1P513**Solid-State ^{17}O NMR of Biological Molecules**

○Kazuhiko Yamada, Miwako Asanuma, Toshio Yamazaki, Hiroshi Hirota
RIKEN Genomic Sciences Center

Oxygen is one of the most common elements in biological compounds. Although ^1H , ^{13}C , and ^{15}N Nuclear Magnetic Resonance (NMR) spectroscopy has made a tremendous impact on the development of protein research, very little ^{17}O NMR studies have been reported on biological molecules. This is because unfavorable nuclear properties of ^{17}O ($I=5/2$, natural abundance = 0.037%, $\gamma = -3.6279 \times 10^7 \text{ rad T}^{-1} \text{ s}^{-1}$, $Q(^{17}\text{O}) = -2.6 \text{ fm}^2$) have made it difficult to carry out ^{17}O NMR experiments. Thanks to the recent development of solid-state NMR methodologies including the introduction of high-field instruments, inventions of effective NMR pulse-sequences, and so on, however, we are able to obtain high-quality solid-state ^{17}O NMR spectra and extract reliable information concerning the ^{17}O NMR parameters. Recently, our group has developed an efficient synthetic route for ^{17}O labeling to α -COOH groups of Fmoc-protected amino acids without racemization (Japanese Patent 2006-8666), making it possible to label ^{17}O into amino acids and peptides. In this presentation, we will report the experimental determination of the carboxyl and carbonyl oxygen Electric-Field-Gradient (EFG) and Chemical Shielding (CS) tensors in the twenty L-amino acids and peptides, showing that solid-state ^{17}O NMR is useful as a new nuclear probe in the investigations of proteins. Our systematic investigations demonstrate that the obtained ^{17}O NMR tensors become a highly sensitive probe to molecular geometries, secondary structures, and hydrogen bonding interactions.

1P515**Development of a High-Sensitivitive, Multi-Frequency EPR system as a New Tool for Research on Metalloproteins with Integer Electron Spin**

○Haruhiko Yashiro¹, Takanari Kashiwagi¹, Masaki Horitani², Hiroshi Hori², Masayuki Hagiwara¹

¹KYOKUGEN, Osaka Univ., ²Dept. of Bioeng, Osaka Univ.

Multi-Frequency Electron Paramagnetic Resonance (MFEPR) is a powerful means to clarify the microscopic information such as electronic structures of magnetic species. One of the most promising applications of MFEPR for biomolecular research is to study metalloproteins with an integer spin or totally integer one due to multinuclear complex, the so-called spin silent system (SSS) which has a larger anisotropy than the conventional EPR frequency (35 GHz). SSSs can be found in many important proteins, such as deoxy hemoglobin (Fe^{2+} $S=2$) and oxygen evolving complex (Mn_4 cluster) etc. In spite of many trials, there has been still no report on MFEPR of SSSs. Here we report our high sensitive MFEPR system in order to be used for samples with dilute magnetic ions. To cover the wide frequency range (35-600GHz) with high sensitivity, we have developed two types of resonators with a simple reflection bridge in a 16T superconducting magnet. For higher frequency region (50-600GHz), we have made a Fabry-Perot Resonator (FPR) which has one to two orders of magnitude higher finesse value than previously reported ones. Below 130GHz, we have made ordinary TE_{011} cylindrical resonators from 35 to 130GHz, since they have theoretically an order of magnitude higher sensitivity than FPR. We could achieve 10^{11} spins/G for FPR and 10^{10} spins/G for TE_{011} resonators at 1.5K as expected. EPR spectra in a couple of metalloprotein samples including one of SSSs have been successfully observed utilizing this MFEPR system. This work was partly supported by MEXT (No.451).

1P514**Potential Measurement of Lipid Bilayer Membrane by Microfluidic FET Device**

○Yoshihiko Kuwana, Katsura Kojima, Yasushi Tamada
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We have reported the preliminary studies for a new type biosensor by utilizing and mimicking the insect sensory functions. Our proposing biosensor is to combine the receptors from insect sensory organs with a field effect transistor (FET) and it is expected to have a high sensitivity and selectivity to the stimulus as well as insects. In order to form lipid membranes, we used polystyrene microchambers with a 200- μm -diameter aperture. Diphytanoyl phosphatidylcholine was dissolved in decane for lipid solution. A lipid bilayer could be easily formed by painting lipid on an aperture in the buffer solution. We fabricated a field effect transistor for potential measurement. To exchange the buffer, microfluidic system was employed. Bilayer membrane potential changes by stimulating with salt solution and it was recorded by our FET device and by a commercial patch clamp amplifier simultaneously. Two results showed a very good accordance. This indicates that our FET device can record the membrane potentials with a quite low noise. We are going to immobilize the pheromone sensitive proteins, such as receptors, ion channels and G proteins, extracted from male silk moth antennae. As a next step, such proteins will be molecularly cloned and expressed on cultured insect cells and transgenic silk moths for our receptor-based biosensor.

1P516**Ab initio calculation of NMR chemical shifts of large molecules by FMO method**

Qi Gao^{1,2}, ○Satoshi Yokojima¹, Takao Kobayashi¹, Shinichiro Nakamura¹

¹Mitsubishi Chemical Group Science and Technology Research Center, INC. and CREST-JST, ²Department of Biomolecular Engineering, Tokyo Institute of Technology

The structure determination of biomolecules is a very important topic in biophysics. The NMR chemical shift is playing one of the key roles in the field. Thus, the accurate determination of the chemical shift is desirable and for that, the ab initio calculation is required. However, the size of the biomolecule is usually too large to perform the ab initio calculations. Moreover the effects of the surrounding environment have to be taken into account, which increase the computational cost further. Therefore, it is very difficult to use the ab initio calculations to determine the NMR chemical shift to identify the structure of biomolecules. The fragment molecular orbital (FMO) method is one of the techniques to overcome the problem, i.e., it can calculate the electronic structure of large systems by dividing the systems into fragments. In this presentation, we show the effectiveness of the method.