Magnetic Resonance Spectroscopy: Principles and Techniques: Lessons for Clinicians

Joshua M. Tognarelli [*], Mahvish Dawood [*], Mohamed I.F. Shariff [*], Vijay P.B. Grover [*], Mary M.E. Crossey [*], I. Jane Cox [†], Simon D. Taylor-Robinson [*], Mark J.W. McPhail [*]

[*]Liver Unit, Division of Diabetes, Endocrinology and Metabolism, Department of Medicine, Imperial College London, London, United Kingdom and [†]The Foundation for Liver Research, Institute of Hepatology, 69-75 Chenies Mews, London WC1E 6HX, United Kingdom

Magnetic resonance spectroscopy (MRS) provides a non-invasive 'window' on biochemical processes within the body. Its use is no longer restricted to the field of research, with applications in clinical practice increasingly common. MRS can be conducted at high magnetic field strengths (typically 11–14 T) on body fluids, cell extracts and tissue samples, with new developments in whole-body magnetic resonance imaging (MRI) allowing clinical MRS at the end of a standard MRI examination, obtaining functional information in addition to anatomical information. We discuss the background physics the busy clinician needs to know before considering using the technique as an investigative tool. Some potential applications of hepatic and cerebral MRS in chronic liver disease are also discussed. (J Clin Exp Hepatol 2015;5:320–328)

The biomedical applications of nuclear magnetic resonance (NMR) are twofold: magnetic resonance imaging (MRI) and magnetic resonance spectroscopy (MRS). The applications of MRS as a research tool are extremely diverse, encompassing studies on isolated cells, body fluids and perfused organs at high magnetic field strengths in an experimental, laboratory-based setting and also *in vivo* studies using clinical MR systems.[1] *In vivo* clinical MRS on whole-body MRI scanners has been used to study the metabolism of well-defined regions of the human body, affording a non-invasive 'metabolic window' on a wide range of biochemical processes in the body, including the composition and function of human organs *in vivo*.[2] Clinical MRS developments have exploited many of the advances in MRI at the magnetic field strengths now used (typically 1.5–3.0 T) and the use of magnetic field gradients. The sensitivity and spatial resolution of MRS is a limiting factor *in vivo*, but parallel utilisation of *in vitro* MR spectroscopy of tissue extracts, body fluids and cell lines at much higher magnetic field strengths (typically 11.7–14.1 T) allows more definitive interpretation of the *in vivo* data. In this article, we aim to equip the clinician with knowledge of the background physics involved in MRS, so that informed decisions can be made for research studies.

NUCLEAR MRS

NMR refers to the behaviour of atoms subjected to a magnetic field. The phenomenon was first described in 1946 by Bloch and Purcell. Atoms with an odd mass number such as ^{1}H, ^{31}P and ^{13}C possess the quantum property of "spin" and behave as dipoles aligning along the axis of an applied magnetic field (Figure 1). During relaxation following excitation, radiofrequency signals are generated which can be expressed as a frequency spectrum. Hydrogen is the most abundant atom in living organisms and using high power magnetic fields on *in vitro* samples, high resolution metabolic spectra can be obtained with clearly defined metabolite peaks of small molecules (<2 kDa).

NUCLEAR SPIN AND ORIENTATIONS

Nuclear resonance occurs because the nuclei of at least one of the isotopes of most elements possess a magnetic moment. A magnetic moment arises because the nucleus may have 'spin', and is also charged. "Spin" can be understood as the nucleus of an atom spinning around its own axis.[3]

When placed in a constant magnetic field, nuclei that possess spin can be excited, the energy of the magnetic moment depends on the orientation of the nucleus with respect to that field.[3] Application of electromagnetic

Keywords: nuclear magnetic resonance, magnetic resonance imaging, magnetic resonance spectroscopy, metabolomics
Received: 31.08.2015; *Accepted:* 26.10.2015; *Available online:* 12 November 2015
Address for correspondence: Joshua Tognarelli, Liver Unit, Department of Medicine, 10th Floor QEQM Wing, St Mary's Hospital, Imperial College London, Praed Street, London W2 1NY, United Kingdom.
Tel.: +44 207 886 6454; fax: +44 207 402 2796.
E-mail: jt2110@imperial.ac.uk
Abbreviations: CPMG: Carr-Purcell-Meiboom-Gill sequence; CSI: chemical shift imaging; FID: free induction decay; K: Kelvin; KEGG: Kyoto Encyclopedia for Genes and Genomes; MR: magnetic resonance; MRI: magnetic resonance imaging; MRS: magnetic resonance spectroscopy; MSEA: metabolite set enrichment analysis; NMR: nuclear magnetic resonance; NOESY: nuclear Overhauser enhancement spectroscopy; PC: principal components; PCA: principal components analysis; PLS-DA: partial least squared discriminant analysis; PRESS: point-resolved spectroscopy; STEAM: stimulated echo acquisition mode; T: Tesla; T_1: spin-lattice relaxation; T_2: spin-spin relaxation; TE: echo time; TMAO: trimethylamine N-oxide; TR: repetition time
http://dx.doi.org/10.1016/j.jceh.2015.10.006

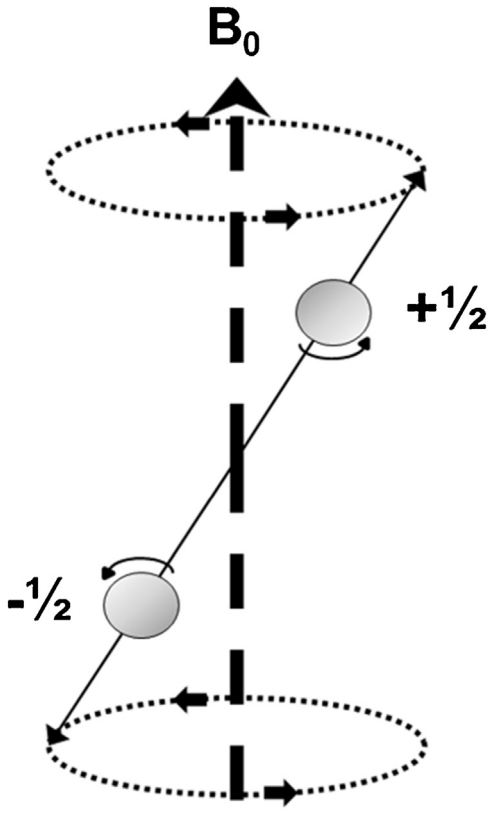

Figure 1 Precession of protons aligned to a magnetic field (B_0).

radiation at a suitable frequency can stimulate transitions between high and low energy states, this transition in energy level providing the basis for NMR spectroscopy, as the energy absorbed can be detected.[3,4]

In an applied magnetic field, the magnetic moments of nuclei become oriented relative to the direction of the applied field in a number of ways, determined by the nuclear spin quantum number, *I*. For protons, $I = ½$, so the number of orientations of proton magnetic moments in the applied field is $(2 \times ½ + 1) = 2$, *i.e.* parallel and anti-parallel to the applied field direction.[5] These two orientations are characterised by two different energy levels (the parallel orientation being lower in energy than the anti-parallel) whose separation ΔE depends on the applied field strength, B_0, as given in equation, where *h* is Planck's constant (Figure 2).[4]

Electromagnetic radiation in the radio-frequency range causes transitions between the two energy levels, giving the possibility of ^{1}H NMR spectra. Since $\Delta E = h\nu$, it follows that the resonance frequency ν is proportional to the field strength B_0.

$$\nu = \frac{\gamma}{2\pi} \times B_0$$

Resonant absorption by nuclear spin will occur only when electromagnetic radiation of the correct frequency (Larmor precession rate) is applied to match the energy difference between the

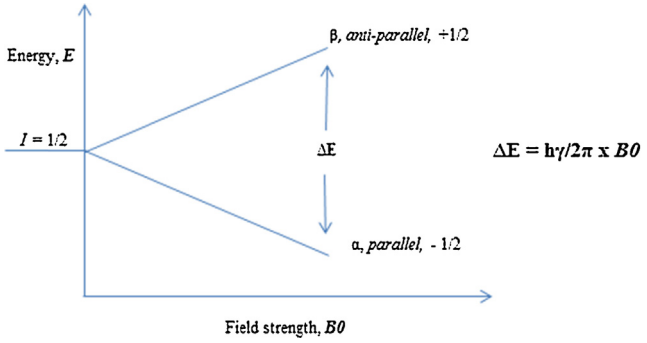

Figure 2 Energy levels depicted on a ½ *I* spin nuclei.

nuclear spin levels in a constant magnetic field of appropriate strength.

CHEMICAL SHIFT

In a molecule, the magnetic field that a particular proton experiences is influenced by that due to the motions of nearby electrons (the chemical environment to which it is subjected). Differently sited protons experience slightly different effective applied fields and resonate at slightly different frequencies; it is this which gives ^{1}H NMR its diagnostic value, as this property can be used to discern different proton environments within molecules.[4] This effect is called 'chemical shift' and if a nucleus in a specific chemical group is shielded to a higher degree by a higher electron density of its surrounding molecular orbital, then the NMR frequency will be shifted "up field" (that is a lower chemical shift), whereas if it is less shielded by surrounding electron density, then the NMR frequency will be shifted "downfield" (that is, a higher chemical shift). Thus, the magnetic environment experienced by each MR sensitive nucleus may be different. Although all nuclei are dominated by the static magnetic field strength, B_0, and by the applied field B_1, they will also experience a local magnetic field due to the magnetic fields of the electrons within their immediate chemical environment. Thus, the degree of shielding or enhancement of the local magnetic field by electron currents depends upon the exact electronic environment, which is a function of the precise chemical structure of the molecule.[4]

SPIN–SPIN COUPLING

When protons occur in more than one kind of environment within a molecule, circumstances may allow their spins to interact with one another. The influence of one proton's spin on another is due to the shielding effects of its electrons, which can cause the magnetic energy experienced by a neighbouring proton to be slightly stronger or weaker if the magnetic moment of the neighbouring proton is parallel or perpendicular to the magnetic force