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GENETIC INVESTIGATION OF GREEN TURTLES (*CHELONIA MYDAS*) HARVESTED FROM A FORAGING GROUND AT MANTANANI, SABAH, MALAYSIA

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***Abstract.*—In recent years, harvesting of sea turtles at foraging grounds in Southeast Asia has increased. However, few studies have been conducted at foraging grounds and it is not known which nesting populations are affected by this exploitation. Using mixed-stock analysis of mitochondrial DNA (mtDNA) control region sequences, we aimed to determine the natal origin of Green Turtles poached from the foraging ground at Mantanani Island, northwest of Sabah, Malaysia. Six mtDNA haplotypes were revealed (D2, C14, C8, C4, C3), including one previously undescribed haplotype. When compared with other foraging grounds in this region, the genetic diversity indices of the carcass samples were h = 0.8474 ± 0.04 and π = 0.0191 ± 0.01. Mixed-stock analysis suggests the possible source populations for the Green Turtle carcasses were from Aru, Sulu Sea, Gulf of Carpentaria, Berau, Sarawak and North-west Shelf. Despite the small sample size, our results highlighted the importance of protecting foraging grounds in Malaysia as exploitation at these areas will affect nearby and distant nesting populations of Green Turtle in Southeast Asia and the Western Pacific. Clearly, enforcement and collaboration, both regional and multinational are urgently needed in order to defeat the sea turtle poaching activities in this region.**

Key Words.—haplotypes; mitochondrial DNA; mixed-stock; poaching

INTRODUCTION

Sea turtles are known to move extensively between nesting and foraging grounds, but in most cases, for example in Malaysia, it is not known which reproductive populations occupy a particular foraging habitat. Studies have shown that nesting sea turtles in Malaysia conduct long distance migrations. For example, Luschi et al. (1996) reported that after the breeding season, Green Turtles (*Chelonia mydas*) from Redang Island, Terengganu, migrated to Sabah (Malaysia), Indonesia and Philippine waters. The nesting Green Turtles tracked from the Sarawak Turtle Islands migrated northwards along the Borneo coast into the Sulu-Sulawesi seas (Liew et al. 2000). Foraging grounds for sea turtles have been identified in Malaysia at Lawas, Sipadan Island and Mantanani. However, the nesting populations that contribute to each of these foraging grounds have not previously been identified. Determining linkages between foraging and nesting grounds is crucial, but it can be very challenging for sea turtles as they exhibit distinct migratory behavior throughout geographically different habitats (Godley et al. 2003). As hatchlings, they enter a pelagic life-history stage for 10 or more years, later recruiting as juveniles to near shore feeding habitats (Bolten and Balazs 1995). They may distribute to different feeding grounds due to food and habitat requirements (Hirth 1980). Once mature, they will return to the area of their birth to nest, and this is known as natal homing (Carr 1967). The wide migration range of hatchlings, juveniles and adult turtles provides opportunities for mixing stocks at the foraging grounds (Bowen and Karl 1997).

Exploitation and trade of sea turtle products is a widespread phenomenon across the Southeast Asian region (Davenport 1988; Shanker and Pilcher 2003; van Dijk and Shepherd 2004; Stiles 2008). Sea turtles are illegally harvested at their foraging grounds in Southeast Asia, especially in Malaysia, Indonesia, and the Philippines, where most of the poaching activities are conducted by Chinese and Vietnamese fishermen (Pilcher et al. 2008). The turtles are caught either by hunting or through incidental catch, and are traded for their meat, shell, and eggs. Though most countries in Southeast Asia have instituted regulations to protect and prevent the harvesting of sea turtles to arrest population declines, poaching is still rampant. Some of the major markets where these turtle products end up are in China, Japan, Vietnam, Taiwan, and Indonesia (Barr 2001; van Dijk and Shepherd 2004; Stiles 2008; Chen et al. 2009; Lam et al. 2011). In Malaysia, poaching has been reported since 2003 (Hassan Omar, unpubl. report); however, the biggest reported poaching incident occurred at Mantanani and Mengalum Islands (northwest of Sabah) in 2007 by fishermen from Hainan, China.

As sea turtle research and conservation in Malaysia concentrate mainly on nesting beaches, there is limited knowledge available about the turtle stocks occupying the foraging grounds throughout Malaysian waters. The only study conducted at foraging grounds was on the population structure and growth rate of juvenile Green Turtles (Pilcher 2010). Up to now, it is not known whether those illegally harvested sea turtles at the northwest of Sabah belonged to the Malaysian nesting populations or to those of neighboring countries. Studies elsewhere have suggested that sea turtles at a given foraging ground often come from various nesting colonies (e.g., Bowen et al. 1996; Dethmers et al. 2010; Amorocho et al. 2012; Nishizawa et al. 2013) hence poaching of sea turtles at foraging grounds in Malaysia might affect several nesting populations in the region.

In the absence of satellite or radio-biotelemetric data to track migratory pathways, molecular genetic markers have proven useful in resolving migration patterns of sea turtles (Bowen and Karl 1997). In this study, we used mitochondrial DNA (mtDNA) control region sequences to determine and identify Green Turtle stocks from the 2007 poaching incident at Mantanani. The natal origin of sea turtles at feeding grounds can be determined because studies of mtDNA variations in nesting populations of Green Turtles have been conducted at almost all the major nesting sites in this region. Genetic isolation among nesting colonies, and corresponding mtDNA haplotype frequency differences, provide opportunities to identify natal origins of sea turtle samples taken from foraging grounds (Bowen and Karl 1997) or from illegal harvests. This study is the first attempt to determine the natal origin of Green Turtle stocks occupying a foraging ground at Mantanani, Sabah using carcass samples. Information on the natal origin of sea turtles has important conservation implications because the mortality caused by poachers around Mantanani Island may affect breeding populations elsewhere in this region. Knowledge obtained from this study will be especially useful to improve regional conservation management strategies and to overcome the illegal poaching activities.

MATERIALS AND METHODS

Sample collection.—We used Green Turtle carcasses from the first vessel confiscated near Mantanani Island (6.71°N, 116.35°E) during the 2007 Hainan poaching incident (Fig. 1). We obtained 20 samples in 2009 from the Department of Fisheries Sabah and University Malaysia Sabah. There was a delay in obtaining the samples as they were used by authorities for the court case, and were released only after the case was settled. The carcass samples consisted of juvenile Green Turtles with curved carapace length ranging from 35 cm to 70 cm. We obtained tissue samples from flippers and preserved them in ethanol to prevent further DNA degradation.

Laboratory analysis.—We conducted genetic analysis at University Malaysia Terengganu. We extracted DNA using QIAGEN DNeasy tissue extraction kit. A 380 bp of the mtDNA control region fragment was amplified using primers TCR5 and TCR6 (Norman et al. 1994). We performed Polymerase Chain Reaction (PCR) amplification in a total volume of 50 µL using a BIO-RAD DNA Engine Peltier thermal cycle. We amplified template DNAs in 50 µL total reaction volumes containing 25–50 ng of turtle genomic DNA, 1 µ/ 50 µL Taq Polymerase (Bioline), 10 mM Tris-HCl buffer, 2.5 mM $MgCl_2$, 0.125 mM deoxynucleotide triphosphates (dNTPs) and 0.2 µM of primer TCR5 and TCR6. Cycling parameters consisted of an initial pre-denaturation at 94° C for 2 min, followed by 30 cycles of 30 s denaturation at 94° C, annealing at 55° C for 35 s and extension at 72° C for 30 s, followed by a final elongation step at 72° C for 2 min. Following PCR, we ran all amplified samples on 1% agarose gel electrophoresis to check for correct sizes. The successful PCR products were then purified using a commercial PCR purification kit (Promega) and sent off to First Base, an outsourced company for sequencing.

Statistical analyses.—We checked the mtDNA sequences using Codon Code Aligner. It was then edited and aligned using Eyeball Sequencing Editor (ESEE3; Cabot 1995). We aligned and compared sequences to reference sequences to identify haplotypes. For determination of turtle stocks, we compared haplotypes to published haplotypes from nesting populations around Southeast Asia and Western Pacific (Dethmers et al. 2006), Taiwan (Cheng et al. 2008), and Japan (Nishizawa et al. 2013). We also referred to mtDNA sequences of Green Turtles of the Pacific and Indian Ocean from the Southwest Fisheries Science Center, NOAA Fisheries Service (NOAA Fisheries, Southwest Fisheries Science Center. 2014. Marine Turtle Research Program. Available from https://swfsc.noaa.gov/textblock.aspx?Division=PRD&id=1226&ParentMenuId=212. [Accessed 14 July 2014]). We also searched the GenBank database (National Center for Biotechnology Information. 2014. Genbank. Available from http://www.ncbi.nlm.nih.gov/genbank/. [Accessed 14 July 2014]) for control region sequences for comparison. We used the program ARLEQUIN 3.1 (Excoffier et al. 2005) to estimate haplotype diversity and nucleotide diversity of the carcass samples and other foraging grounds identified previously in this region. Exact tests of population differentiation (Raymond and Rousett 1995) were employed to assess differences of the Mantanani samples with other foraging grounds

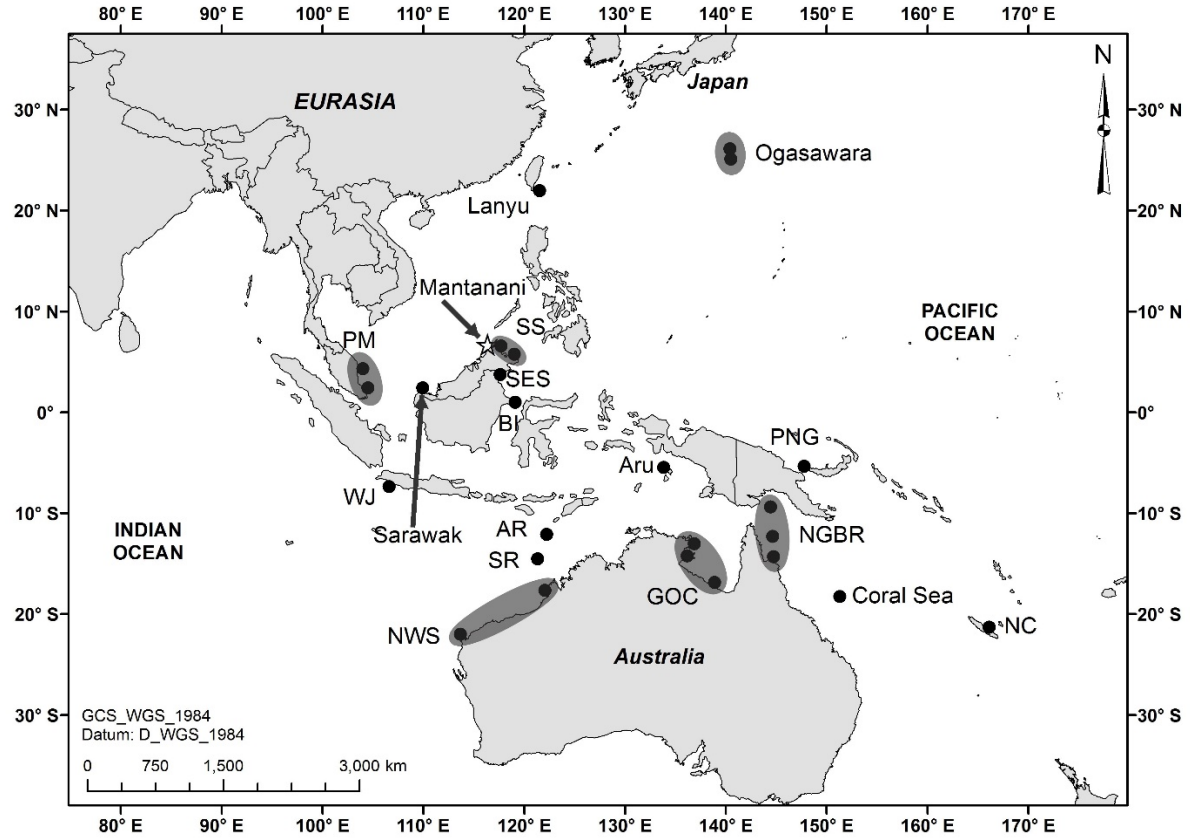

FIGURE 1. Location of Mantanani foraging ground of Green Turtles (*Chelonia mydas*) and the 17 genetically distinct breeding populations from previous studies (Dethmers et al. 2006; Cheng et al. 2008; Nishizawa et al. 2013): Northern Great Barrier Reef, (NGBR), Coral Sea, New Caledonia (NC), Papua New Guinea (PNG), Gulf of Carpentaria (GOC), Aru, Ashmore Reef (AR), Scott Reef (SR), North-west Shelf (NWS), Berau Islands (BI), Southeast Sabah (SES), Sulu Sea (SS), Sarawak, Peninsular Malaysia (PM), West Java (WJ), Lanyu and Ogasawara.

identified by Dethmers et al. 2010 and Nishizawa et al. 2013.

We determined the proportional contributions of the original stocks to the Green Turtle carcass samples using Bayesian mixed-stock analysis (MSA) as implemented in the program BAYES (Pella and Masuda 2001) to estimate a single mixed stock and set of source population. We performed two MSA approaches on the Mantanani samples: the uniform prior probabilities (MSA_1) and informative prior (MSA_2) weighted with the size of each possible source population following Dethmers et al. (2010). In the MSA analysis, we used 17 chains to run each of the contributing stock and we ran 50,000 Markov Chain Monte Carlo (MCMC) for every chain. Each chain was started with 95% contribution from one of the potential rookeries of origin and a burn-in of 25,000 runs to calculate the posterior distribution of all chains combined. We used the Gelman and Rubin shrink factor to check the convergence of each chain. Value of the shrink factor < 1.2 indicated convergence (Pella and Masuda 2001). We removed any novel haplotypes from these analyses that were not previously detected at the contribution stocks. The source populations were from 17 genetically separated rookeries (Fig. 1) previously reported for the Southeast Asia and Western Pacific (Dethmers et al. 2006), Taiwan (Cheng et al. 2008), and Japan (Nishizawa et al. 2013). The rookeries included the Northern Great Barrier Reef, Coral Sea Platform, New Caledonia, Papua New Guinea, Gulf of Carpentaria, Aru, Berau Island, Southeast Sabah (Sipadan), Sulu Sea, (Philippines / Sabah Turtle Islands), Sarawak, Peninsular Malaysia, Ashmore Reef, Scott Reef, West Java, North-west Shelf, Ogasawara, and Lanyu.

RESULTS

Haplotypes composition.—We detected six haplotypes from the 20 Green Turtle samples (Table 1). Among the six haplotypes, five haplotypes, C3, C4, C8, C14, and D2 (known in SWFSC haplotype nomenclature as CmP49, CmP87, CmP89, CmP91, and CmP57, respectively), were previously identified by Dethmers et al. (2006) for the Southeast Asia and Western Pacific

TABLE 1. Frequencies of Green Turtle (*Chelonia mydas*) mtDNA haplotypes from Mantanani foraging ground (FG) and 17 possible nesting rookeries from Southeast Asia and the Western Pacific (Dethmers et al. 2006), Taiwan (Cheng et al. 2008), and Japan (Nishizawa et al. 2013): Northern Great Barrier Reef, (NGBR), Coral Sea (CS), New Caledonia (NC), Gulf of Carpentaria (GOC), Ashmore Reef (AR), Scott Reef (SR), North-west Shelf (NWS), Papua New Guinea (PNG), Aru (AI), Berau Islands (BI), West Java (WJ), Sulu Sea (SS), Southeast Sabah (SES), Sarawak (SW), Peninsular Malaysia (PM), Ogasawara (OG) and Lanyu (LY). MG5 is a novel haplotype found in Mantanani and was excluded from the MSA.

	Nesting Rookeries																	
Haplotype	NGBR	CS	NC	GOC	AR	SR	NWS	PNG	AI	BI	WJ	SS	SES	SW	PM	OG	LY	FG
A1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A2	2	30	2	0	1	1	1	0	0	0	0	0	0	0	0	0	0	0
A3	0	0	0	1	9	0	0	16	1	0	0	0	0	0	0	0	0	0
A4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A6	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
B1	42	0	4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
B3	2	9	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
B4	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
B5	0	0	0	0	0	0	0	0	0	0	0	0	0	2	3	0	0	0
C1	0	1	0	65	3	5	36	0	0	0	0	0	0	0	0	0	0	0
C2	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
C3	0	1	1	45	7	11	3	1	0	7	17	13	18	2	22	4	14	2
C4	1	0	0	0	0	0	0	0	0	0	0	0	0	18	1	0	0	2
C5	0	0	0	0	0	0	0	0	0	9	6	1	3	0	0	0	0	0
C7	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
C8	0	0	0	20	0	0	0	0	0	0	0	0	0	0	0	0	0	4
C9	0	0	0	0	0	2	2	0	0	0	0	0	0	0	0	0	0	0
C12	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
C13	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
C14	0	0	0	1	0	0	1	0	27	5	0	0	1	0	1	0	0	5
D2	0	0	0	0	0	0	0	0	0	7	0	53	8	0	0	0	0	5
E1/E2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
J1	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
J2	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
CMP50	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	13	0	0
CMP39	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	53	0	0
CMP54	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
CMP53	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0
CMJ19	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	12	0	0
CMJ15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0
CMJ16	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	0	0
CMJ20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0
CMJ38	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0
CMJ32	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0
CMP95	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0
CMP37	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0
MG5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2
Total	52	41	10	132	20	19	45	18	28	29	23	67	30	22	27	103	12	20

Green Turtles. The remaining one haplotype, MG5 (Genbank accession number: KM62224) was detected for the first time and has never been described elsewhere. We found haplotype D2 (25%) and C14 (25%) to have the highest percentage in the carcass samples, followed by C8 (20%). Haplotypes C4, C3 and MG5 have 10% occurrence in the carcass samples. We estimated the genetic diversity of the Mantanani samples as $h = 0.8474 \pm 0.04$ and $\pi = 0.0191 \pm 0.01$, and are quite high compared to the other foraging grounds (Table 2). Exact tests revealed that the Mantanani samples were highly differentiated from the previously reported foraging grounds in this region (exact $P < 0.001$).

Mixed stock analysis.—Overall, the MSA resultsshowed the Green Turtle carcass samples fromMantanani were derived from multiple nesting populations (Table 3). The MSA for the uniform and weighted priors showed different results because of the small sample size used in this study. Based on uniform prior, we estimated that the main contributors are from Aru (mean = 25%), Sulu Sea (mean = 24%), and Gulf of Carpentaria (mean = 23%). We observed small contributions from Sarawak (mean = 11%) and Berau (mean = 9%). MSA results, based on weighted prior, estimate lower probabilities and the main contributors are from Gulf of Carpentaria (mean = 16%), Sulu Sea (mean = 13%), Aru (mean = 10%) with small contributions from Berau (mean = 5%), North-west Shelf (mean = 4%), and Sarawak (mean = 3%).

TABLE 2. Genetic diversity of the carcass samples of Green Turtles (*Chelonia mydas*) from Mantanani compared with other foraging grounds in Southeast Asia., Western Pacific (Dethmers et al. 2010), and Japan (Nishizawa et al. 2013).

Feeding Grounds	Haplotypes	Haplotype diversity *(h)* ± SD	Nucleotide diversity (π) ± SD	Sample size
Mantanani	6	0.8474 ± 0.04	0.0191 ± 0.01	20
Cocos Keeling Islands	3	0.4524 ± 0.07	0.0011 ± 0.00	36
Ashmore Reef Feeding	12	0.6139 ± 0.05	0.0136 ± 0.01	65
Fog Bay	12	0.7707 ± 0.04	0.0202 ± 0.01	67
Field Island	17	0.7472 ± 0.05	0.0228 ± 0.01	62
Cobourg Peninsula	15	0.7849 ± 0.03	0.0294 ± 0.01	91
Aru Island Feeding	8	0.7218 ± 0.06	0.0338 ± 0.02	40
Sir Edward Pellew Island	7	0.6428 ± 0.03	0.0100 ± 0.00	102
Yaeyama	22	0.8355 ± 0.02	0.0490 ± 0.02	142
Ginoza	9	0.8789 ± 0.04	0.0506 ± 0.03	20
Nomaike	12	0.6913 ± 0.08	0.0275 ± 0.01	38
Muroto	11	0.6316 ± 0.06	0.0235 ± 0.01	60
Kanto	11	0.7438 ± 0.04	0.0310 ± 0.02	47

DISCUSSION

We found six haplotypes (C3, C4, C8, C14, D2, and MG5) from the genetic analyses of the Green Turtle carcass samples from the 2007 Hainan poaching incident. All of these haplotypes except for MG5 were previously reported by Dethmer et al. (2006) to have originated from the nesting populations of Green Turtle from Southeast Asia and Western Pacific. The most common haplotypes we found were D2 and C14. Haplotype D2 was previously reported to occur in high percentage for the nesting populations around the Sulu Sea (Sabah Turtle Islands and Philippines Turtle Islands). Haplotype C14 was previously reported to occur in Green Turtle nesting populations in Indonesia especially those from Aru. Haplotype C8 is a unique haplotype and can only be found in the Australian nesting populations, and a dominant haplotype for Green Turtle nesting at Gulf of Carpentaria (Dethmer et al. 2006). Occurrence of this haplotype in the Malaysian foraging grounds revealed the first evidence of the long distance migration of Green Turtles from Australia to the foraging grounds in Malaysia. Haplotype C4 is reported as the dominant haplotype for the Green Turtles from Sarawak and C3 is the common haplotype for the nesting populations of Green Turtles in Southeast Asia and Western Pacific (Dethmers et al. 2006).

Apart from the five known haplotypes, one novel haplotype (MG5) was found in the carcass samples. The presence of a novel haplotype is common in sea turtle mixed stock analysis, and had been reported in several studies (e.g., Naro-Maciel et al. 2007; Dethmers et al. 2010; Nishizawa et al. 2013). The appearance of novel haplotypes may be due to insufficient genetic studies of some nesting populations in this region, or due to depleted stocks of rare nesting populations that have not yet been sampled (Bolten et al. 1998). Although large areas of nesting rookeries have been sampled for Southeast Asia, Taiwan, Japan, and the western Pacific region, there are still some nesting grounds with low sample size or that have never been investigated (such as from Thailand, Vietnam, and many others) and these may contribute to the missed haplotypes. Therefore, more studies need to be done on the genetic makeup of nesting populations over a larger area to solve for the missing rare haplotypes in this region. Collaboration with local government agencies and non-governmental agencies (NGOs) may help in collecting adequate samples, not only in Malaysia but for the whole of Southeast Asia and Western Pacific region.

The six haplotypes we identified suggest that this area represents a critical site used as feeding or stopover habitat for the Green Turtle stocks in Southeast Asia and Western Pacific. Based on the mark recapture study conducted by Pilcher (2010), Mantanani may be used as a temporary juvenile (1–6 y olds) foraging ground where they settle after the oceanic development phase, before moving to a more productive foraging grounds. It is especially important to protect this area from poachers as the Mantanani foraging ground can help sustain a large number of Green Turtles from different source population in this region.

Overall the MSA (Pella and Masuda 2001) results suggests that the Green Turtle carcass samples from Mantanani represented mixed-stock derived from multiple nesting populations in Southeast Asia and Western Pacific. The range of possible proportional contributions varied among sites. The different MSA approaches (uniform and weighted priors) used resulted in different contributions. Both results support the observation by Bowen et al. (2007) that nesting populations that are large or nesting populations that are

TABLE 3. Bayesian Mixed Stock Analysis of the control region haplotypes of Mantanani Green Turtle (*Chelonia mydas*) using uniform (MSA_1) and informative priors weighted with population size (MSA_2). Possible stocks contributors are in bold.

Nesting Grounds	MSA	Mean (±SD)	2.5%	Median	97.5%
North Great Barrier Reef	MSA_1	0.003 ± 0.013	0.0000	0.0000	0.0364
	MSA_2	0.011 ± 0.029	0.0000	0.0002	0.0959
Coral Sea Platform	MSA_1	0.003 ± 0.012	0.0000	0.0000	0.0340
	MSA_2	0.001 ± 0.007	0.0000	0.0000	0.0058
New Caledonia	MSA_1	0.004 ± 0.015	0.0000	0.0000	0.0389
	MSA_2	0.000 ± 0.000	0.0000	0.0000	0.0000
Papua New Guinea	MSA_1	0.003 ± 0.013	0.0000	0.0000	0.0369
	MSA_2	0.000 ± 0.004	0.0000	0.0000	0.0001
Gulf of Carpentaria	**MSA_1**	**0.231 ± 0.135**	**0.0000**	**0.2358**	**0.4912**
	MSA_2	**0.163 ± 0.155**	**0.0000**	**0.1614**	**0.4773**
Aru	**MSA_1**	**0.249 ± 0.142**	**0.0000**	**0.2455**	**0.5488**
	MSA_2	**0.107 ± 0.168**	**0.0000**	**0.0000**	**0.5258**
Berau Islands	**MSA_1**	**0.097 ± 0.222**	**0.0000**	**0.0000**	**0.8087**
	MSA_2	**0.523 ± 0.391**	**0.0000**	**0.6487**	**0.9938**
Southeast Sabah	MSA_1	0.021 ± 0.075	0.0000	0.0000	0.2847
	MSA_2	0.000 ± 0.004	0.0000	0.0000	0.0000
Sulu Sea	**MSA_1**	**0.243 ± 0.149**	**0.0000**	**0.2524**	**0.5247**
	MSA_2	**0.127 ± 0.167**	**0.0000**	**0.0111**	**0.5174**
Sarawak	**MSA_1**	**0.112 ± 0.087**	**0.0000**	**0.0951**	**0.3209**
	MSA_2	**0.025 ± 0.059**	**0.0000**	**0.0000**	**0.2104**
Peninsular Malaysia	MSA_1	0.008 ± 0.031	0.0000	0.0000	0.0982
	MSA_2	0.001 ± 0.013	0.0000	0.0000	0.0000
Ashmore Reef	MSA_1	0.004 ± 0.017	0.0000	0.0000	0.0458
	MSA_2	0.000 ± 0.003	0.0000	0.0000	0.0000
Scott Reef	MSA_1	0.005 ± 0.018	0.0000	0.0000	0.0549
	MSA_2	0.000 ± 0.003	0.0000	0.0000	0.0000
West Java	MSA_1	0.005 ± 0.019	0.0000	0.0000	0.0549
	MSA_2	0.000 ± 0.004	0.0000	0.0000	0.0000
North-west Shelf	MSA_1	0.003 ± 0.014	0.0000	0.0000	0.0369
	MSA_2	**0.041 ± 0.048**	**0.0003**	**0.0242**	**0.1747**
Ogasawara	MSA_1	0.003 ± 0.013	0.0000	0.0000	0.0365
	MSA_2	0.000 ± 0.000	0.0000	0.0000	0.0000
Lanyu	MSA_1	0.006 ± 0.020	0.0000	0.0000	0.0714
	MSA_2	0.000 ± 0.000	0.0000	0.0000	0.0000

closer would probably contribute more to a feeding population. Hence, these stocks occur in higher proportion compared with other nesting populations. However, the small sample size used in this study resulted in large confidence intervals, and values are therefore considered general indicators of source contribution (Bass et al. 2006; Naro-Maciel et al. 2007; Dethmers et al. 2010). Improvement in the accuracy of the MSA will require larger sample size and sequencing longer fragments. Nesting beaches in Southeast Asia should also be studied in detail to avoid missing haplotypes.

Even though the sample size used in this study was small, the MSA confirmed that the Green Turtle samples from the 2007 Hainan poaching incident were derived from multiple stocks. Our results suggest that exploitation of sea turtles at the northwest of Sabah may affect not only the Malaysian nesting populations but other distant nesting populations such as those in Indonesia and Australia. Our results also support natal philopatry in foraging Green Sea turtles in Malaysia. Effective protection of sea turtles foraging at the northwest of Sabah may enhance nesting populations hundreds or thousands of kilometers away. These findings contribute to the knowledge about sea turtles at the foraging grounds in Malaysia, a region in which limited study has been conducted. This information is especially useful in formulation of regional conservation management strategies, and to strengthen protection through international collaboration and enforcement to defeat illegal harvesting of sea turtles at their foraging grounds.

This study also proved that genetic analysis can be used to investigate illegal harvests of wild animals using degraded carcasses. Previously in Malaysia, all confiscated sea turtle samples were thrown away or destroyed after being used as evidence in a court as it was thought to have no valuable use (Sabah Fisheries Department, pers. comm.). Confiscated wildlife samples should be donated to researchers at universities for further genetic investigations. Despite the degraded condition of the carcasses, some DNA samples can still be obtained and used successfully for mtDNA analysis. Authorities should also make it a standard practice to

collect tissue samples of the confiscated turtles as evidence and properly preserve samples for later DNA analysis for verification.

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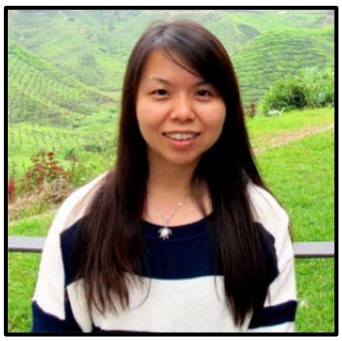

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