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A Brief Conservation Medical Comment of Parasitological Surveys on Stranded Whales

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ストランディングしたクジラ類の寄生虫学的調査における保全医学的なコメント

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Introduction

The Wild Animal Medical Center (abbreviated to WAMC; Fig. 1) was established in the animal teaching hospital of Rakuno Gakuen University in 2004 (Asakawa and Taniyama, 2005). The center's mission was to focus on Conservation Medicine, which integrates three scientific disciplines: veterinary medicine (VM), conservation ecology (CE), and human medicine (HM) (Aguirre et al., 2002). The centers efforts are directed primarily in the region surrounding Hokkaido, Japan (Asakawa, 2010). Since 2006, the WAMC has joined the Hokkaido Stranding Network, organized by Associate Prof. Takashi Matsuishi, Hokkaido University, and the Stranded Marine Mammal Research Group of the National Museum of Nature and Science, organized by Dr. Tadasu K. Yamada, Japan. This paper provides an overview of recent parasitological surveys of stranded whales performed by the networking group between 2006 and 2010. This paper originated from the chapter titled "Parasite Studies" in a necropsy manual developed for a training seminar

Fig. 1　A stranded whale carried to WAMC

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(titled "What and how we learn from stranded marine mammals") held by the National Museum of Nature and Science, Shinjuku Branch, Tokyo, Japan, August 30 and 31, 2010.

Parasitic agents of marine mammals

Parasitism is defined as is a trophic association between 2 individuals (or species) in which one (parasite) derives its "food" from another, living organism (host) (Price, 1980). In the case of other benefits (i.e., not nutrition), the counterpart is defined as phoresy (commensal organisms). In general, parasites (or infectious agents, pathogens etc) are divided into three groups:

1) microparasites = virus, bacteria, and fungi
2) mesoparasites = protozoa, e.g., the genera *Toxoplasma/Neospora*, the order Ciliophora
3) macroparasites = helminthes, arthropods, and other animals

This discussion will focus primarily on the animals (parasitic helminthes and arthropods), although half of the animal kingdom's taxa, including Cestoda, Trematoda, Acanthocephala, Nematomorpha, Nematoda, Pentastomida, and Copepoda represent macro parasites (Price, 1980).

Complicating factors during studies of whale parasites

Parasites are ubiquitous in marine mammals and some have direct and/or indirect pathogenicity to a host individual. However, our understanding is very fragmentary relative to that in humans, domestic animals, or even fish. The primary reasons relate to difficulties in quantifying factors related to disease as a phenomena: viz, it is impossible to assess parasitic significance for the host population without the ability to calculate basic epidemiological proportions (e.g., prevalence, incidence, mortality rate, etc). Furthermore, the number of parasites in a host individual is difficult to determine because of the size of the host (whale).

Measuring the long-term effect of parasites is extremely difficult. In particular, the delayed effects may be most important in assessing the parasitic impact to a targeted host population. For example, researchers have detailed microbiological profiles of West Nile virus (WNV) and population biological data for the Japanese crane (*Grus japonensis*), an endangered avian species in Japan. Both national and local governments have funded in situ conservation projects in east Hokkaido to try to forecast the change in the population of cranes as a result of infection with WNV (Onuma et al., 2010). However, we do not have such detailed data describing the parasites or populations of marine mammals.

Samples that are collected for parasitological study from marine mammals are biased by the method of collection and/or body condition. Furthermore, the samples may not represent the actual state in nature. For example, it is well known that grey whales carry large amphipod populations (ca. 5000 per an individual, e.g., http://wildwhales.org/). However, the majority of amphipods are likely lost prior to examination (e.g., the Tomakomai carcass in 2007, Murase et al., in submission). Hence, site/habitat ecological analyses of host-parasite relationship between an individual whale and amphipod species (cf. Kaliszewska et al., 2005) cannot be applied in these instances.

Further complicating matters, minute sized parasites from marine mammal have yet to be described taxonomically. In addition, helminth taxonomists have become an "endangered species". Hence, any parasites obtained from surveys of whales should be held in storage for future identification. Helminth fixation is generally achieved using a 70% ethanol solution. The sample tends to degenerate if the final concentration is below 70% ethanol, so it is best to use a 99% solution in the field. Formalin is a remarkable fixative which pathologists often use for fixing histological specimens. However, formalin should not be used if a researcher is intending to analyze the genetic material in the sample. Formalin solutions should be used at a final concentration of 5% (cf. 10% for pathological samples). Following this, the helminth s fixed are cleared in lacto-phenol solution for microscopic observation, and photos/drawings/measurements are recorded with a camera lucida, as with other invertebrate morphological procedures. Some platihelmiths (e.g., trematodes and cestodes) are stained with acid carmin and/or hematoxylin solution.

Studies of the host-parasite relationship between whales and helminthes incorporates three scientific fields: VM, CE, and HM

1) VM: Studies are applied to clinical and/or epidemiological view points such as monitoring the health of captive dolphins associated with phoresy or external parasites in an aquarium. Most external parasites are, in fact, commensal organisms. All members of the family Coronulidae including the genera *Chelonibia, Platylepas, Coronula, Cryptolepas*, and *Xenobalanus* have been documented in the skin of mammals (whale, manatee and dugong), sea turtles, and gar fish (Lepidosteus), and are regarded as obligate commensal organisms (Morris et al., 1980). However, it is thought that settlement of *X. globicipitis* and another cirripedian group (the genera *Lepas* and *Conchoderma*: Lepadidae) may have increased due to reduced locomotor activity in dolphins and/or impaired regenerative and immune functioning of the skin prior to death (Aznar et al., 1994).

   The amphipodian species appear to be omnivorous and eat both fibrous algae and the cutaneous scales of their hosts. They are also burrowers that penetrate the horny layer of the hosts' epidermis and/or cracks between sessile barnacles (Arvy, 1982). However, it is unknown whether they are a carrier or vector of micro pathogens to whale hosts. Hence, monitoring of such cirripedian and amphipodian species should be performed, particularly when determining the health of captive animals.

2) CE: In some instances, understanding a parasite life cycle may clarify the ecology/evolution/zoogeography of the host-parasite relationship. That is, parasites can be used as biological tags or indicators of the ecological traits of the host (Aznar et al., 1994). However, there is often liitle detailed information describing their life cycle. For example, the whale genera, *Physeter* and *Phocoenoides*, (both living animals and fresh carcasses) appear to be a food resource for sharks in Japanese waters. This is based on recovery of cycticercoids of the genus *Phyllobothrium* (Phyllobothriidae: Cestoda) from fat tissue of the whales (Kuramochi, 2003) and their adult cestodes from a shark intestine, respectively.

3) HM: In Japan, many cases of acute anisakisiasis (typical zoonoses in Japan) are reported every year. Hence, whale helminths may also be regarded as human pathogens. For example, 3rd stage larvae of the genera Anisakis and/or Pseudoterranova (Anisakidae: Nematoda) parasitize the internal body cavity/muscle of marine fish (2nd intermediate host), and the adult nematodes persist inside the stomach of whales. However, humans are not suitable hosts (accidental host) for such parasites. Thus, a person eating raw fish (e.g. *Sashimi or Sushi*) containing live larvae will experience pain as the larvae enter the stomach mucosal membrane, but the larvae will also die in the accidental host (human) within several days.

   Parasite studies performed by the WAMC focus on conservation medicine. The motto of this field is, "One World, One Health", meaning that most zoonotic agents can easily enter human, captive animals, and wildlife and there is no academic barrier among the three scientific fields (VM, CE, and HM). The parasites mentioned above represent an unique conservation medical model. Given this, I believe the phrase "One Parasitology" could be added to the motto.

Helminths recorded from whales in waters around Japan

The helminths of several genera, including trematodes, cestodes, nematodes, and acanthocephalans, found in whales in waters around Japan and adjoining seas are documented below. These records are restricted to the helminth family and the genus names and its host abbreviations refer principally to Kuramochi (2003), who based the paper on records published during the mid 1990s. If more recent data are needed, researchers should consult the "Helminthological Abstract", a database published by CABI, UK. The host names' abbreviations are shown next, namely; BB: *Balaenoptera borealis*, BA: *B. acutorostrata*, PM: *Physeter macrocephalus*, KB: *Kogia breviceps*, BBa: *Berardius bairdii*, ZC: *Ziphius cavirostris*, Msp *Masoplodon* sp., OO: *Orcinus orca*, GM: *Globicephala macrorhynchus*, PC: *Pseudorca crassidens*, PE: *Peponocephala electra*, SB: *Steno bredanensis*, LO: *Lagenorhynchus obliquidens*, GG: *Grampus griseus*, TT: *Tursiops truncatus*, SC: *Stenella coeruleoalba*, DD: *Delphinus dussumierii*, LB: *Lissodelphis borealis*, PD: *Phocoenoides dalli* (dalli-type), PT: *P. dalli* (truei-type), PP: *Phocoena phocoena*, NP: *Neophocaena phocaenoides*, respectivelly.

TREMATODA

Campulidae: *Campula* (NP, DD), *Lecithodesmus* (BA, GM, PC, SB, DD), *Odhneriella* (GM, PC, SB, LO), *Hadwenius* (LO, PD), *Orthosplanchus* (NP)

Zasitrematidae: *Nasitrema* (NP, PC, TT, PD, GM, PE, LO)

Ophisthorchiidae: *Delphinicola* (DD, NP)

CESTODA

Terabothriidae: *Tetrabothrius* (TT, PD, BA, LO), *Trigonocotyle* (GM, GG, OO)

Diphyllobothriidae: *Diphyllobothrium* (DD, GM, BA, OO, GG), *Diplogonoporus* (BB, BA)

Phyllobothriidae (only cycticercoid): *Phyllobothrium* (PM, PD)

NEMATODA

Anisakidae: *Anisakis* (BA, GM, SB, LO, SC, LB, PD, PP, PM, KB, TT, PT), *Pseudoterranova* (PD)

Pseudaliidae: *Halocercus* (PD, PT, NP), *Stenurus* (PE, PD), *Pseudostenurus* (NP), *Pharurus* (PD, PT)

Crassicaudidae: *Crassicauda* (BBa, GG, Msp, TT)

ACANTHOCEPHALA

Plymorphidae: *Bolbosoma* (PC, BA, PT)

It is my impression that, if this fauna is compared with that of cattle or other domestic animals, an observer will be surprised by the low number of taxa (families, genera, and species) comprising the cetacean helminth faunas. Is this real, or not? If it is real, did the ancestors of contemporary marine mammals have greater numbers of parasites? If so, why? Although we do not have an answer for these questions, future researchers will likely address them.

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和 文 要 旨

一般における自然環境保護意識が高まり，海棲哺乳類，特にクジラ類の関心は目覚ましい。エコ・ツーリズム（ホエールウォッチング）における対象，食料としての利用，アニマル・セラピーとしての活用などの資源として側面ばかりか，国内外の動物福祉・愛護などの立場からの発言も活況を呈している。このようにクジラ類にまつわる話題は喧しい。酪農学園大学野生動物医学センターWAMC では 2006 年以来，国立科学博物館・北海道大学などとの共同で北海道沿岸にストランディング（漂着・迷入・混獲）したクジラ類の回収を行い，内外寄生虫や感染症の疫学調査に関するサンプリングを実施してきた。今後の研究展開上，保全医学的な側面から留意すべき点についてコメントした。なお，本稿は国立科学博物館が 2010 年 8 月に実施したアジア地域の研究者を対象にした研修〝What and how we learn from stranded marine mammals〟の寄生虫学研究紹介のために執筆したハンドアウト（未公表）を基に作成し，今回の出版については当該研修会主催者で博物館・山田格先生のご許可頂いた。