivo and in vitro NMR spectroscopy of cystic ovarian tumors. *NMR Biomed.* **22**, 1093–1099 (2009)
40. Dž. Belkić, K. Belkić, Iterative averaging of spectra as a powerful way of suppressing spurious resonances in signal processing. *J. Math. Chem.* **55**, 304–348 (2017)

41. Dž. Belkić, K. Belkić, Synergism of spectra averaging and extrapolation for quantification of in vivo MRS time signals encoded from the ovary. *J. Math. Chem.* **55**, 1067–1109 (2017)
42. Dž. Belkić, K. Belkić, Encoded in vivo time signals from the ovary in magnetic resonance spectroscopy: poles and zeros as the cornerstone for stability of response functions of systems to external perturbations. *J. Math. Chem.* **55**, 1110–1157 (2017)
43. R. Zurmühl, *Praktische Mathematik* (Springer, Berlin, 1953)
44. C. Lanczos, *Applied Analysis* (Prentice Hall, New York, 1956)
45. F.B. Hildebrand, *Introduction to Numerical Analysis* (McGraw Hill, New York, 1956)
46. E. Bertholdi, R.R. Ernst, Fourier spectroscopy and the causality principle. *J. Magn. Reson.* **11**, 9–19 (1973)
47. L. Vanhamme, T. Sundin, P. van Hecke, S. van Huffel, MR spectroscopy quantitation: a review of time-domain methods. *NMR Biomed.* **14**, 233–246 (2001)
48. Š. Miwerisová, M. Ala-Korpela, MR spectroscopy quantitation: a review of frequency-domain methods. *NMR Biomed.* **14**, 247–259 (2001)
49. S.W. Provencher, Automatic quantitation of localized in vivo ^1H spectra with LCMODEL. *NMR Biomed.* **14**, 260–264 (2001)
50. Dž. Belkić, K. Belkić, Exact quantification by the nonparametric fast Padé transform using only shape estimation of high-order derivatives of envelopes. *J. Math. Chem.* **56**, 268–314 (2018)
51. Dž. Belkić, K. Belkić, Explicit extraction of absorption peak positions, widths and heights using higher order derivatives of total shape spectra by nonparametric processing of time signals as complex damped multi-exponentials. *J. Math. Chem.* **56**, 932–977 (2018)
52. Dž. Belkić, K. Belkić, Validation of reconstructed component spectra from non-parametric derivative envelopes: comparison with component lineshapes from parametric derivative estimations with the solved quantification problem. *J. Math. Chem.* **56**, 2537–2578 (2018)
53. Dž. Belkić, K. Belkić, Derivative NMR spectroscopy for J-coupled multiplet resonances using short time signals (0.5KB) encoded at low magnetic field strengths (1.5T). Part I: Water suppressed. *J. Math. Chem.* **59**, 364–404 (2021)
54. Dž. Belkić, K. Belkić, Derivative NMR spectroscopy for J-coupled multiplet resonances using short time signals (0.5KB) encoded at low magnetic field strengths (1.5T). Part II: water Unsuppressed. *J. Math. Chem.* **59**, 405–443 (2021)
55. Dž. Belkić, K. Belkić, Derivative NMR spectroscopy for J-coupled resonances in analytical chemistry and medical diagnostics. *Adv. Quantum Chem.* **84**, 171 pp (2021). <https://doi.org/10.1016/bs.aiq.2021.05.001>
56. W.W.F. Pijnappel, A. van den Boogaart, R. de Beer, D. van Ormondt, SVD-based quantification of magnetic resonance signals. *J. Magn. Reson.* **97**, 122–134 (1992)
57. C.V. Trinidad, A.L. Tetlow, L.E. Bantis, A.K. Godwin, Reducing ovarian cancer mortality through early detection: approaches using circulating biomarkers. *Cancer. Prev. Res.* **13**, 241–252 (2020)
58. T. Berg, T.J. Nøttrup, U.B.S. Peen, H. Roed, Treatment and outcomes of a Danish ovarian cancer population. *Dan. Med. J.* **67**, A06190346 (2020)
59. B.R. Carr, M. Moroney, J. Sheeder, S.G. Eckhardt, B. Sawyer, K. Behbakht, J.R. Diamond, Survival and clinical outcomes of patients with ovarian cancer who were treated on phase 1 clinical trials. *Cancer* **126**, 4289–4293 (2020)
60. S. Petersen, P. Shahiri, A. Jewell, L. Spoozak, J. Chapman, S. Fitzgerald-Wolff, S. Min Lai, D. Khabele, Disparities in ovarian cancer survival at the only NCI-designated cancer center in Kansas. *Am. J. Surg.* **221**, 712–717 (2021)
61. G. Chornokur, E. Armankwah, J. Schildkraut, C. Phelan, Global ovarian cancer health disparities. *Gynecol. Oncol.* **129**, 258–264 (2013)
62. L.F.A.G. Massuger, P.B.J. van Vierzen, U. Engelke, A. Heerschap, R. Wevers, ^1H magnetic resonance spectroscopy. A new technique to discriminate benign from malignant ovarian tumors. *Cancer* **82**, 1726–1730 (1998)
63. A. Esseridou, G. Di Leo, L.M. Sconfienza, V. Caldiera, F. Raspagliesi, B. Grijuela, F. Hanozet, F. Podo, F. Sardanelli, In vivo detection of choline in ovarian tumors using 3D MRS. *Investig. Radiol.* **46**, 377–382 (2011)
64. F.H. Ma, Y.A. Li, J. Liu, H.M. Li, G.F. Zhang, J.W. Qiang, Role of proton MR spectroscopy in the differentiation of borderline from malignant epithelial ovarian tumors: a preliminary study. *J. Magn. Reson. Imaging* **49**, 1684–1693 (2019)
65. S.M. Mansour, M.M.M. Gomma, P.N. Shafik, Proton MR spectroscopy and the detection of malignancy in ovarian masses. *Br. J. Radiol.* **92**, 20190134 (2019)

66. D. Ben Sellem, K. Elbayed, A. Neuville, F.-M. Moussallieh, G. Lang-Averous, M. Piotto, J.-P. Bellocq, I.J. Namer, Metabolomic characterization of ovarian epithelial carcinomas by HRMAS-NMR spectroscopy. *J. Oncology* **2011**, 174019 (2011)
67. M. Kyriakides, N. Rama, J. Sidhu, H. Gabra, H.C. Keun, M. El-Bahrawy, Metabonomic analysis of ovarian tumour cyst fluid by proton nuclear magnetic resonance spectroscopy. *Oncotarget* **7**, 7216–7226 (2016)
68. I.C. Smith, D.E. Blandford, Diagnosis of cancer in humans by ¹H NMR of tissue biopsies. *Biochem. Cell. Biol.* **76**, 472–476 (1998)
69. J.C. Wallace, G.P. Raaphorst, R.L. Somorjai, C.E. Ng, M. Fung Kee Fung, M. Senterman, I.C. Smith, Classification of ¹H MR spectra of biopsies from untreated and recurrent ovarian cancer using linear discriminant analysis. *Magn. Reson. Med.* **38**, 569–576 (1997)
70. E. Kolwijck, R.A. Wevers, U.F. Engelke, J. Woudenberg, J. Bulten, H.J. Blom, L.F.A.G. Massuger, Ovarian cyst fluid of serous ovarian tumors contains large quantities of the brain amino acid N-acetylaspartate. *PLoS ONE* **5**, e10293 (2010)
71. M.Y. Fong, J. McDunn, S.S. Kakar, Identification of metabolites in the normal ovary and their transformation in primary and metastatic ovarian cancer. *PLoS ONE* **6**, e19963 (2011)
72. B. Zand, Altered ovarian cancer metabolism increases neuronal N-acetylaspartate to promote tumor growth. M.Sc. thesis, UT GSBS (University of Texas, Graduate School of Biomedical Sciences), Dissertations Theses (Open Access), n° 378 (2013)
73. M. Bagnoli, A. Granata, R. Nicoletti, B. Krishnamachary, Z.M. Bhujwalla, R. Canese, F. Podo, S. Canevari, E. Iorio, D. Mezzanzanica, Choline metabolism alteration: a focus on ovarian cancer. *Front. Oncol.* **6**, 153 (2016)
74. E. Iorio, D. Mezzanzanica, P. Alberti, F. Spadaro, C. Ramoni, S. D'Ascenzo, D. Millimaggi, A. Pavan, V. Dolo, S. Canavari, F. Podo, Alterations of choline phospholipid metabolism in ovarian tumor progression. *Cancer Res.* **65**, 9369–9376 (2005)
75. E. Iorio, A. Ricci, M.E. Pisanu, M. Bagnoli, F. Podo, S. Canevari, Choline metabolic profiling by magnetic resonance spectroscopy, in *Ovarian Cancer: Methods and Protocols, Methods in Molecular Biology*, ed. by A. Malek, O. Tchernitsa (Springer, New York, 2013), pp. 255–270
76. K. Glunde, J. Jiang, S.A. Moestue, I.S. Gribbestad, MRS/MRSI guidance in molecular medicine: targeting choline and glucose metabolism. *NMR Biomed.* **24**, 673–690 (2011)
77. N. Mori, R. Delsite, K. Natarajan, M. Kulawiec, Z. Bhujwalla, K. Singh, Loss of p53 function in colon cancer cells results in increased phosphocholine and total choline. *Molec. Imaging* **3**, 319–323 (2004)
78. M. Engskog, M. Björklund, J. Haglöf, T. Arvidsson, M. Shoshan, C. Pettersson, Metabolic profiling of epithelial ovarian cancer cell lines: evaluation of harvesting protocols for profiling using NMR spectroscopy. *Bioanalysis* **7**, 157–166 (2015)
79. K. Sonkar, V. Ayyappan, C. Tressler, O. Adelaja, R. Cai, M. Cheng, K. Glunde, Focus on the glycerophosphocholine pathway in choline phospholipid metabolism of cancer. *NMR Biomed.* **32**, e4112 (2019)
80. K. Belkić, Dž. Belkić, The challenge of ovarian cancer: steps towards early detection through advanced signal processing in MRS. *Isr. Med. Assoc. J.* **19**, 517–525 (2017)
81. B. Sitter, T. Bathen, B. Hagen, C. Arentz, F.E. Skjeldestad, I.S. Gribbestad, Cervical cancer tissue characterized by high-resolution magic angle spinning MR spectroscopy. *Magn. Res. Mater. Phys. Biol. Med. (MAGMA)* **16**, 174–181 (2004)

Authors and Affiliations

Dževad Belkić^{1,2} · Karen Belkić^{1,2,3,4}

✉ Dževad Belkić
Dzevad.Belkic@ki.se

¹ Department of Oncology and Pathology, Karolinska Institute, P.O. Box 260, 171 76 Stockholm, Sweden

² Department of Medical Radiation Physics and Nuclear Medicine, Karolinska University Hospital, P.O. Box 260, 171 76 Stockholm, Sweden

³ School of Community and Global Health, Claremont Graduate University, Claremont, CA, USA

⁴ Institute for Health Promotion and Disease Prevention Research, University of Southern California School of Medicine, Los Angeles, CA, USA