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Molecular Identification, Fatty Acid Profile and Trace Elements in a Stranded Fin Whale in Sabah (Borneo, Malaysia): Implications on Migration Routes and Trophic Ecology of Southern Fin Whales

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Fin whales are a cosmopolitan species found in the largest water masses of the world. In Malaysia, as well as other tropical countries in the Southeast Asian region, literature on fin whales is limited, and as a result, there is confusion regarding their distribution range in the region. This study utilizes the fresh tissue of the skin and blubber of a dead fin whale that was stranded in Sabah (Borneo, Malaysia) on the coast of the South China Sea to confirm the species identity, possible properties of the species' diet, and any trace element contamination. The DNA profile results confirmed that the whale belonged to *Balaenoptera physalus*. Further investigation of its cytochrome *b* gene sequence indicated that it was closely related to the southern fin whale (*Balaenoptera physalus quoyi*). This finding indicates that fin whales indeed migrate to warm tropical waters and that their continuous global distribution spans the equatorial region. The dominant fatty acids, such as C18:0, C16:1, C18:1N9T and C16:0 profiles, were consistent with the pelagic plankton diet that the whale would have had during its migration in the tropical waters of the South China Sea. The whales are likely pelagic feeders and thus need to be offshore, which would explain why they are rarely seen in shallow coastal areas during migration in these waters. The concentrations of K, Ca, Sc, Mg and Al ranged from 0.45 μg g^{-1} to 7.80 μg g^{-1}, while Cr, Cd, As and Pb were either very low or could not be detected. This is consistent with concentrations of trace elements previously reported for other baleen whale genera from the Southern Ocean. Our study demonstrates the importance of the South China Sea as a migration route for the southern fin whale, since it is a rich food source with relatively low contaminant levels. The South China Sea is therefore well-suited to ensure these whales' survival during migration.

Key words: Southern Hemisphere fin whale, Migration routes, Tropical waters, South China Sea.

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BACKGROUND

Fin whales (*Balaenoptera physalus*), also known as finback whales or common rorquals, are cetaceans belonging to the parvorder of baleen whales. They are a cosmopolitan species, found in the largest water masses of the world, ranging from the equator to the polar regions (Bose and Lien 1989; Aguilar 2002; Jefferson et al. 2008; Aulich et al. 2019), though they are rarely found near the ice edge (Gambell 1985). The Society of Marine Mammalogy recognises three subspecies of fin whales: *B. physalus physalus* from the Northern Hemisphere, *B. physalus quoyi* from the Southern Hemisphere and *B. physalus patachonica*, a pygmy subspecies also found in the Southern Hemisphere (Clarke 2004; Edwards et al. 2015; Anonymous 2017; Aulich et al. 2019). In addition, some suggest that northern fin whales can be divided into two subspecies themselves, each from a different ocean region (Archer et al. 2013), although this is still under debate (Cabrera et al. 2019). The Northern and Southern Hemisphere subspecies perform seasonal stereotypical migrations each year. During the summer, the respective subspecies will migrate to the polar regions, and in the winter, they will move towards the equator (Lockyer and Brown 1981; Mizroch et al. 1984; Meredith and Campbell 1988; Aulich et al. 2019). However, due to seasonal differences between the Northern and Southern Hemispheres, their movements toward the equator are seasonally separated with no evidence of population overlap in these waters (Mizroch et al. 1984; Edwards et al. 2015; Aulich et al. 2019). To date, the published literature on Northern Hemisphere fin whales is quite extensive covering various aspects of the subspecies (Fujino 1960; Berube et al. 2002; Anonymous 2009; Aulich et al. 2019). On the other hand, published literature is lacking for the subspecies from the Southern Hemisphere (Findlay et al. 2016). Based on sightings and historical catch records, the southern fin whales are larger in size than the other species (Aulich et al. 2019) and are expected to have wider distribution ranges, especially in the tropics.

Although southern fin whales are generally known to migrate out of Antarctica during the winter, the range of this migration is poorly understood due to the difficulty of tracking their movements which are usually in the deep open ocean (Findlay et al. 2016). However, they could reach near middle latitudes, between approximately 21°S–65°S (*e.g.*, Best 2003; Acevedo et al. 2012; Findlay et al. 2016; Aulich et al. 2019). To date, there is no confirmed record of their occurrence in equatorial regions, specifically below the latitude of 21°S. There is limited literature on the occurrence and possible migration routes of fin whales in the waters of Malaysia and other tropical countries of the Southeast Asia region, and no literature confirming the identity of a fin whale in this region. A recent review by Edwards et al. (2015) does not specifically indicate the occurrence of fin whales within these waters, but instead argues that the global distribution of fin whales shows an equatorial hiatus between approximately 20°N and 20°S. This argument is based on the distribution range of fin whales suggested in Mizroch et al. (1984), Meredith and Campbell (1988), Perry et al. (1999), Mizroch et al. (2009), Reilly et al. (2013) as well as environmental envelope modeling by Kaschner et al. (2006), Kesner-Reyes et al. (2012) and one study from an anonymous author (2013a). Given the limited information on fin whales in this region (*e.g.*, Edwards et al. 2015; Aulich et al. 2019), more research is needed to identify their subspecies as well as to understand their migration routes and trophic ecology. This information, including the availability of relevant data from hard-to-obtain fin whale specimens, is indeed crucial to support conservation initiatives. For this purpose, specimens from any source, including those that have been stranded and dead, can be utilized for further study. To date, many in-depth studies have been conducted on stranded and dead baleen whales around the world and have contributed very useful findings (*e.g.*, Domingo et al. 1990; Heyning 2002).

The global decline of the fin whale population is attributable to commercial whaling's severe overexploitation of the Southern Hemisphere subspecies from the 1930s to the 1960s (Findlay et al. 2016). Unfortunately, their populations are expected to continue to decline due to various emerging threats around the world (Mullins 2008; Bogomolni et al. 2010; Peltier et al. 2012 2014; Findlay et al. 2016). This paper presents the findings of a study on a fin whale that was stranded and died in a river in Sabah (Borneo, Malaysia) connected to the South China Sea. To the best of our knowledge, recording a fin whale in this location in the tropics is relatively new and thus is vital for understanding the species' migratory route and feeding habits. This study utilizes the fresh tissue of the skin and blubber of the stranded specimen to confirm the species identity, possible properties of the species' diet, and the presence of contaminants. The species identity was confirmed through molecular analysis to establish DNA profiles and phylogeny. The properties of the diet were assessed through profiling the fatty acids and contaminants that were estimated to be present, based on the concentration of selected trace elements. It is expected that these results could provide vital information on the real identity of fin whale subspecies that may occur in tropical waters. Moreover, their possible migration route and source of diet were also

discussed by comparing the fatty acid profiles among baleen whale species and other fauna of the different regions, as well as the presence of a contaminant in the body to anticipate the level of pollution in their environment.

MATERIALS AND METHODS

Stranding site and collection of tissue samples

A baleen whale about 15.80 m long was stranded and died on the 2nd of August 2012 at the Sitompok River (Lat. 05°34'672"N; Long.115°39'710"E) near Kuala Penyu, a coastal town overlooking the South China Sea on the western shores of Sabah (Borneo, Malaysia) (Fig. 1). Fresh samples of blubber and skin were taken from the dorsal top, approximately next to the dorsal fin of the stranded baleen whale. Attempts to collect extra samples from the other parts of the body were deemed too difficult, as it was submerged at the time of sampling. All samples were placed in zip-lock plastic bags and kept fresh under ice inside a cooler box until laboratory analysis began.

Molecular analysis

DNA from the whale skin tissue was extracted using the method described by Phillips and Simon (1995). The DNA pellet was resuspended in 100 µl

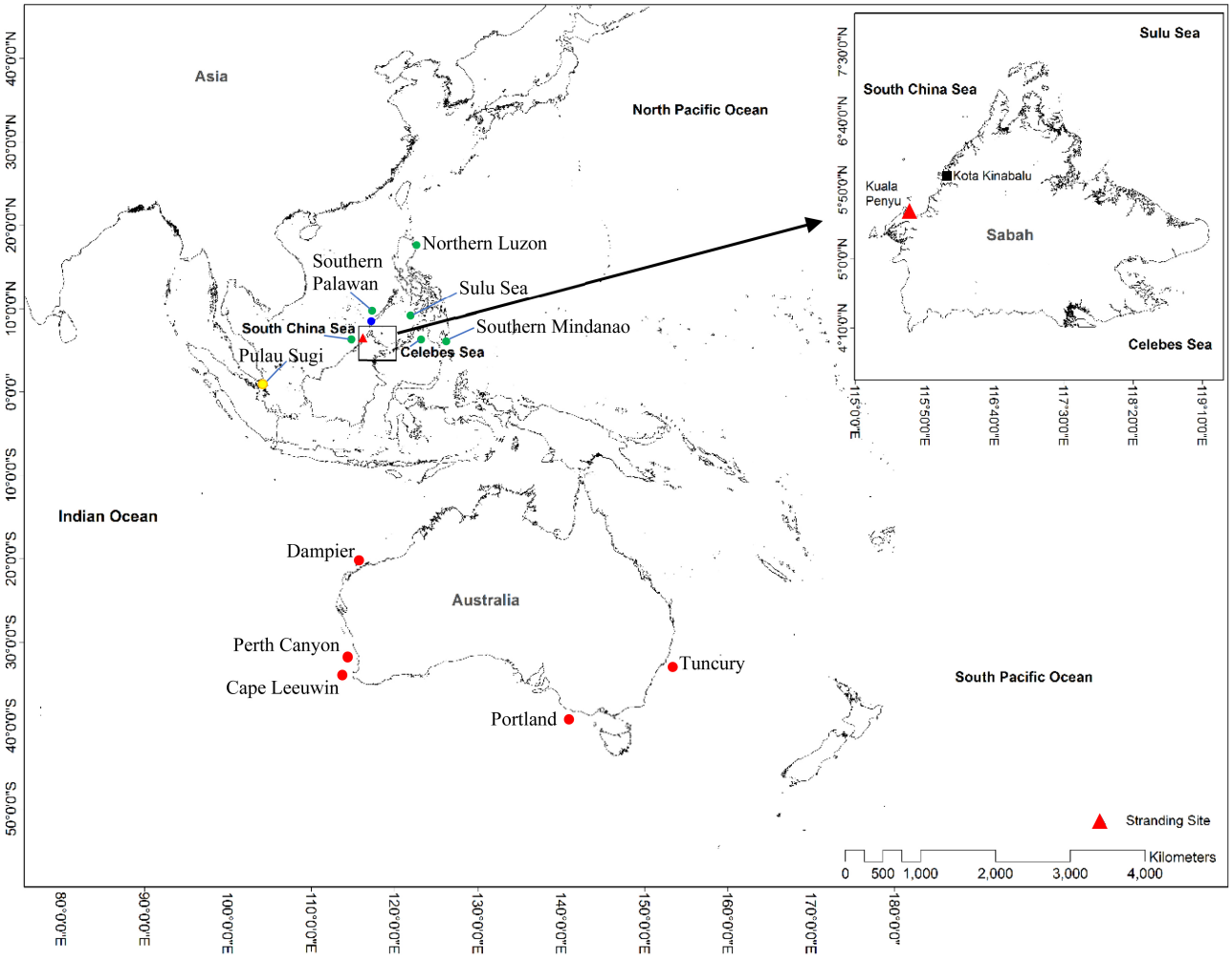

Fig. 1. Stranding site (red-filled triangle) of the fin whale at the Sitompok River (Lat. 05°34'672"N; Long.115°39'710"E) near Kuala Penyu (KP), a coastal town overlooking the South China Sea on the western shores of Sabah (Borneo, Malaysia) (inset map). The approximate location of the sighting of possible fin whales reported by De Boer (2000) is marked with a blue-filled circle. The distribution ranges of rorquals species, including fin whales, in the Philippine waters reported by Slijper et al. (1964) and Acebes (2014) are marked with green-filled circles. The locations of fin whales' migration ranges in Australian waters according to Aulich et al. (2019) are shown using red-filled circles. The stranding site of the unconfirmed fin whale species at Pulau Sugi (Junge 1950) is indicated by a yellow-filled circle.

1X TE buffer and stored at -20°C until used. The amplification of 16S rRNA and cytochrome *b* (cytb) genes was carried out using the polymerase chain reaction (PCR) primers described by George et al. (2011) and Bijukumar et al. (2012), respectively. The PCR cocktails contained 1X PCR Buffer (Promega), 0.4 µM forward and reverse primers, 1U DNA Taq Polymerase and 2 µl DNA sample in a 50 µl reaction. The PCR amplification was performed as follows: initial denaturation at 95°C for 2 min, 30 cycles at 94°C for 30 sec, 55°C (16S) and 46°C (cytb) for 1 min, 72°C for 1 min, and the final extension step at 72°C for 5 min. PCR products were separated and visualized using a 1.5% agarose gel stained with 0.05% RedSafe Nucleic Acid Stain (iNtRON). The PCR products of 16S rRNA and cytb genes were purified using MEGAquick-spinTM PCR and Agarose Gel DNA Extraction Kit (iNtRON, Korea) according to the manufacturer's instructions. Then, the gene fragments were cloned into pGEMT Easy Vector (Promega) and transformed into *E. coli* JM109. The recombinant plasmids harbouring correct DNA fragments were then extracted and purified using DNA-spinTM Plasmid DNA Purification Kit sequencing. The bidirectional nucleotide sequencing was carried out by a service provider (AITbiotech Ptd Ltd, Singapore). The nucleotide sequences of the two gene fragments were analysed using DNAstar Lasergene Version 7. The forward and reverse sequences were assembled using the SeqMan module.

The homology search of the two genes against gene sequences in the gene database was done by using BLAST analysis (https://blast.ncbi.nlm.nih.gov/Blast.cgi). Then, the nucleotide sequences of the two genes were aligned with the same gene sequences from the members of Balaenoptera using the MEGA4 by Clustal W (Thompson et al. 1994) algorithm. The phylogenetic tree was constructed using Neighbour-Joining and Maximum-Likelihood analysis at the bootstrap value of 1000 resamplings.

Fatty acids

Three subsamples each from the skin and blubber were analysed for fatty acids. Step by step procedure for the extraction of lipids using chloroform and methanol (2:1, v/v) was carried out following the method of Folch et al. (1957) with minor modifications. The esterification was performed according to the method by Yoshiraka and Satoh (1989) where hexane was added to the extracted lipids instead of boron trifluoride in methanol, as was commonly used in other procedures. Gas chromatography was then performed on FAMEs using a Shimadzu GC-2010 fitted with a 60 mm × 0.25 mm ID column and coated with a 0.25 µm thick 70% Cyanopropyl Polysilphenylene-siloxane film (Shimadzu Corporation, Kyoto Japan).

After gas-chromatographic analysis, triplicate subsamples were averaged to produce one value for each fatty acid. Fatty acid percentages were divided by the total amount of identified fatty acids in the sample to standardize the data. Only fatty acids with amounts greater than or equal to 1% were considered for the analysis. The total amounts of saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs) were estimated for the skin and blubber, respectively.

Trace elements

The preparation of sample tissue for trace element analysis was conducted following the method of Yap et al. (2003) with slight modifications. Three subsamples of 1 g each from the skin and blubber were weighed, dried, and digested individually in 30 ml concentrated nitric acid following the method by Yap et al. (2002) with slight modifications. They were initially placed in a hot-block digester at 40°C for 1 hr followed by full digestion at 140°C for at least 3 hr. The fully digested subsamples were diluted in 100 ml of deionized distilled water and filtered through GF/C filter paper (47 mm, 0.45 µm, Whatman) in a funnel into Scintillation Vials. The Scintillation Vials containing the resulted solutions were stored in a refrigerator at 4°C until the analysis of trace elements.

All solutions of each subsample were tested for Al, Sr, Pb, Ba, As, Cr, Ti, Cd, Co, U, Ni, Be, Ag, Mg, Fe, Zn, Mn, Cu, Na, K, Ca, Sc, Si, Rb, V, Bi, Ga and Cs using Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES) (Optima 5300DV, Perkin Elmer, USA). The concentration of the trace elements was expressed in µg g^{-1} per wet weight of tissue. All equipment during the analysis was acid-washed with 10% HCl (Analar) and then rinsed with distilled water to avoid possible contamination. A blank sample and certified reference material DOLT-5 from National Research Council Canada were analysed once in every ten samples to check the accuracy of the reading (Yap et al. 2002). The recovery rates were 80 – 117% for Al, As, Cd, Ca, Cr, Co, Cu, Fe, Pb, Mg, Mn, Ni, K, Ag, Na, Sr, U, V and Zn. Additionally, ultrapure water was used as a blank sample to estimate the concentration levels of Ba, Be, Bi, Cs, Ga, Rb, Sc, Si and Ti following the methods of Lee et al. (2017).

Statistical analysis

One-way ANOVA was used to compare the percentage contributions of fatty acids and the

concentrations of trace elements in the skin and blubber of the stranded baleen whales. The percentage data were subjected to arc-sine transformation before statistical analysis. The homogeneity of variances was tested using Levene's test, and the multiple comparisons between skin and blubber were performed using a Tukey HSD post hoc test. The significance level was set at $p < 0.05$. Both tests were performed using SPSS for Windows version 23 (SPSS version 23, IBM, Armonk, NY, USA).

RESULTS

DNA of species

The DNA profile showed that the stranded baleen whale (Baleen whale KP/Sabah/02082012) belonged to the fin whale species of *Balaenoptera physalus* (Fig. 2a). The baleen whale was placed on the phylogenetic tree and clustered together with other fin whales, namely U13103, Z18633 and X61145, based on the DNA sequences deposited into the DNA library downloaded from http://www.ncbi.nih.gov.

Further analysis of the phylogeny of cytochrome *b* gene sequencing (Table S1) indicated that the stranded fin whale was closely related to the DNA of a specimen with accession number KC572845, available at the Dryad Digital Repository (doi:10.5061/dryad.084g8) (Fig. 2b). This specimen was obtained from the Southern Hemisphere (Antarctica) at Latitude 55°.95'N; Longitude 9°.26'E and represented *Balaenoptera physalus quoyi* (see Archer et al. 2013).

Fatty acids

There were 14 fatty acids in both the skin and the blubber with percentage contributions of greater than 1%. These 14 fatty acids consist of six saturated fatty acids, six monounsaturated fatty acids, and only two polyunsaturated fatty acids (Table 1). Both SFAs and MUFAs are the major components of fatty acids in the tissues, with a substantially higher total percentage of contributions than the PUFAs. The total contributions of SFAs were higher in the blubber (55%) than in the skin (46%), but the difference was not significant ($p > 0.01$). In contrast, the total contributions of MUFAs were much higher in the skin (51%) than in the blubber (44%), but again these differences were not significant ($p > 0.01$). The total contributions of PUFAs are low, less than 4% in both skin and blubber.

The dominant SFAs included C18:0 (29%), C16:0 (13%) and C14:0 (9%), while the dominant MUFAs included C16:1 (21%), C18:1N9T (17%) and C18:1N9C (9%) in both the skin and blubber. There were, however, no significant differences ($p > 0.01$) in the percentage contributions of particular fatty acids between the skin and blubber.

Trace elements

As expected, the concentration of sodium (Na) was the highest out of all the trace elements in the whale tissue. The concentration in the blubber (30.11 μg g^{-1}) was two times higher ($p < 0.01$) than in the skin (13.46 μg g^{-1}) (Table 2). This was followed by potassium (K), calcium (Ca) scandium (Sc) and magnesium (Mg) which ranged between 1 to 7.80 μg g^{-1}, though the difference was not significant ($p > 0.01$) between skin and blubber. Aluminium (Al), iron (Fe), zinc (Zn), silicon (Si), rubidium (Rb), strontium (Sr) and vanadium (V) ranged between 0.01 and 0.44 μg g^{-1}, and their concentrations were almost the same for both skin and blubber.

Other elements such as bismuth (Bi), manganese (Mn), barium (Ba), chromium (Cr), titanium (Ti), copper (Cu), cadmium (Cd), cobalt (Co), nickel (Ni), gallium (Ga), silver (Ag) and caesium (Cs) were generally less than 0.01 μg g^{-1}. Among these, copper (Cu), nickel (Ni) and gallium (Ga) were significantly ($p < 0.01$) higher in the blubber than in the skin. Beryllium (Be), uranium (U), arsenic (As) and lead (Pb) were either not present or below the detection limit.

DISCUSSION

The results of DNA analysis and cytochrome *b* gene phylogeny confirmed that the beached baleen whale belonged to *Balaenoptera physalus quoyi*. This proves that fin whales, in particular the subspecies from the Southern Hemisphere, indeed migrate to warm tropical waters. Our findings further support the proposition that fin whales exhibit a continuous global distribution as was suggested in Aguilar (2009) and Anonymous (2013b), although their occurrence in the tropics may be rare. Though the exact purpose of their migration to these waters is not known for sure, this region should be given priority for conservation efforts. This is especially critical since the migration to subtropical or tropical waters may be closely related to their winter breeding season (*e.g.*, Mackintosh 1942; Gambell 1974; Aguilar 2002; Acevedo et al. 2012). However, close monitoring of the exact location of their breeding sites is necessary since they may not occur in shallow coastal waters (Reeves et al. 2002; Jefferson et al. 2008) and are therefore difficult to ascertain. To date, there is no confirmed report of the presence of northern fin whales in the South China Sea, which represents the

tropical Southeast Asian region. Future studies on fin whales, particularly those found in this region, should consider providing a genetic analysis and description of morphology to confirm their species identity given that the presence of several baleen whales has been reported in this area. This is necessary to understand the global distribution of the species and its taxonomy where distribution gaps may suggest reproductively separate

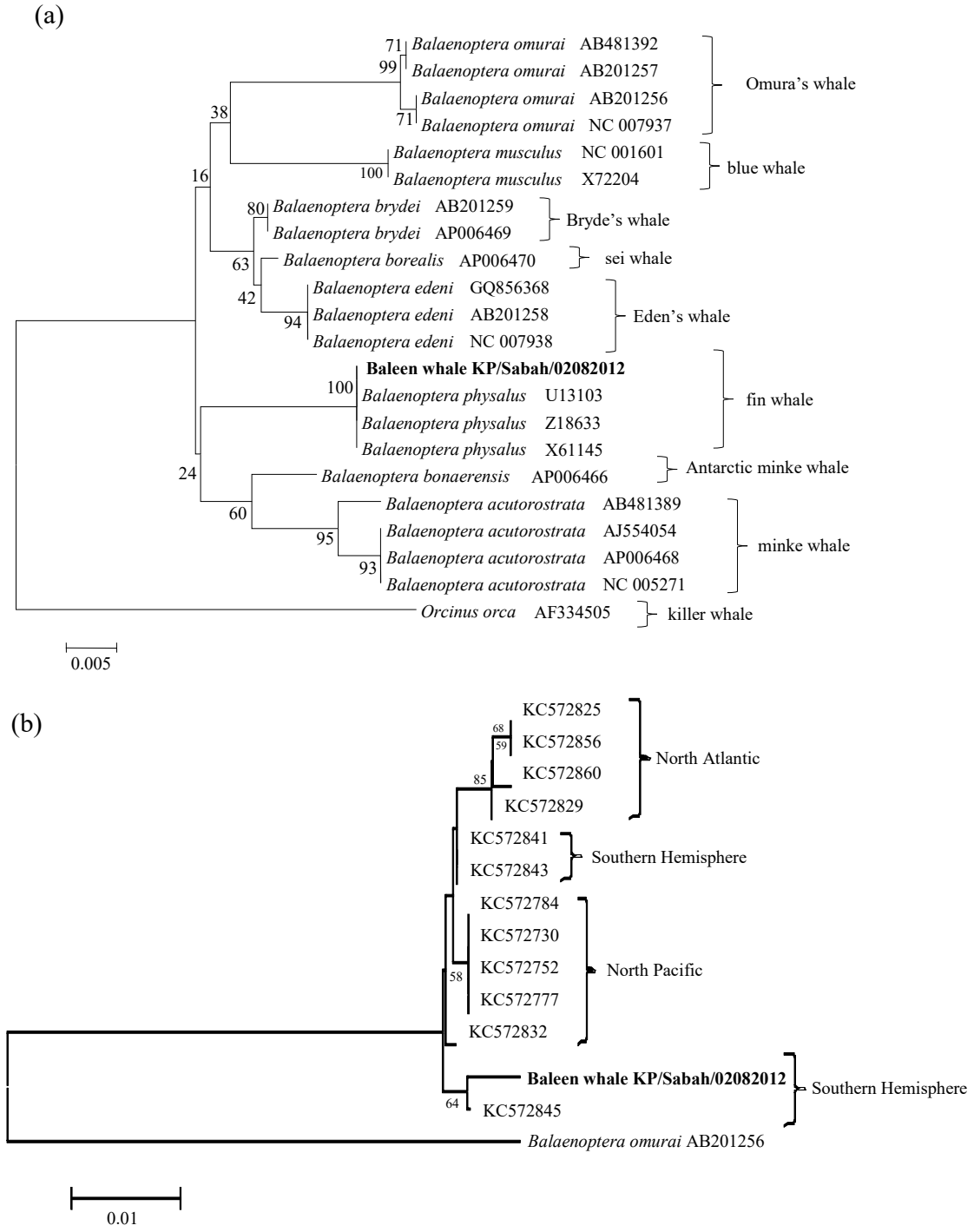

Fig. 2. (a) The phylogenetic tree showing the stranded baleen whale (Baleen whale KP/Sabah/02082012) clustered together with the fin whale *Balaenoptera physalus* (U13103, Z18633 and X61145). (b) The phylogenetic analysis of the cytochrome *b* gene sequence indicating that the stranded fin whale (Baleen whale KP/Sabah/02082012) is closely related to the specimen of fin whales from the southern hemisphere with accession number KC572845, which represents *Balaenoptera physalus quoi.*

populations (*e.g.*, Edwards et al. 2015), especially in less explored equatorial regions.

There have been no previous reports on stranded fin whales, whether alive or deceased in Malaysia's territorial waters. However, other genera of baleen whales, such as the blue whale, Bryde's whale, and sei whale, as well as several species of dolphin, have been reported, particularly within the coastal waters of Sabah, Malaysia (Madin et al. 2013). There are also few records of strandings from other Southeast Asian countries. As far as it is known, one possible case has been reported which occurred on the coast of Pulau Sugi, a small island located between Singapore and the Sumatran coast in July 1936 (Junge 1950). In the most recent work by Mustika et al. (2009), a review of the whale stranding events throughout Indonesian waters from 1987 to 2007 did not mention any fin whale species. This indicates that it is rare to find stranded fin whales, whether alive or deceased, in tropical waters and when they are found, ought to be fully utilized for research purposes. The possible reason for the low number of reported cases is not clear. Nevertheless, fin whales prefer habitats that are located deep offshore in major oceans and rarely venture inshore. This fact would limit the chance of finding any stranded or deceased specimens on the shore. Furthermore, their seasonal presence in the tropical waters may contribute to the few reported cases, especially from Southeast Asian countries (see below).

Although fin whale strandings are rare within Southeast Asian countries, reports of their possible presence in the South China Sea and adjacent waters have been recorded through live sightings. For example, De Boer (2000) reported a sighting of three individuals fin whales during the high-effort survey in April 1999, at Latitude 07.32°N, Longitude 115.45°E of the South China Sea, which is approximately 111 nmi from the Sitompok River, the present stranding site, based on the estimated distance obtained from Google Earth Pro. In Philippine territorial waters, fin whales have been sighted around northern Luzon, southern Palawan, southern Mindanao and the Sulu Sea (Slijper et al.

Table 1. The percentage contribution (%) of fatty acids in the skin and blubber of the stranded southern fin whale

	Skin	Blubber
Fatty Acids	% (Mean ± SD)	% (Mean ± SD)
SFAs		
C14:0	9 ± 1	8 ± 1
C16:0	10 ± 4	13 ± 1
C17:0	2 ± 1	2 ± 0
C18:0	23 ± 12	29 ± 3
C20:0	1 ± 1	1 ± 0
C22:0	1 ± 0	2 ± 0
Total	46 ± 19	55 ± 5
MUFAs		
C14:1	2 ± 0	2 ± 1
C16:1	21 ± 1	20 ± 2
C24:1	1 ± 0	1 ± 0
C18:1N9C	9 ± 3	8 ± 9
C18:1N9T	17 ± 12	11 ± 1
C22:1N9	1 ± 0	1 ± 0
Total	51 ± 16	44 ± 13
PUFAs		
C18:2N6C	1 ± 1	1 ± 0
C18:3N6	2 ± 1	3 ± 0
Total	3 ± 2	4 ± 0

Table 2. Concentrations (μg g^{-1}) of selected trace elements in the skin and blubber of the stranded southern fin whale (nd = not detected)

Elements	Skin (mean ± SD)	Blubber (mean ± SD)
sodium (Na)	13.46 ± 3.00	30.10 ± 5.04
potassium (K)	7.80 ± 1.65	5.80 ± 0.97
calcium (Ca)	3.33 ± 1.09	4.66 ± 1.77
scandium (Sc)	1.87 ± 1.15	1.57 ± 3.23
magnesium (Mg)	1.08 ± 0.13	1.10 ± 0.36
aluminium (Al)	0.44 ± 0.33	0.34 ± 0.10
iron (Fe)	0.28 ± 0.24	0.30 ± 0.23
zinc (Zn)	0.15 ± 0.05	0.12 ± 0.04
silicon (Si)	0.11 ± 0.03	0.12 ± 0.02
rubidium (Rb)	0.06 ± 0.00	0.07 ± 0.00
strontium (Sr)	0.01 ± 0.00	0.10 ± 0.14
vanadium (V)	0.01 ± 0.00	0.01 ± 0.00
bismuth (Bi)	0.00 ± 0.00	0.00 ± 0.00
manganese (Mn)	0.00 ± 0.00	0.00 ± 0.00
barium (Ba)	0.00 ± 0.00	0.00 ± 0.00
chromium (Cr)	0.00 ± 0.00	0.00 ± 0.00
titanium (Ti)	0.00 ± 0.00	0.00 ± 0.00
copper (Cu)	0.00 ± 0.00	0.02 ± 0.02
cadmium (Cd)	0.00 ± 0.00	0.00 ± 0.00
cobalt (Co)	0.00 ± 0.00	0.00 ± 0.00
nickel (Ni)	0.00 ± 0.00	0.00 ± 0.00
gallium (Ga)	0.00 ± 0.00	0.00 ± 0.00
silver (Ag)	0.00 ± 0.00	0.00 ± 0.00
cesium (Cs)	0.00 ± 0.00	0.00 ± 0.00
beryllium (Be)	nd	nd
uranium (U)	nd	nd
arsenic (As)	nd	nd
lead (Pb)	nd	nd

1964; Acebes 2014) (Fig. 1). Past sighting records and the findings from the present study agree with De Boer's (2000) suggestions that the South China Sea and adjacent waters including the Sulu Sea and Balabac Strait are an important migration route for fin whales and other baleen whale genera during a particular time of year. Interestingly, these waters had been within the distribution range of various rorquals species, including fin whales, in the past, based on the history of whaling activities in the Philippines during the nineteenth century (Slijper et al. 1964; Acebes 2014). In Australian waters, Aulich et al. (2019) reported the seasonal arrival of separate fin whale subpopulations at several locations along the western, northern and eastern coastal areas which likely act as a waystation for feeding during migrations (Fig. 1). While there is no confirmed record of fin whale migration outside of Australian waters, such as in the Southeast Asian region, their migration range could reach up to tropical waters including the South China Sea based on the findings of the present study. However, more studies about their migration through tropical waters as well as other areas outside of the Southern Ocean are needed since this phenomenon has not been fully explored (Jefferson et al. 2008; Aguilar 2009; Mizroch et al. 2009; Toro et al. 2016).

There have been no previous studies of fatty acids in southern fin whales from specimens obtained from the Southern Ocean itself or during migration across the South China Sea. However, studies on other species of Southern Ocean baleen whales, such as southern humpback whales migrating in Australian waters, indicate that C18:1N9C was the most dominant fatty acid in the blubber tissues followed by C16:1N7C, C16:0, C18:1N7C, and C20:5N3 (Waugh et al. 2012). This is slightly different from the fatty acid profile of the stranded fin whale where C18:0 was the most dominant followed by C16:1N7C, C18:1N9T, C16:0, C18:1N9C and C14:0 (Fig. 3). It is difficult to determine the cause of this variation as both baleen species adapt to the same diet, the *Euphausia superba*, when they are in the Southern Ocean (Bannister and Hedley 2001; Paterson et al. 2001; Aguilar 2009; Double et al. 2014). However, the differences in diet during migration could be the reason given that the former is known to feed on *Nyctiphanes australis* in Australian waters (Gill et al. 1998; Virtue et al. 1995). Unfortunately, there have been no studies on the diet of southern fin whales during migration, but it is known that they feed on plankton, particularly copepods, in the Southern Ocean (Aguilar 2009) and perhaps during migration, especially in tropical waters. This plankton diet could be the whales' migratory diet given the fatty acid profiles of plankton feeders such as mesopelagic fish from the South China Sea where C16:0, C18:0, C16:1 and C18:1N9 were dominant (Wang et al. 2019) were much closer to the dominant fatty acid profiles in the stranded southern fin whale. In this study, both C18:1N9C and C18:1N9T contributed the highest fractions (*i.e.*, 26% in skin tissue) of the total MUFAs with no contributions from C18:1N7 and C20:1N9 components. Taken together, the C18:1N9 can be attributed to prey feeding on the phytoplankton within the pelagic regions of the South China Sea, and the absence of C18:1N7 and C20:1N9 components suggests this did not extend to the South China Sea coastal regions. These regions supply seston from rivers and bottom resuspension to prey that can be characterized by their high C18:1N7 and C20:1N9 fraction (Wang et al. 2019). This further suggests that the fin whale is most likely a pelagic feeder which is also the reason for their rare sighting in the shallow coastal areas during migration in the South China Sea (Fig. 3). However, more studies are needed to understand the relationship between fatty acids and the whales' diet, as the whales may have a broad spectrum of feeding strategies during migration, as in the case of southern humpback whales (Eisenmann et al. 2016). This is also shown in figure 3 where several other fatty acids (*i.e.*, C20:0, C20:5N3C, C22:6N3C, C22:5N3C) of the southern humpback whales, which are of a minor concentration or not detected in fin whales, suggest that both species have a different diet during migration and possibly the migration route itself.

As far as we know, there have been no other studies of trace elements in the tissues of southern fin whales. However, the concentration pattern is very similar to the findings of Martino et al. (2013) who also studied trace elements from the skin of the southern right whale (*Eubalaena australis*) (Fig. 4), a species known to feed on *Euphausia superba* in the Southern Ocean (Seyboth et al. 2016). Martino et al. (2013) show that nonessential elements such as Al had the highest concentration with an average of 9.75 μg g^{-1} in the tissue (w/w) followed by Au, Sn, Cd, Li, Sb, Ag, Be, Hg, As, Pb, Ni, Ba, Cr, Sr and Ti in the range of below 0.05 μg g^{-1} or not detected. Similarly, in this study, Al was among the highest trace element concentrations, with 0.44 μg g^{-1} (*i.e.*, in the skin), followed by Cd, Ag, Be, As, Pb, Ni, Ba, Cr, Sr and Ti in descending order. Apart from this, Martino et al. (2013) also reported that essential elements such as Mg were the highest, with an average of 187.02 μg g^{-1} in the tissue (w/w) followed by Mn, Cu, Fe and Zn. This is also consistent with the findings of this study where Mg is among the highest concentrations with an average of 1.10 μg g^{-1} (*i.e.*, in blubber) followed by other elements such as Mn, Cu, Fe and Zn. This further indicates that the pattern of trace element concentrations in the Southern Ocean may be quite similar among baleen whale species with the

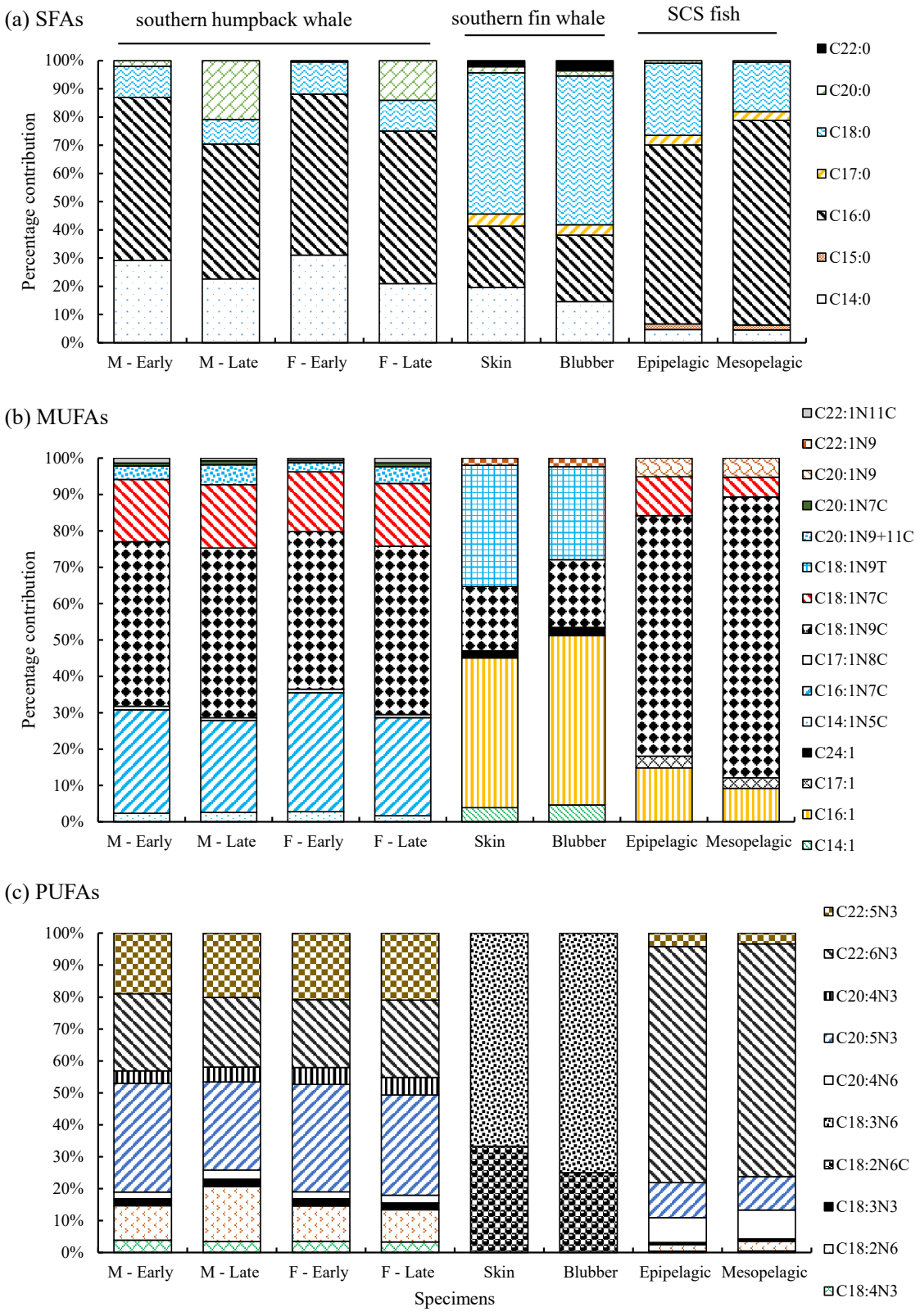

Fig. 3. Comparison of the percentages of fatty acid profiles for (a) SFA, (b) MUFA and (c) PUFA in the tissues of adult male (M) and female (F) southern humpback whales during the early and late migrations extracted from the results of Waugh et al. (2012), epipelagic and mesopelagic (*i.e.*, average) fish in the South China Sea (SCS) extracted from the supplementary data of Wang et al. (2019) and the southern fin whale in the present study.

same diet. However, the actual concentration levels may differ due to the sample preparation protocol as well as the total number of specimens involved. In the present study, samples were only taken at one position of the body of a single-stranded specimen, while Martino et al. (2013) biopsied at least 10 adult female southern right whales.

The concentrations of metal elements in the tissues of the stranded southern fin whales are much lower than those reported for fin whale subspecies in

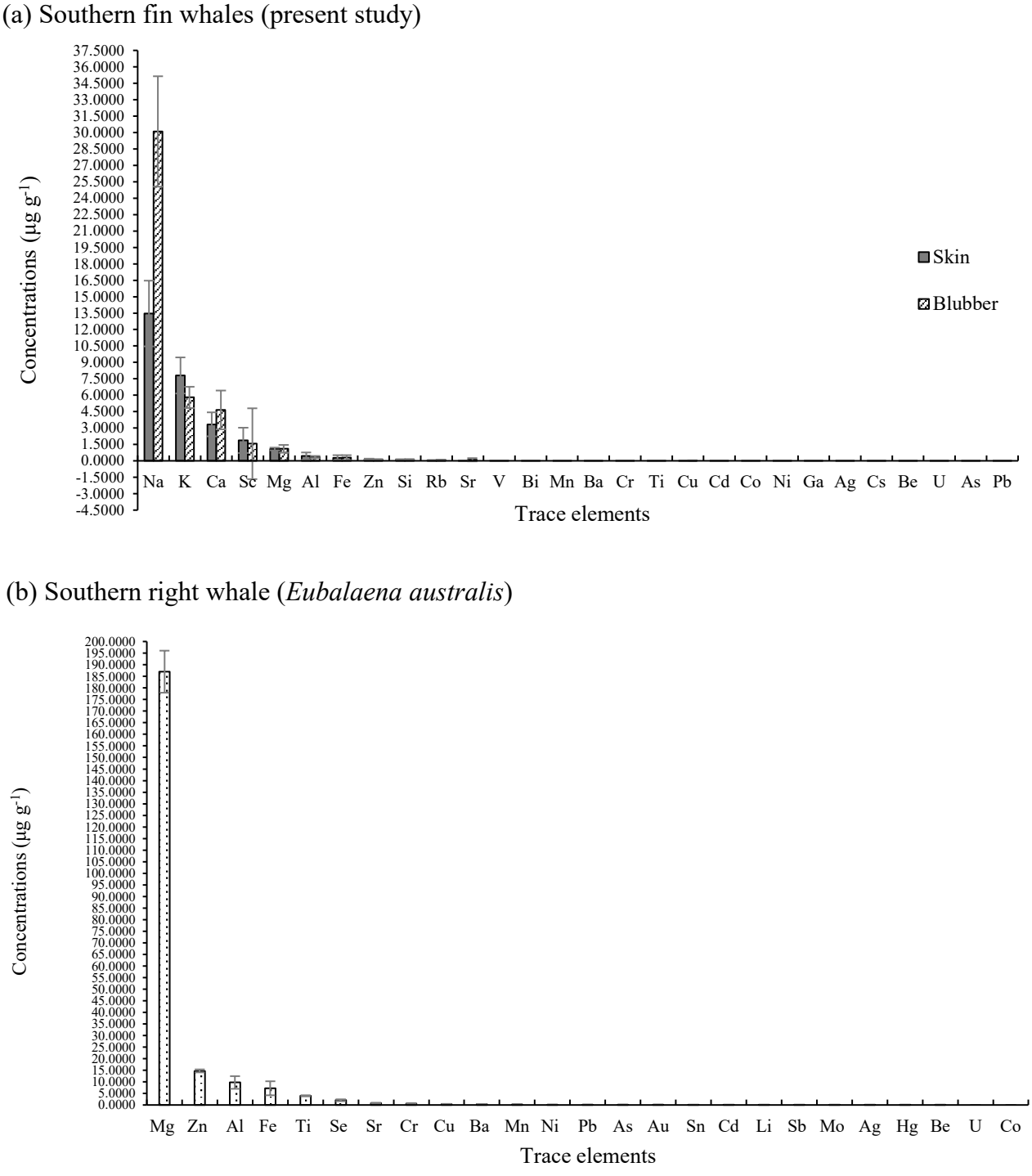

Fig. 4. (a) Concentrations of trace elements (Mean ± SD) in the skin and blubber of the southern fin whale recorded in the present study compared to (b) the concentrations of trace elements in the skin of southern right whales (*Eubalaena australis*) extracted from the results of Martino et al. (2013).

the Northern Hemisphere. For example, Wise Jr et al. (2019), studied the metal levels in the skin of male and female fin whales in the Gulf of Maine and showed that concentrations are several times higher than the concentration recorded in the present study. In their supplementary data, Ti, Cr, Zn, Fe and Mg ranged between 5.01 μg g^{-1}–9.08 μg g^{-1}, 5.16 μg g^{-1}–16.41 μg g^{-1}, 15.68 μg g^{-1}–40.41 μg g^{-1}, 74.06 μg g^{-1}–321.91 μg g^{-1} and 101.55 μg g^{-1}–468.80 μg g^{-1} respectively, while in the present study these are 0.02 μg g^{-1}, 0.30 μg g^{-1}, 1.10 μg g^{-1} and 0.15 μg g^{-1} respectively. Wise Jr. et al. (2019) also reported that metals with low concentration levels, such as Be, U, Ba, As, Cd, V, Pb and Cu ranged between 0.01 μg g^{-1}–0.02 μg g^{-1}, 0.01 μg g^{-1}–0.02 μg g^{-1}, 0.05 μg g^{-1}–0.11 μg g^{-1}, 0.28 μg g^{-1}–0.73 μg g^{-1}, 0.01 μg g^{-1}–0.06 μg g^{-1}, 0.09 μg g^{-1}–1.01 μg g^{-1}, 0.07 μg g^{-1}–1.67 μg g^{-1}, and 0.89 μg g^{-1}–3.78 μg g^{-1}, respectively. However, these were substantially very low (*i.e.*, < 0.01) or undetected in the present study. In another study, Hernandez et al. (2000) reported that the concentration of Cr (1.91 μg g^{-1}), Cd (0.07 μg g^{-1}) and Pb (0.15 μg g^{-1}) in the blubber of the Mediterranean fin whale was substantially higher than the concentration recorded in the present study but comparable with the concentrations reported for fin whales from the Gulf of Maine. Interestingly, Martino et al. (2013) also noted that the average levels of Cr in southern right whales were substantially lower than the concentration reported for North Atlantic right whales. It has been widely suggested that the heavily industrialized coasts of the North Atlantic may contribute to the high concentration of contaminants in baleen whale tissues. Wise Jr. et al. (2019) indicate that heavy coastal development, industry, and marine traffic such as that occurring in the Gulf of Maine could contribute to chronic exposures to environmental chemicals through ingestion, dermal absorption and inhalation that will eventually bioaccumulate in the tissue of baleen whales. The relatively low levels of metal elements in the stranded fin whale tissue could have been due to the whale's pelagic eating habits in the offshore region that minimize the ingestion of contaminated food as well as dermal absorption and inhalation, unlike those in shallow coastal waters. However, studies involving large samples are needed to understand the level of contamination in fin whales in the Southern Ocean to accurately represent the entire population.

CONCLUSIONS

This study is the first to confirm that the subspecies of fin whale known as *Balaenoptera physalus quoyi* or the southern fin whale can cross the tropics, especially the South China Sea and its surrounding waters, during migration. The findings demonstrate a continuous global distribution of fin whales, although their occurrence in the tropics may be rare. Future studies on baleen whales found within these waters should consider the analysis of DNA as well as a complete description of the morphological features to avoid confusion with other baleen whale species. The dominant fatty acids in the tissue of the stranded southern fin whale reveal that they rely on tropical food sources for diet, in particular particular zooplankton, during their migration outside of the Southern Ocean. Although the southern fin whale has a wide migration range and thus may be vulnerable to contamination, the concentration of trace elements in the tissue is generally low and consistent with the results reported for other baleen whale genera from the Southern Ocean. Our findings prove the importance of the South China Sea and adjacent waters as a pristine migration route that is also rich in sources of food for transient baleen whales. Future studies to understand migration season, frequencies and duration are urgently needed to minimize conflicts with ever-increasing human activities in the region.

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Authors' contributions: JM initiated the study and drafted the manuscript. NMHT conducted the FA and TE analysis. MTML and JR conducted the molecular analysis. TY and JBG pre-reviewed the manuscript.

Competing interests: JM, NMHT, MTML, TY and JBG declare that they have no conflict of interests.

Availability of data and materials: DNA sequences (Table S1) of the stranded southern fin whale *Balaenoptera physalus quoyi* were deposited into GenBank with the accession number 'Baleen whale KP/Sabah/02082012'.

Consent for publication: All of the authors agreed to publish the paper.

Ethics approval consent to participate: Sampling of baleen whale tissue for research and teaching purposes was carried out with the permission of the Sabah Wildlife Department (Kota Kinabalu, Sabah) to comply

with the Sabah Wildlife Conservation Enactment 1997.

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Supplementary Materials

Table S1. Information on the Cytochrome *b* gene sequence (472 bp) of a stranded baleen whale (Baleen whale KP/Sabah/02082012). (download)