Electron paramagnetic resonance (EPR) measures the signal from unpaired electrons and is a technique already widely used in the bioscience and pharmaceutical industries. Dr Joshua R. Biller of TDA Research, Inc, USA is championing the novel use of EPR to answer research questions. He explains that rapid innovations in EPR will allow a wider group of researchers to image tumours, learn about the structure and function of proteins, and observe the mechanisms used by enzymes, amongst many other possibilities.

Electron paramagnetic resonance (EPR) spectroscopy is the study of the interaction between a substance and electromagnetic radiation. The output is called a spectrum and can be viewed as the signature of the sample. Whilst spectroscopy is vital to progressing knowledge about the structure of atoms and molecules, and in analysing materials with unknown chemical composition, it also has practical applications. Spectroscopy can be used to improve the structure of drugs, to search for biomarkers of disease, and is the basis of medical imaging.

A well-known form of spectroscopy is nuclear magnetic resonance (NMR) spectroscopy. NMR is a technique used to explore magnetic fields around the very centre of atoms, an area called the nucleus. An atom is the smallest particle of a chemical element that can exist, and it is made up of protons, electrons, and neutrons. NMR is most often used in chemistry to identify different molecules, as each molecule has its own distinct NMR spectrum. EPR is one of a broader category of spectroscopic techniques based on the magnetic properties of an atom. A second type of magnetic spectroscopy which measures information about unpaired electrons is called electron paramagnetic resonance (EPR).

THE MAGNETIC PROPERTIES OF ELECTRONS

Usually, electrons exist in pairs as this is the preferred lowest energy state. The magnetic characteristic of an electron is cancelled out when it is paired, and compounds with all paired electrons are termed diamagnetic. However, many compounds exist with unpaired electrons which can interact with an external magnetic field, and this is called paramagnetic.

Electron paramagnetic resonance (EPR) spectroscopy is the study of molecules or atoms with unpaired electrons. It was first discovered by Yevgeny Zavoisky in 1944 in Kazan, Russia. The technique characterises the paramagnetic material by placing it in a magnetic field and applying microwave radiation. The EPR spectrum provides information on the type of chemical environment that the unpaired electron resides in.

In EPR, the environment surrounding an unpaired electron can be carefully measured and analysed. This information can be used to learn more about materials, including metals like iron, nickel, or chromium, as well as a variety of organic radicals.

Historically, EPR instruments have been developed to operate at increasingly higher magnetic field strengths, since the sensitivity of the experiment increases with the applied magnetic field. However, advances in computer power and improvement in the quality of electronics now allow quality EPR experiments in magnetic fields with low strength. This leads to more opportunities to use the technique outside a laboratory setting, as has been done with NMR. One benefit of low field EPR is sample preparation, which is very different from the traditional high field approaches. For example, low field EPR can be used to analyse much larger sample sizes.

Over the years, there have also been changes in how data are collected from EPR. Rapid-scan allows the magnetic field to be repeatedly scanned much quicker than conventional EPR, leading to an increase in signal acquired per unit time. Pulsed EPR allows different types of interactions between the unpaired electron and surrounding nuclei to be separated more efficiently.

EPR was once considered inflexible, expensive, and only useful in a laboratory setting. However, advancements in technology mean that this impression no longer rings true and the EPR community are keen to show others the benefits of the technology. One example of this outreach is the NSF-funded SHARED-EPR network (shared-epr.org). The website is a portal to the EPR community designed to bring together scientists from the fields of chemistry, biochemistry, physics, materials science, medicine, and biology to disseminate and advance the field of EPR spectroscopy.

Dr Joshua Biller of TDA Research, Inc, USA, claims that EPR is often under-recognised and under-appreciated, and his aim is to change this.

HOW DOES EPR WORK?

The EPR sample is held in the magnetic field in a structure called a resonator. This is where the microwave interacts with the sample and a signal is detected. Newer resonators have a much wider range of geometries than in older spectrometers which permits analysis of a much wider range of samples, and also a wider range of sample sizes. This is one limitation to wider application of EPR which has now been overcome.

Dr Biller explains that another previous limitation of EPR was a lack of computer power; the hardware did not have a large enough capacity to analyse the data. Due to a rapid progression of technologies, it is now possible to use miniaturised electronic devices with sufficient processing power, to analyse results. One of the main advantages of this improvement in computing power and resonator geometries is that EPR hardware can now be configured to analyse samples in situ (in their natural environment).

USING EPR TO ANSWER RESEARCH QUESTIONS

Dr Biller highlights several novel uses for EPR, in treating cancer and learning more about neurodegenerative diseases.

Low oxygen levels are seen in tumours with uncontrollable growth and proliferation, as the formation of abnormal blood vessels can lead to reduced transport of oxygen and nutrients to the tumour. Tumour hypoxia has shown to be linked to a poor prognosis and increased resistance to treatment, therefore the ability to modify treatment based on oxygen levels may help improve survival. One major limitation has been the inability to identify patients for whom this approach may be helpful.

In pre-clinical imaging, EPR is used to image tumour oxygen levels, to help guide radiotherapy treatment. This is done through oxygen-sensitive spectroscopy and imaging, which is used to work...
Electron ‘spins’ are everywhere, and new advances in technology mean EPR researchers can measure EPR sensitive molecules in the places they exist naturally.

EPR researchers can measure EPR sensitive molecules in the places they exist naturally. EPR can be applied to many research problems, including neurodegenerative diseases, and may play a vital role in uncovering new ways to combat these conditions. The technology can also be used in the pharmaceutical industry to monitor product stability and shelf life. The same types of information that are important to medical researchers, like redox state, oxygen concentration, and pH, are also of interest to researchers looking at technologically advanced materials like carbon fibre and those interested in corrosion research. In situ materials research represents an untapped potential for future EPR applications.

Since its discovery in the 1940s, EPR has become an increasingly elegant and accessible technology, and with a focus on securing funding to continue to explore the abilities of EPR, its research potential is boundless.