Enhanced Sampling Method in Molecular Simulations Using Genetic Algorithm for Biomolecular Systems

Yoshitake Sakae,[a] John E. Straub,[b] and Yuko Okamoto *[a,c,d,e,f]

We propose a molecular simulation method using genetic algorithm (GA) for biomolecular systems to obtain ensemble averages efficiently. In this method, we incorporate the genetic crossover, which is one of the operations of GA, to any simulation method such as conventional molecular dynamics (MD), Monte Carlo, and other simulation methods. The genetic crossover proposes candidate conformations by exchanging parts of conformations of a target molecule between a pair of conformations during the simulation. If the candidate conformations are accepted, the simulation resumes from the accepted ones. While conventional simulations are based on local update of conformations, the genetic crossover introduces global update of conformations. As an example of the present approach, we incorporated genetic crossover to MD simulations. We tested the validity of the method by calculating ensemble averages and the sampling efficiency by using two kinds of peptides, ALA3 and $(AAQAA)_3$. The results show that for ALA3 system, the distribution probabilities of backbone dihedral angles are in good agreement with those of the conventional MD and replica-exchange MD simulations. In the case of $(AAQAA)_3$ system, our method showed lower structural correlation of α-helix structures than the other two methods and more flexibility in the backbone ψ angles than the conventional MD simulation. These results suggest that our method gives more efficient conformational sampling than conventional simulation methods based on local update of conformations. © 2018 Wiley Periodicals, Inc.

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Introduction

Molecular dynamics (MD) and Monte Carlo (MC) simulation methods are very powerful tools for studying biomolecular systems such as proteins and DNA. However, there are also some problems, one of which is that the biomolecular systems generally have a large number of degrees of freedom and are characterized by many local minima separated by high-energy barriers. Therefore, conventional MD and MC simulations tend to get trapped in states of local minima. In order to solve this problem, various sampling and optimization methods such as simulated annealing (SA),[1] replica-exchange method (REM),[2,3] multicanonical algorithm (MUCA),[4,5] simulated tempering (ST),[6,7] and their extensions (for example, see, Refs. 8–18) have been proposed. Genetic algorithm (GA)[19,20] has also been recognized by researchers as powerful tools for various optimization problems including biological systems. The GA mimics the process of natural evolution and uses the optimization procedures of natural gene-based evolution, that is, mutation, crossover, and replication. For a certain optimization problems, this algorithm has been found to be an excellent strategy to find global minima. The conformational search or optimization approaches for biomolecules using the GA has also been performed.[21–25]

We have also proposed conformational search method referred to as the parallel simulated annealing using genetic crossover,[26–30] which is a hybrid algorithm combining MC (or MD) simulated annealing[1] and GA. While GA is usually used for finding the global-minimum energy state, we have proposed to use genetic crossover to just introduce global update of conformations to enhance conformational sampling in conventional MC or MD simulations.[31,32] The methods can be respectively referred to as the parallel Monte Carlo using genetic crossover (PMC/GAc) and the parallel molecular dynamics using genetic crossover (PMD/GAc). Whereas the selection rule after genetic crossover in usual GA is to choose the lowest-energy conformations, that in PMC/GAc or PMD/GAc is based

[a] *Y. Sakae, Y. Okamoto*
Department of Physics, Graduate School of Science, Nagoya University, Nagoya, Aichi, 464-8602, Japan
E-mail: okamoto@tb.phys.nagoya-u.ac.jp
[b] *J. E. Straub*
Department of Chemistry, Boston University, Boston, Massachusetts, 02215-2521
[c] *Y. Okamoto*
Information Technology Center, Nagoya University, Nagoya, Aichi, 464-8601, Japan
[d] *Y. Okamoto*
Structural Biology Research Center, Graduate School of Science, Nagoya University, Nagoya, Aichi, 464-8602, Japan
[e] *Y. Okamoto*
Center for Computational Science, Graduate School of Engineering, Nagoya University, Nagoya, Aichi, 464-8603, Japan
[f] *Y. Okamoto*
JST-CREST, Nagoya, Aichi, 464-8602, Japan

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on the Metropolis criterion.[31,32] Hence, the purpose for introducing genetic crossover here was not finding the global-minimum-energy state, but obtaining accurate ensemble averages of physical quantities at a certain temperature. Furthermore, we have shown that PMC/GAc or PMD/GAc can be naturally combined with the REM.[32]

In PMC/GAc or PMD/GAc, there is one complication: the "child" conformations that are produced from a pair of "parent" conformations by genetic crossover often have high-potential energy and are unstable due to steric hindrance. We simply introduced short relaxation simulations with restraints on the backbone structures in order to obtain stable "child" conformations.[32] However, this introduction of extra simulations may break the detailed balance condition, which may result in giving inaccurate ensemble averages of physical quantities. In this article, we propose to employ Chen and Roux's method,[33] which guarantees the detailed balance condition, for this relaxation simulation in PMC/GAc or PMD/GAc. We applied this modified PMD/GAc method to two kinds of peptide systems, ALA3 and $(AAQAA)_3$ in order to confirm the validity for the ensemble average calculations of physical quantities and the conformational sampling efficiency. The simulation results were compared with those of conventional MD and replica-exchange molecular dynamics (REMD)[3] simulations.

This article is organized as follows. In "Methodology" section, we explain the present methods. In "Computational Details" section, we explain the details of the simulations. In "Results and Discussion" section, we present the results. "Conclusions" section is devoted to conclusions.

Methodology

Parallel MC or MD using genetic crossover

In Figure 1, we show the flow charts of the PMC/GAc or PMD/GAc method[31,32] and the original GA for comparison. We first prepare M initial conformations of the system in study, where M is the total number of "individuals" in GA and is usually taken to be an even integer. We then alternately perform the following two steps:

1. For the M individuals, regular canonical MC or MD simulations at temperature T are carried out simultaneously and independently for certain MC or MD steps.
2. $M/2$ pairs of conformations are selected from "parental" group randomly, and the crossover operation is performed to create preliminary "child" conformations. Perform short simulations of the "parent" conformations with restraint on the backbone structures to obtain final, stable "child" conformations (propagation process). The obtained final "child" conformations are "selected" or accepted from the parents with the following Metropolis criterion:

$$w(\mathrm{p}\to\mathrm{c}) = \min\left(1, \exp\left\{-\beta\left[E_\mathrm{c} - E_\mathrm{p}\right]\right\}\right), \qquad (1)$$

where E_p and E_c stand for the potential energy of the parental conformation and the final child conformation of the parent–child pair, respectively, and β is the inverse temperature defined by $\beta = 1/k_\mathrm{B}T$ (k_B is the Boltzmann constant).

In Step 2, we can employ various kinds of GAc operations. Here, we just present a case of the two-point crossover.[30] The following procedure is carried out:

1. Consecutive amino acids of length n residues in the amino-acid sequence of the conformation are selected randomly for each pair of selected conformations.
2. Dihedral angles (in only backbone or all dihedral angles) in the selected n amino acids are exchanged between the selected pair of conformations.

Note that the length n of consecutive amino-acid residues can, in general, be different for each pair of selected conformations.

We need to deal with the produced "child" conformations with care. Because the produced conformations often have unnatural structures by the crossover operation, they have high-potential energy and are unstable. Therefore, a propagation process is introduced before the selection operation. As the propagation process, we perform a short MD simulation with restraint potentials $E_\mathrm{rst}(\theta)$ of the (backbone) dihedral angle θ in the selected n amino acids as follows:

$$E_\mathrm{rst}(\theta) = k_\theta(\theta - \theta_\mathrm{child})^2 \qquad (2)$$

where k_θ is the force constant, and θ_child is a dihedral angle proposed by exchanging dihedral angles between "parent" conformations by the crossover operation. The initial conformations for these propagation simulations are the ones before the crossover. Namely, by these propagation simulations, the corresponding backbone conformations of the n amino acids gradually transform from the ones before the crossover to the ones after the crossover. For the propagation process, we follow Chen and Roux's method,[33] which guarantees the detailed balance condition. Here, we simply perform the propagation simulation in the microcanonical NVE ensemble.

Computational Details

We applied the present method, conventional MD, and REMD, to two peptides, ALA3 (ACE-ALA-ALA-ALA-NME) and $(AAQAA)_3$ (ACE-ALA-ALA-GLN-ALA-ALA-ALA-ALA-GLN-ALA-ALA-ALA-ALA-GLN-ALA-ALA-NME). The potential energy of the solvated peptide systems is represented by the AMBER14SB[34] force field and TIP3P water model.[35] The total numbers of atoms of ALA3 system and $(AAQAA)_3$ system were 2241 and 10,053, respectively. For both systems, different initial structures for each conventional MD, each replica of REMD, or each individual of PMD/GAc are prepared (see Supporting Information Fig. S1). Before the production run, we perform the equilibrium simulations for 2.0 ns with NPT ensemble with periodic boundary conditions using the particle mesh Ewald (PME) method. The cutoff distances of 8.0 Å and 10.0 Å were used for the direct space

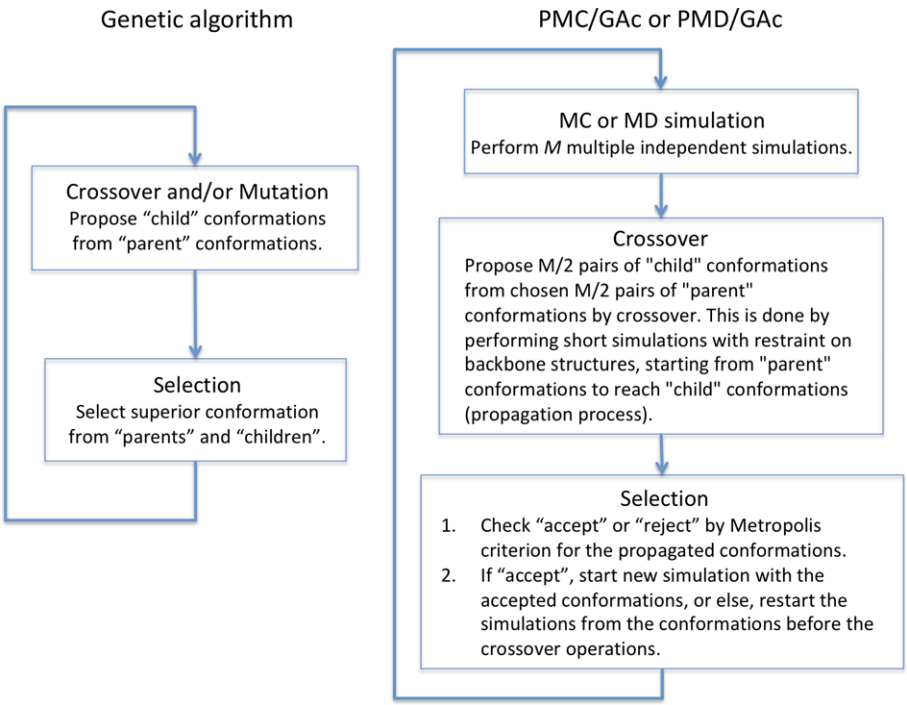

Figure 1. Flow charts of general genetic algorithm (left side) and PMC/GAc or PMD/GAc (right side). [Color figure can be viewed at wileyonlinelibrary.com]

sum of PME and van der Waals interactions of ALA3 system and $(AAQAA)_3$ system, respectively. Temperature was controlled with the Langevin thermostat with a collision frequency of 1.0 ps^{-1} at 300 K. Pressure regulation was achieved with isotropic position scaling with the Berendsen barostat with pressure around 1.0 atm and a pressure relaxation time of 1.0 ps. For all bonds involving hydrogens in the fragments, we used the SHAKE algorithm[36] as constraint algorithm to carry out the simulations with 2.0 fs as the time step. For production run, we performed all MD simulations using a Langevin dynamics integrator with NVT ensemble. AMBER14 program package[37] was used for MD simulations. The crossover and selection operations of PMD/GAc were performed by our own Python program and AmberTools14.[37]

Results and Discussion

In order to confirm the validity of PMD/GAc method, we compared with conventional MD and REMD by using ALA3 peptide. Each PMD/GAc simulation for sampling was carried out for every 200 ns with 28 individuals (M = 28). Namely, the total simulation time for sampling was 5.6 μs. We performed 1000 crossover operations for each individual, which selected consecutive amino-acid residues of length between 1 and 3, during simulations. For the propagation process of the crossover operation, we performed short simulations of 20 ps with the restraint potential (k_θ = 300 kcal/(mol rad^2) in eq. 2) for backbone dihedral angles, ϕ and ψ. On the other hand, the conventional MD simulations were performed 28 times for 200 ns independently. The acceptance ratio at the selection operation was 45.5%. For REMD simulations, the simulation time was 200 ns for each replica, and each simulation used 28 replicas. Replica exchange was tried every 1000 MD steps (2 ps). The 28 temperatures for REMD were distributed from 280 to 500 K: 280, 286, 292, 300, 305, 312, 319, 325, 332, 340, 347, 355, 362, 370, 378, 386, 395, 403, 412, 421, 430, 440, 449, 459, 469, 479, 489, and 500 K. In Figure 2, we show the probability distributions of ϕ and ψ angles of ALA3 peptide obtained from conventional MD, REMD, and PMD/GAc simulations. For estimating probability distributions at 300 K from the REMD simulations, the weighted histogram analysis method[38,39] was used. All backbone probability distributions from PMD/GAc were in good agreement with those of the conventional MD and REMD simulations.

In order to confirm the sampling efficiency of the PMD/GAc method, we also used $(AAQAA)_3$ peptide. Each PMD/GAc simulation for sampling was carried out for 200 ns with 56 individuals (M = 56). Namely, the total simulation time for sampling was 11.2 μs. For the crossover operation, we selected consecutive amino-acid residues of length between 2 and 15. The acceptance ratio at the selection operation was 25.8%. The conventional MD simulations were performed 56 times for 200 ns in order to match the number of the individuals of the PMD/GAc simulation. The REMD simulations were also performed 200 ns for each replica, and each simulation used 56 replicas. The 56 temperatures for REMD were distributed from 291 to 500 K: 291, 294, 297, 300, 303, 306, 309, 312, 315, 318, 321, 324, 327, 331, 334, 337, 341, 344, 347, 351, 354, 358, 361, 365, 369, 372, 376, 380, 383, 387, 391, 395, 399, 403, 407, 411, 415, 419, 423, 427, 431, 436, 440, 444, 449, 453, 458, 462, 467, 471, 476, 481, 485, 490, 495, and 500 K. The other simulation conditions of all simulation methods were the same as in the ALA3 system. In Supporting Information Figures S2–S4, we show the time series of the fraction of the amino acids identified as α-helix from the conventional MD, REMD, and PMD/GAc simulations. In Supporting Information Figures S5–S7, we show the conformations of lowest potential energy obtained from the conventional MD, REMD, and

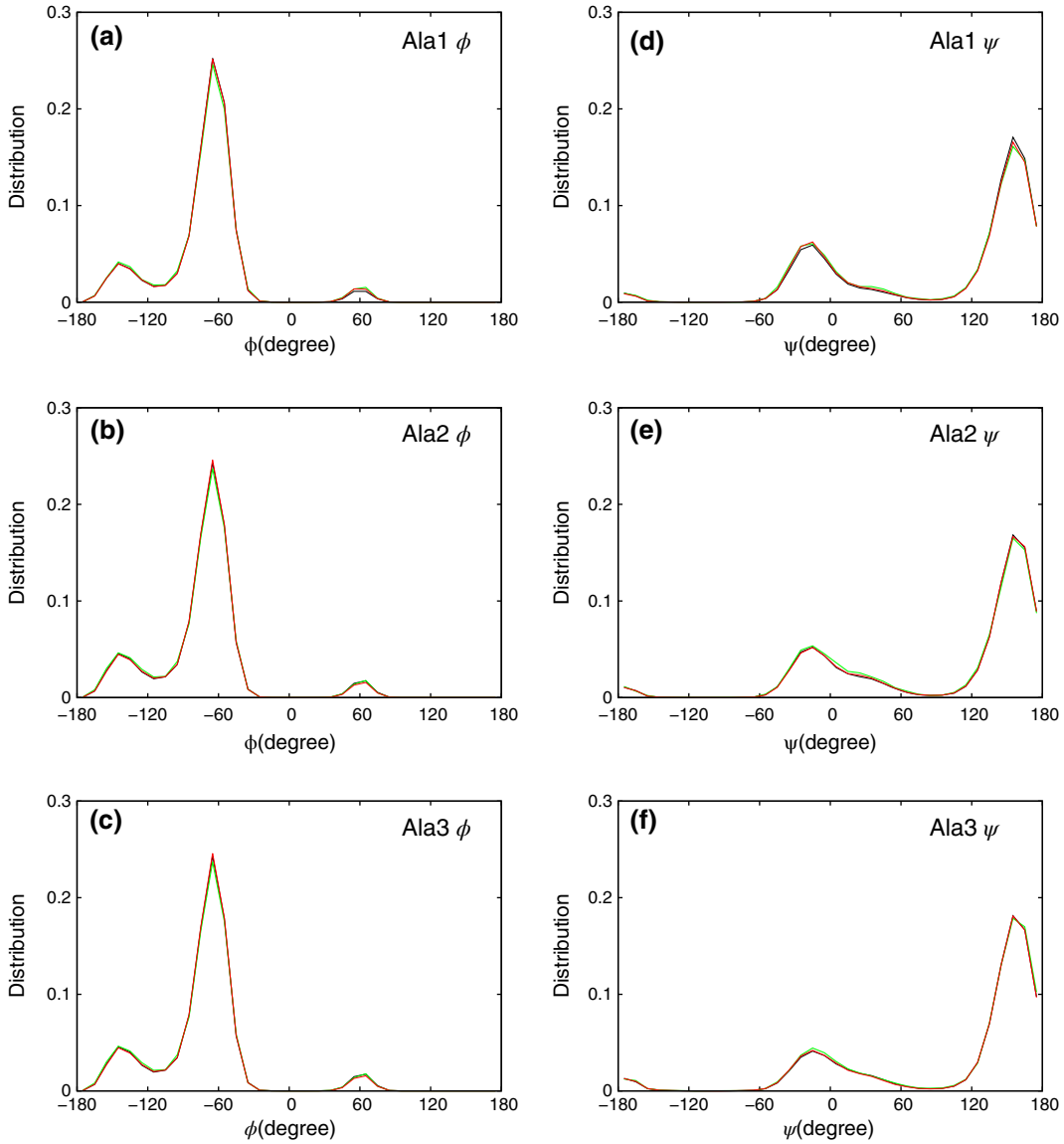

Figure 2. Probabilities of ϕ and ψ angles of ALA3 peptide obtained from MD (black line), REMD (green line), and PMD/GAc (red line). a), b), and c) stand for ϕ angles of residue 1, 2, and 3, respectively. d), e), and f) stand for ψ angles of residue 1, 2, and 3, respectively. [Color figure can be viewed at wileyonlinelibrary.com]

PMD/GAc simulations. The three methods do give quite different dynamical behaviors. However, they all converge to give canonical ensemble averages as shown below. In Figures 3 and 4, we show the probability distributions of ϕ and ψ angles of $(AAQAA)_3$ peptide obtained from conventional MD, REMD, and PMD/GAc simulations. The probabilities of ϕ of PMD/GAc roughly agreed with those of the conventional MD and REMD, however, in the case of the ψ angles, some peak heights were different from those of conventional MD and REMD. The convergence of average dihedral angles in the middle residues is shown in Supporting Information Figure S8. We see that as the simulation time becomes longer, the results of the conventional MD and REMD simulations tend to approach those of the PMD/GAc simulation.

As an indicator for the sampling efficiency of simulation methods, we used time-autocorrelation function $C_{\alpha\text{-helix}}(t)$ defined by,

$$C_{\alpha\text{-helix}}(t) = \frac{\langle A(t)A(0)\rangle - \langle A(0)\rangle^2}{\langle A(0)^2\rangle - \langle A(0)\rangle^2}. \quad (3)$$

Here, $A(t)$ is the number of amino acids identified as α-helix in the amino-acid sequence at lag time t and $\langle A(t)A(0)\rangle$ is defined by

$$\langle A(t)A(0)\rangle = \frac{\sum_{i=1}^{N} A(t+t_i)A(t_i)}{N}, \quad (4)$$

where t_i is the time of MD step i on the trajectory and N is the total number of the trajectory. We used the DSSP algorithm[40] for the criterion for α-helix formations. When $C_{\alpha\text{-helix}}$ is large for a long time, in general, a conformation of the peptide or protein keeps α-helix structure intact or keeps non-α-helix structures such as extended or random coil structures. On the

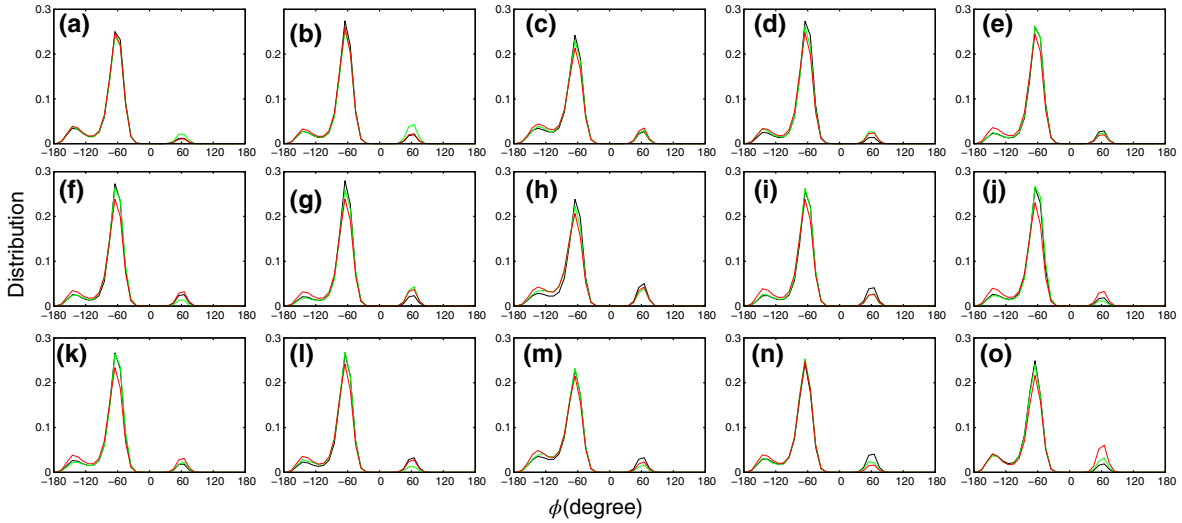

Figure 3. Probabilities of ϕ angles of $(AAQAA)_3$ peptide obtained from MD (black line), REMD (green line), and PMD/GAc (red line). a), b),..., o) stand for ϕ angles of residue 1, 2,..., 15, respectively. [Color figure can be viewed at wileyonlinelibrary.com]

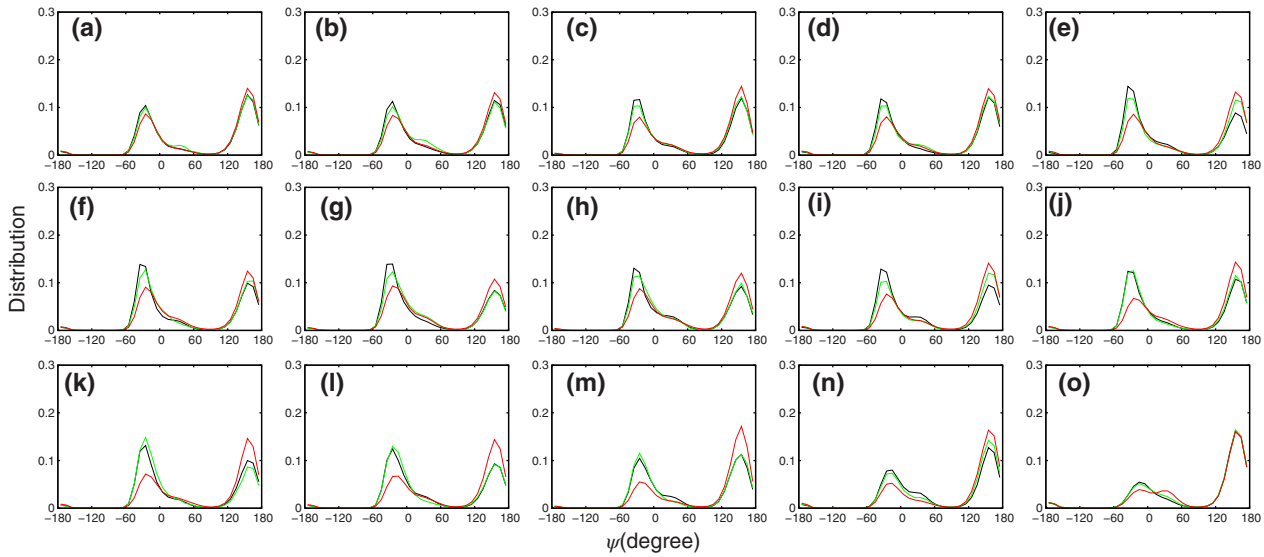

Figure 4. Probabilities of ψ angles of $(AAQAA)_3$ peptide obtained from MD (black line), REMD (green line), and PMD/GAc (red line). a), b),..., o) stand for ψ angles of residue 1, 2,..., 15, respectively. [Color figure can be viewed at wileyonlinelibrary.com]

contrary, in the case of small $C_{\alpha\text{-helix}}$ for a short time, the conformation of α-helix structure is quickly changing to other conformations during the simulation. In Figure 5, we show $C_{\alpha\text{-helix}}$ estimated from the conventional MD, REMD, and PMD/GAc simulations. $C_{\alpha\text{-helix}}$ of the conventional MD is always higher value than those of REMD and PMD/GAc simulations. In the case of PMD/GAc, although the $C_{\alpha\text{-helix}}$ is higher than that of REMD before a short time (~9.3 ns), it becomes lower than that of REMD in a long time, and finally, it is close to zero at 40 ns.

In Figure 6, the frequencies of flips of ψ angles from each independent simulation of the conventional MD, replica of REMD, and individual of PMD/GAc, are plotted. This flip frequency was evaluated by counting the number of back-and-forth moving between ($-85.0 \le \psi \le 85.0$) and ($-85.0 > \psi$ or $85.0 < \psi$) (degrees). Namely, we considered that the frequency of flips depends on the conformational change of an amino acid between α-helix region and

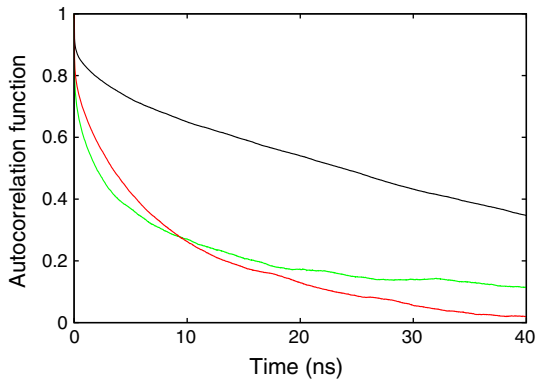

Figure 5. Autocorrelation function $C_{\alpha\text{-helix}}$ of the number of amino acids identified as α-helix in $(AAQAA)_3$ peptide, which is defined by eq. 3. Black line, green line, and red line stand for the functions of MD, REMD, and PMD/GAc, respectively. [Color figure can be viewed at wileyonlinelibrary.com]

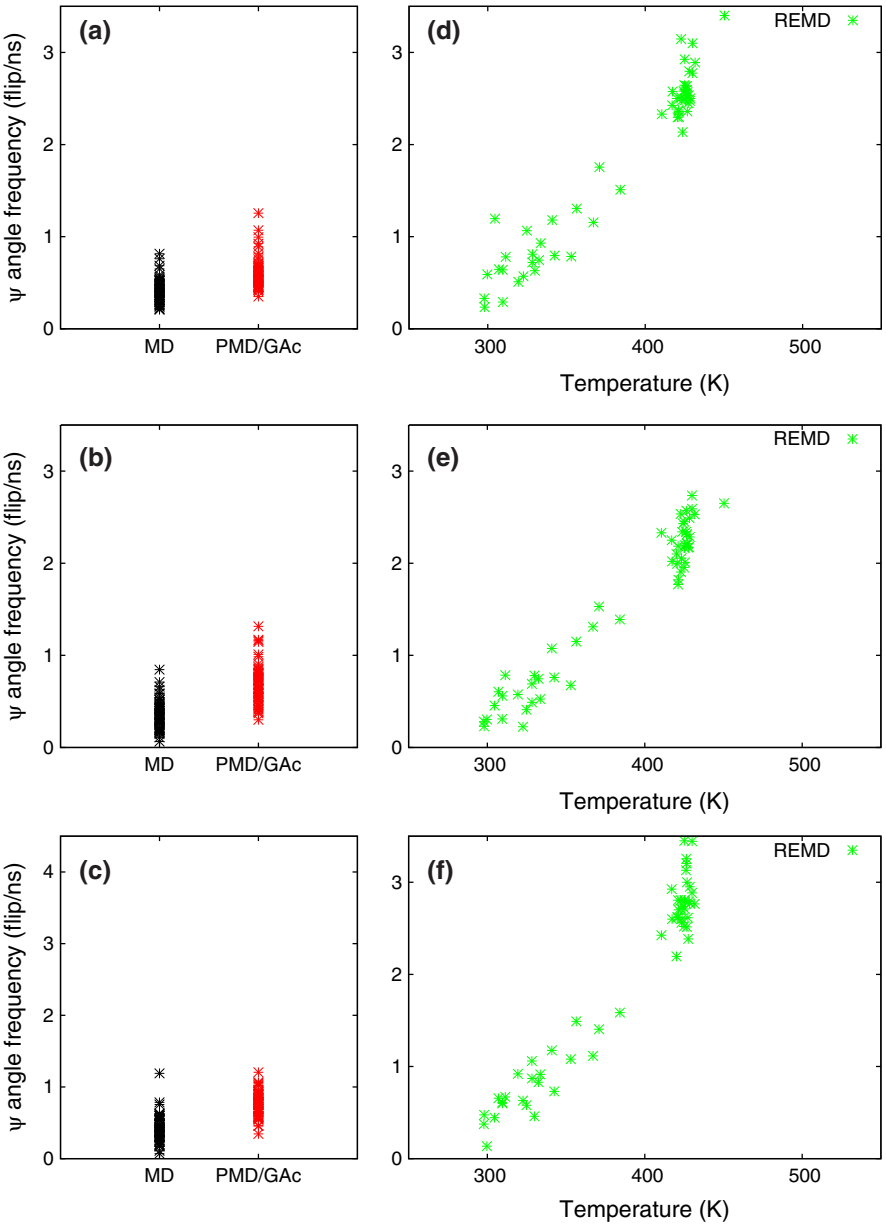

Figure 6. Frequency of flip of ψ angle for Gln3 (a, d), Gln8 (b, e), and Ala11 (c, f) in $(AAQAA)_3$ peptide. The black, green, and red points stand for each independent simulation of conventional MD, replica of REMD, and individual of PMD/GAc, respectively. For conventional MD and PMD/GAc, temperature was 300 K. For REMD, frequency as a function of average temperature for each replica is plotted. [Color figure can be viewed at wileyonlinelibrary.com]

extended region on the Ramachandran plot. The frequencies of PMD/GAc have higher values than those of the conventional MD as a whole. On the other hand, the frequency of REMD depends on the temperature strongly. Although some values of REMD at high temperatures have very high frequencies, those at low temperatures have low frequencies. In addition, we estimated the frequency ratio of flips of ψ angles between conventional MD and PMD/GAc in Figure 7. Especially, the frequencies of the residues around the middle of the amino-acid sequence increased largely for PMD/GAc.

We consider that one of the reasons of the difference of the probability in the case of $(AAQAA)_3$ peptide is the difference of the sampling efficiency between conventional MD, REMD, and PMD/GAc simulation methods because in the case of the smaller system (ALA3 peptide), the probability obtained from the three simulation methods was almost identical and, in the case of $(AAQAA)_3$ system, the correlation functions of α-helix structure obtained from the three methods were different. In particular, the

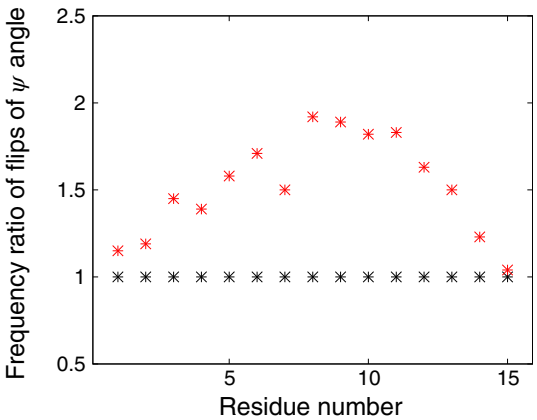

Figure 7. Frequency ratio of flip of ψ angle for $(AAQAA)_3$ peptide. The black points present the ratio of MD/MD (=1.0) and the red points present the ratio of (PMD/GAc)/MD. [Color figure can be viewed at wileyonlinelibrary.com]

efficiency by the crossover and selection operations increases at the residues around the middle of the amino-acid sequence in comparison with conventional MD. This tendency may be true even for larger systems.

Conclusions

In this study, we proposed PMD/GAc method based on GA as a sampling method of molecular simulation and applied this method to ALA3 and $(AAQAA)_3$ peptides. The results show that the ensemble averages of physical quantities obtained from the PMD/GAc method were in good agreement with those of conventional MD and REMD in the case of ALA3 system. On the other hand, in the case of $(AAQAA)_3$ system, there was a difference of the quantities among conventional MD, REMD, and PMD/GAc methods. The correlation time of α-helix structure obtained from PMD/GAc simulation was lower than those of conventional MD and REMD simulations, suggesting the superiority of PMD/GAc. In addition, $(AAQAA)_3$ peptide during PMD/GAc simulation was more flexible than that of conventional MD.

In the future, we will apply PMD/GAc method to larger protein systems because from the present results, there is a possibility that the larger proteins are even more effectively sampled. We are also considering the combination of genetic crossover with other sampling methods such as REMD, MUCA, ST, Smart Darting Method,[14] etc. Since PMD/GAc is suitable for parallel processing, it can be easily applied to large-scale computer systems.

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Additional Supporting Information may be found in the online version of this article.

[1] S. Kirkpatrick, C. D. Gelatt, Jr., M. P. Vecchi, *Science* **1983**, *220*, 671.
[2] K. Hukushima, K. Nemoto, *J. Physical Soc. Japan* **1996**, *65*, 1604.
[3] Y. Sugita, Y. Okamoto, *Chem. Phys. Lett.* **1999**, *314*, 141.
[4] B. A. Berg, T. Neuhaus, *Phys. Lett. B* **1991**, *267*, 249.
[5] B. A. Berg, T. Neuhaus, *Phys. Rev. Lett.* **1992**, *68*, 9.
[6] A. P. Lyubartsev, A. A. Martsinovski, S. V. Shevkunov, P. N. Vorontsov-Velyaminov, *J. Chem. Phys.* **1992**, *96*, 1776.
[7] E. Marinari, G. Parisi, *Europhys. Lett.* **1992**, *19*, 451.
[8] Z. Li, H. A. Scheraga, *Proc. Natl. Acad. Sci. USA* **1987**, *84*, 6611.
[9] I. Andricioaei, J. E. Straub, *Phys. Rev. E* **1996**, *53*, R3055.
[10] U. H. E. Hansmann, *Physica A* **1997**, *242*, 250.
[11] D. J. Wales, J. P. K. Doye, *J. Phys. Chem. A* **1997**, *101*, 5111.
[12] U. H. E. Hansmann, Y. Okamoto, *Phys. Rev. E* **1997**, *56*, 2228.
[13] Y. Sugita, A. Kitao, Y. Okamoto, *J. Chem. Phys.* **2000**, *113*, 6042.
[14] I. Andricioaei, J. E. Straub, A. F. Voter, *J. Chem. Phys.* **2001**, *114*, 6994.
[15] T. Whitfield, L. Bu, J. Straub, *Physica A* **2002**, *305*, 157.
[16] A. Mitsutake, Y. Okamoto, *J. Chem. Phys.* **2009**, *130*, 214105.
[17] J. Kim, T. Keyes, J. E. Straub, *J. Chem. Phys.* **2010**, *132*, 224107.
[18] J. Kim, J. E. Straub, *J. Chem. Phys.* **2010**, *133*, 154101.
[19] J. H. Holland, Adaptation in Natural and Artificial Systems, The University of Michigan Press, Ann Arbor, **1975**.
[20] D. E. Goldberg, Genetic Algorithms in Search, Optimization, and Machine Learning, Addison-Wesley, Reading, **1989**.
[21] T. Dandekar, P. Argos, *Protein Eng.* **1992**, *5*, 637.
[22] S. Sun, *Protein Sci.* **1993**, *2*, 762.
[23] R. Unger, J. Moult, *J. Mol. Biol.* **1993**, *231*, 75.
[24] A. A. Rabow, H. A. Scheraga, *Protein Sci.* **1996**, *5*, 1800.
[25] J. Lee, H. A. Scheraga, S. Rackovsky, *J. Comput. Chem.* **1997**, *18*, 1222.
[26] T. Hiroyasu, M. Miki, M. Ogura, Proceedings of the 44th Institute of Systems, Control and Information Engineers; **2000**, pp. 113–114.
[27] T. Hiroyasu, M. Miki, M. Ogura, Y. Okamoto, *J. IPS Japan* **2002**, *43*, 70.
[28] T. Hiroyasu, M. Miki, M. Ogura, K. Aoi, T. Yoshida, Y. Okamoto, Proceedings of the 7th World Multiconference on Systemics, Cybernetics and Informatics (SCI 2003), **2003**, *XIV*, pp. 117–122.
[29] Y. Sakae, T. Hiroyasu, M. Miki, Y. Okamoto, *Pac. Symp. Biocomput.* **2011**, *16*, 217.
[30] Y. Sakae, T. Hiroyasu, M. Miki, Y. Okamoto, *J. Comput. Chem.* **2011**, *32*, 1353.
[31] Y. Sakae, T. Hiroyasu, M. Miki, K. Ishii, Y. Okamoto, *J. Phys. Conf. Ser.* **2014**, *487*, 012003.
[32] Y. Sakae, T. Hiroyasu, M. Miki, K. Ishii, Y. Okamoto, *Mol. Sim.* **2015**, *41*, 1045.
[33] Y. Chen, B. Roux, *J. Chem. Theory Comput.* **2015**, *11*, 3572.
[34] J. Maier, C. Martinez, K. Kasavajhala, L. Wickstrom, K. Hauser, C. Simmerling, *J. Chem. Theory Comput.* **2015**, *11*, 3696.
[35] W. L. Jorgensen, J. Chandrasekhar, J. D. Madura, R. W. Impey, M. L. Klein, *J. Chem. Phys.* **1983**, *79*, 926.
[36] J. Ryckaert, G. Ciccotti, H. J. Berendsen, *J. Comp. Phys.* **1977**, *23*, 327.
[37] D. A. Case, V. Babin, J. T. Berryman, R. M. Betz, Q. Cai, D. S. Cerutti, T. E. Cheatham, Ill, T. A. Darden, R. E. Duke, H. Gohlke, A. W. Goetz, S. Gusarov, N. Homeyer, P. Janowski, J. Kaus, I. Kolossváry, A. Kovalenko, T. S. Lee, S. LeGrand, T. Luchko, R. Luo, B. Madej, K. M. Merz, F. Paesani, D. R. Roe, A. Roitberg, C. Sagui, R. Salomon-Ferrer, G. Seabra, C. L. Simmerling, W. Smith, J. Swails, R. C. Walker, J. Wang, R. M. Wolf, X. Wu, P. A. Kollman, AMBER14, University of California, San Francisco, **2014**.
[38] S. Kumar, J. M. Rosenberg, D. Bouzida, R. H. Swendsen, P. A. Kollman, *J. Comput. Chem.* **1992**, *13*, 1011.
[39] J. D. Chodera, W. C. Swope, J. W. Pitera, C. Seok, K. A. Dill, *J. Chem. Theory Comput.* **2007**, *3*, 26.
[40] W. Kabsch, C. Sander, *Biopolymers* **1983**, *22*, 2577.

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