MLF Experimental Report	提出日 Date of Report
課題番号 Project No. 2017A0183 実験課題名 Title of experiment Determination of muon stopping sites and electronic structures in proteins 実験責任者名 Name of principal investigator Yoko Sugawara 所属 Affiliation Kitasato University, School of Science	装置責任者 Name of responsible person Yasuhiro Miyake 装置名 Name of Instrument/（BL No.） Muon S1(ARTEMIS) 実施日 Date of Experiment 2017/5/25 9:00 - 2017/5/29 9:00

試料、実験方法、利用の結果得られた主なデータ、考察、結論等を、記述して下さい。（適宜、図表添付のこと）
Please report your samples, experimental method and results, discussion and conclusions. Please add figures and tables for better explanation.

1. 試料 Name of sample(s) and chemical formula, or compositions including physical form.

Glycine-^{14}N: $H_2{}^{14}NCH_2COOH$

Glycine-^{15}N: $H_2{}^{15}NCH_2COOH$

Glycylglycine-^{14}N: $H_2{}^{14}NCH_2CO{}^{14}NHCH_2COOH$

2. 実験方法及び結果（実験がうまくいかなかった場合、その理由を記述してください。）
Experimental method and results. If you failed to conduct experiment as planned, please describe reasons.

The electron transfer process is highly important in the field of bioscience. Cytochrome *c* is a small heme protein and the member of the respiratory chain in mitochondria. It transfers an electron from protein complex III to IV, coupled with redox reactions of Fe located at the center of a porphyrin ring. Nagamine and his co-workers have been investigating electron transfer processes in cytochrome *c* by a labelled-electron method with positive muons [1]. The LF μSR data showed a level crossing resonance (LCR) signature for cytochrome *c* (Fig. 1(a)). The observed LCR data was similar to that of polyglycine at around 20 G (Fig. 1(b)) [2]. Polyglycine is the simplest oligomer of amino acids. It made of aliphatic (–CH_2–) parts, peptide bonds (–CONH–) and terminal –COOH (or –COO^-) and –NH_2 (or –NH_3^+) groups. Candidates of muon stopping sites are peptide bonds (–CONH–) and terminal –COOH or –NH_2. Under such a background, we carried out μSR measurements of glycine and glycylglycine. Measurements of μSR time spectra under the various longitudinal fields from 0 G to 3.5 kG at 300 K, 200 K, 100 K, 50 K and 8 K using the single pulse mode. The μSR of glycine-^{14}N and -^{15}N were compared to examine the effect of quadrupole, because nitrogen-14 (^{14}N) has quadrupole moment (I= 1, Q = 0.016 barn) and nitrogen-15 (^{15}N) has no quadrupole (I = –1/2, Q = 0).

2. 実験方法及び結果（つづき） Experimental method and results (continued)

Muon relaxation time spectra at zero field are shown in Fig. 2(a). There exists distinct difference between the relaxation spectra of glycine (Gly) and those of glycylglycine (Glygly), but no significant difference between the relaxation rates of glycine-^{14}N and -^{15}N.

Low LF μSR data of glycine were fitted well using the product of Kubo-Toyabe and Lorentzian functions. LCR was not detected around 20 G (Fig. 2(b): upper half) contrary to the cases of cytochrome c and polyglycine. On the other hand, low LF μSR data of glycylglycine was approximately fitted with the Lorentzian function, and LCR was observed (Fig. 2(b): lower half). In addition, the missing fraction of the initial asymmetry of the glycylglycine under zero magnetic field was approximately 20% of the full asymmetry, and larger than that of glycine (approximately 10%) (Fig.2(a)).

Taking the results of theoretical calculations into account [3], it was concluded that muon would stop at –COOH (or –COO$^-$) moiety in glycine, and at CO moiety of peptide bonds in oligoglycine. The results indicate that LCR data of cytochrome c would originate from muon stopping at peptide bonds.

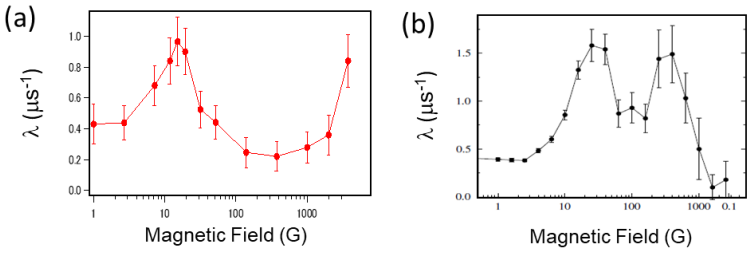

Fig. 1 LF dependence of the relaxation rates (λ) of cytochrome c at 18K (a) and polyglycine at 5K (b)

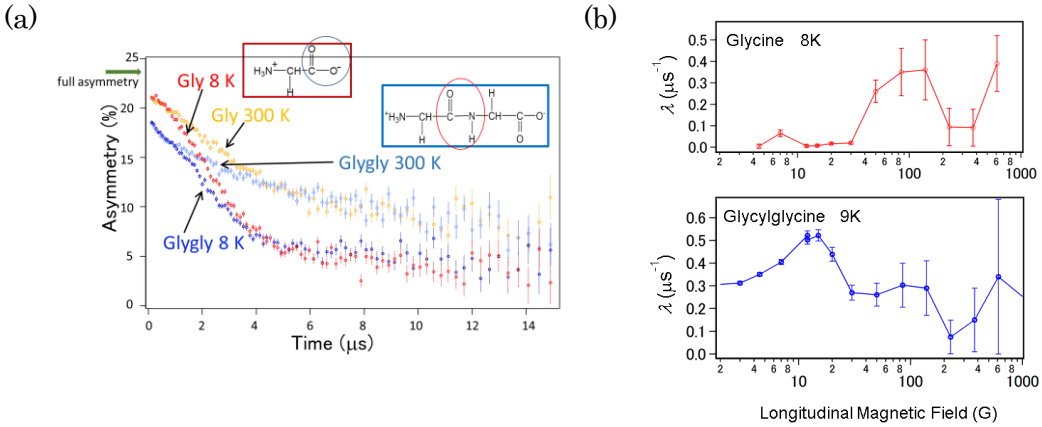

Fig. 2 Temperature dependence of μSR at ZF (a) and LF dependence of relaxation rate (λ) (b) in glycine and glycylglycine.

[1] K. Nagamine *et al*., Physica B, **289-290**, 631(2000).; K. Nagamine and E. Torikai, J. Phys. Condensed Matter **16,** S4797 (2004).

[2] F. L. Pratt *et al*. (private communication).

[3] A. D. Pant *et al*., JPS Conference Proceedings, **21**, 011038(2018).