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Chitinolytic bacteria in the minke whale forestomach

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The North-eastern Atlantic minke whale (*Balaenoptera acutorostrata*) is a small baleen whale, feeding on fish and crustaceans. Molecular evidence for the inclusion of whales within the order of Artiodactyla have recently been found (Graur and Higgins, 1994, Mol Biol Evol, 11, 357-364). Like ruminants, whales have multi-chambered stomach systems with an initial non-glandular forestomach. It is lined with a keratinized stratified squamous epithelium which resembles that of the rumen, but lacks papillary projections, and the mean pH of the forestomach is 6.43 (SD 0.28, n = 12) (Olsen et al, 1994, J Zool, 234, 55-74). High concentrations of indigenous bacteria reside in the forestomach, initiating the digestion of the prey (Olsen et al, 1994, Appl Environ Microbiol, 60, 4445-4455). Even though the concentration of volatile fatty acids (VFAs) is high, ranging from 49-486 mM (n = 8) (Mathiesen et al, 1995, in: Whales, seals, fish and man, Blix et al, eds, 351-359), the total forestomach production of VFAs produced by bacterial fermentation is found to represent only 16.9 % of the average energy expenditure of minke whales (n = 5) (Olsen and Mathiesen, 1996, Brit J Nutr, 75, 21-31). This does not necessarily indicate a reduced importance of the forestomach bacterial system. In fact, mink whales are superior to the monogastric harp seals (*Phoca groenlandica*) in utilising crustaceans (Mårtensson et al, 1994, Br J Nutr, 72, 713-716; Mårtensson et al, 1994, Mar Mam Sci, 10, 325-331). A major structural component of the crustacean exoskeleton is chitin, the (1,4)-β-linked unbranched homopolymer of *N*-acetyl-D-glucosamine (NAG), which is the cellulose analogy in the marine ecosystem. Chitin is hydrolysed to free NAG by the chitinolytic system, which consists of two hydrolases acting consecutively. Chitinase hydrolyses the polymers of NAG, while chitobiase hydrolyses chitobiose and triose. The high digestibility of crustaceans in minke whales may be related to the occurrence of chitinolytic bacteria in the forestomach. The mean bacterial population growing in an anaerobic culture medium containing colloidal chitin ranged from (6-7) x 10^8 cells per ml forestomach fluid from krill-eating minke whales (n = 3). The total viable bacterial population were enumerated using an anaerobic rumen-like culture medium (M8W), and ranged from (0.6 - 120) x 10^8 bacterial cells per ml forestomach fluid (n = 5). *Eubacterium* spp. (25 %), *Streptococcus* spp. (18.2 %), *Clostridium* spp. (13.6 %), and *Bacteroides* spp. (11.4 %) were most common of the bacterial strains (n = 44) isolated from the forestomach fluid (10^3-10^5 dilution) of one krill-eating minke whale using the M8W medium. Utilisation of NAG were observed in 72.7 % of the bacterial strains isolated, while 53.3 % hydrolysed chitobiose and 11.6 % were chitinolytic. The prey is not chewed prior to swallowing, and the primary function of the bacterial chitinase may therefore be to disrupt the chitinous envelopes of the prey allowing access to the soft inner tissues by other bacteria.