Are Little Terns *Sternula albifrons* breeding in Okinawa and East Japan different population?

M.Hayakawa[a], T.Fujii[b], R.Murofushi[b], W.Kitamura[cd], M.Suzuki[e], K.Matsubara[f], S.Kanazawa[e], A.Moriyama[g]

a: Uekusa Gakuen Univ. b: Japanese Society for Preservation of Birds c: Tokyo City Univ. d: Little Tern Project e:Nagoya City Univ. f: Kwansei Gakuin Univ. g: Chubu Univ.

INTRODUCTION

Little Tern is a migratory bird and listed as a Vulnerable (VU) species on Red List of Japanese Ministry of the Environments. Maintenances of breeding sites, wintering sites, stopover sites are necessary for the protection of the migratory bird.

We have revealed, using geolocators, that Little Tern's population breeding in East Japan (Tokyo, Chiba) migrate to Philippines, Indonesia, Papua New Guinea, Southeastern Australia via Okinawa and Taiwan. In addition, we have heard that the population bred in Taiwan wintered in northwestern Australia, (personal data by Miss Chang in Taiwan). Then, where do Little Terns breeding in Okinawa which locates between East Japan and Taiwan winter?

Final aim of this study is to reveal geographical relation of Little Tern between the populations in Japan and in east Asia using the combinatorial analyses of their migratory route and genetic affiliation. And this will mean the need of the reexamination of the protection policy of Little Tern.

solid line
Migration routes Ⅰ (from East JAPAN)
→ East Australia, Indonesia, Papua New Guinea, Philippines, et al.

dotted line
Migration route Ⅱ (from Taiwan)
→ North-west Australia

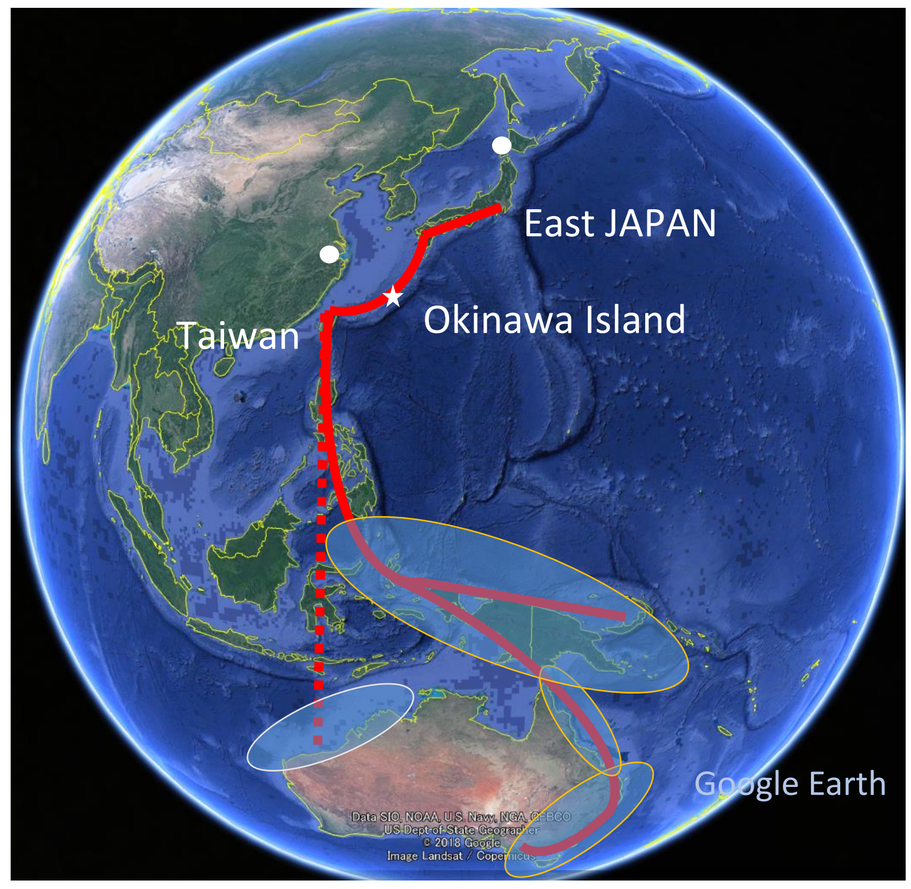

Fig.1 Difference of the migration routes of East Japan breeding population and the Taiwan breeding population.

MATERIALS & METHODS

1 **GPS & geolocator analysis:** To research migration routes, we used 8 GPSs (V3177Pin-point10 or V4291PP-SOB-10, Migrate Tec. Co.) in 2017 and 20 geolocators (Intigeo-W65A9, Bio Track Co.) from 2015 to 2017 in Okinawa.

2 **DNA analysis:** We sequenced mitochondrial D-loop domain for 47 individuals from the 4 sites (Chiba, Tokyo, Fukuoka and Okinawa), and constructed haplotype networks by Popart to assess genetic difference between each populations.

Fig.2 Geolocator that attached to leg.

Fig.3 GPS worn in waist.

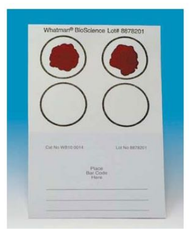

Fig.4 FTA card for Blood sample collection. (pic. By Lab Shop)

RESULTS

GPS & geolocator analysis: We couldn't recover the GPS. And we could recover only one geolocator. The Little Tern which bred in Okinawa wintered in the eastern part of Australia.

Fig.5 Migration route of Little Tern which bred in Okinawa.

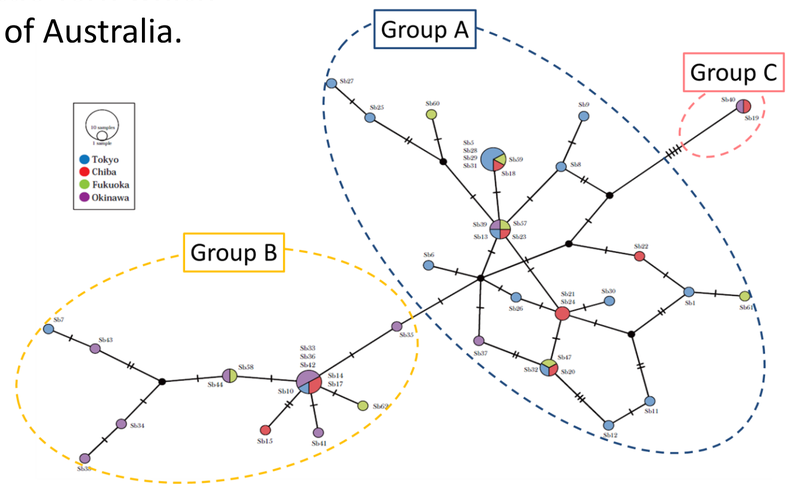

Fig.6 Haplotype Network of the mtDNA D-loop domain.

The network for the mtDNA D-loop domain (1029bp) from 47 samples was constructed with the TCS method implemented in PopART. On this network, haplotype at the remote position means far genetically. The population that bred in Japan were shared to 3 groups (A, B, C).

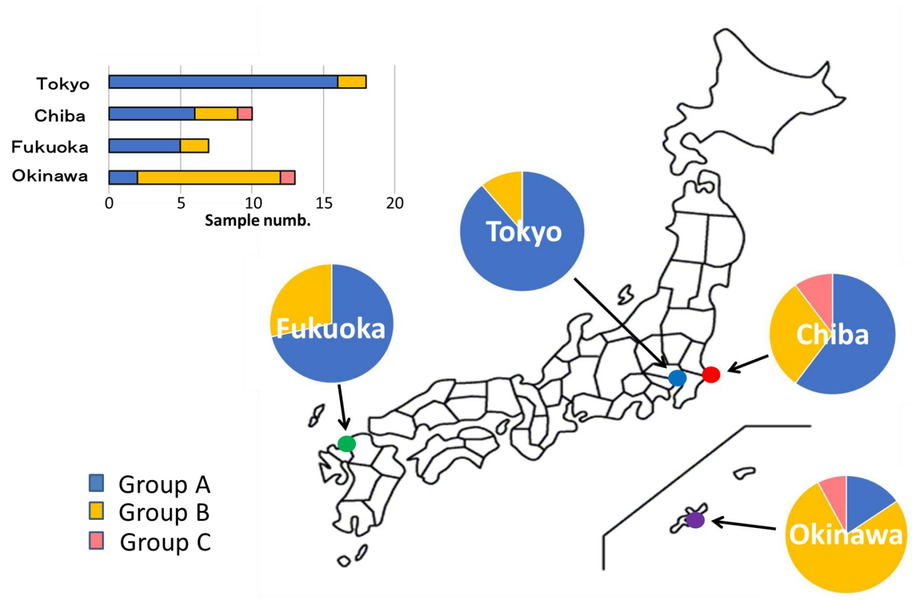

Fig.7 Distribution of mtDNA D-loop domain haplotype in each domestic nesting site.

Each group and color in nesting site support figure 6. The haplotype of the mtDNA D- loop domain showed a different genetic structure between 4 domestic nesting sites. There were many B types in Okinawa whereas a tendency with many A types was seen in the other places including East Japan.

Table1 Index of the genetic divergence (Fst) between groups.

	Tokyo	Chiba	Fukuoka	Okinawa
Tokyo	0	-	-	-
Chiba	-0.0057	0	-	-
Fukuoka	-0.0295	-0.0803	0	-
Okinawa	0.2260 (p<0.05)	0.0954 (p<0.05)	01382 (p<0.05)	0

DNA analysis: We analyzed the genetic structure in the population of each nesting site. 29 haplotypes from the mtDNA D-loop domain of 47 samples which we collected in 4 domestic nesting sites were detected. Genetic haplotype of Little Terns which nested in Japan was greatly divided into 3 groups, and it was suggested by haplotype network that genetic structure of the group of Okinawa was different from the other places (Tokyo, Chiba, Fukuoka). In addition, only population of Okinawa varied in a genetic structure than population of the other places, and the possibility that formed a sub-group was suggested by the comparison of Fst and the base variation site.

DISCUSSION

It was revealed by geolocator's data that the breeding individual in Okinawa wintered in the eastern part of Australia like individuals breeding in other Japanese sites. However, the possibility that an individual is to migrate to northwest Australia is thought, because the obvious migration route of the Little Tern is only one case. In addition, the population to breed in Okinawa include the possibility that a wintering site is different because having possibilities to form a sub-group genetically.

We try to recover the GPSs and more geolocators in next year to get further insights to assess migration routes of the domestic populations.

ACKNOWLEDGEMENT

This work was supported by JSPS KAKENHI (15K06935), The Mitsui & Co. Environment Fund(R15-0004).
Cooperation: Little Tern Project, Kenji Takehara, Takurou Hattori, Tatsuo Sato, Toshimitsu Nuka, Yoshimitsu Shigeta, Yutaka Tokuchi.

Contact information: Masaharu Hayakawa: m-hayakawa@uekusa.ac.jp
Takashi Fujii: fujii@jspb.org