D. BURNS/SCUWRS

A gentle way to age

The day started much like any other. Researcher Daniel Burns and his colleagues were keeping a respectful distance behind a pair of courting humpback whales. Then a third whale made an unexpected appearance. The new arrival began to press his attentions on the increasingly harassed female. Taking fright, she manoeuvred to place the research boat between herself and her suitors, leaving Burns and his team sandwiched between three 35-tonne whales, all shoving in different directions. "The adrenaline was pumping," says Burns, recalling how he braced himself on the bow of the six-metre vessel as it lurched precariously between the massive animals, each measuring about twice the length of the boat. But he and his team got what they came for: a sample of whale 'dandruff'.

Using a kitchen sieve strapped to the end of a stick, Burns and his team trail behind whales and quickly scoop up flakes of skin — some as big as the palm of a hand — before they sink. The flakes are naturally shed by the animals when they launch themselves out of the water (see above) or slap their tail fins on the surface.

Scooping up skin might not have the glamour normally associated with whale watching, but Burns and his colleagues think the flakes could offer a non-invasive way of working out a whale's age. If they are right, one of the key arguments in favour of killing whales for scientific purposes will be dead in the water.

Burns, a PhD student at the Southern Cross University Whale Research Centre in Lismore, New South Wales, goes out in virtually all weather, from dawn until dusk. He is part of a research team, led by Peter Harrison, that is developing genetic techniques to age humpback whales (*Megaptera novaeangliae*) from their sloughed skin. Currently, the most accurate way to age baleen whales such as humpbacks, which lack the teeth often used to age whales, is to count the lamination rings that form in their ear wax. Because the ear canal is closed to the outside world, wax accumulates in layers. Such laminations are conventionally thought to form twice a year in humpbacks. But counting them requires killing and dissecting the whale.

"It's better to have the approximate age of a live whale than the exact age of a dead whale."
— Peter Harrison

Showing the years

"Being able to age whales is something of a holy grail," says Phil Clapham, a whale biologist at the National Marine Mammal Laboratory in Seattle, Washington. Knowing whales' ages would enable biologists to model population dynamics, to better understand whale behaviour and assess how well the creatures are recovering from the devastating slaughter of the past century. It could also reveal whether mating tactics change with age, why individuals associate with each other at certain stages, and answer basic questions, such as exactly how long humpback whales live. Earlier reports suggested half a century, but researchers now suspect they could live for twice as long as that.

Harrison's team — and other groups around the world — hope that clues to a whale's age lie in the tips of its chromosomes, where there are stretches of short, repeated DNA sequences called telomeres. Like countdown timers, telomeres gradually shorten with age in some species, including humans. Studies of humans and birds suggest that individuals can be assigned to broad age classes[1,2], based on the length of their telomeres — although variability between individuals and in the rate of telomere loss during an individual's lifetime make it difficult to determine age precisely. Whale researchers are optimistic that, if whales are also shown to lose telomere sequences with age, telomere analysis could provide a non-lethal means to assign individuals to broad age groups.

The need is urgent, say researchers, because last year Japan declared that it will double its catch of minke whales for scientific research. It will also include, for the first time since the International Whaling Commission (IWC) imposed a moratorium on commercial whaling in 1986, humpback and fin whales in its annual catch for research — with a yearly take of 50 of each species[3]. One of Japan's main scientific arguments for increasing its catch is the need to collect samples to determine population structure. Harrison's team and others hope that the telomere method could scotch this claim. "If the technique works, it would put a large nail in the coffin of Japan's argument for a

scientific whaling programme," says Clapham.

Using telomeres to estimate the age of whales — and other cetaceans, such as dolphins and porpoises — is untested water. Telomeres shorten with age in some animals, but not in others[4]; some even lengthen[5]. To show that the telomere ageing method will work in whales, researchers need a sample of known age.

Harrison's team is fortunate to have access to an extensive photo library of whales created by Trish Franklin, a historian, and her husband Wally, a retired airline executive. The couple have been meticulously photographing and documenting the animals since 1989. "It started off as an interest, which then become a passion — and later an obsession," says Wally, who, along with his wife, is now undertaking a PhD under Harrison's supervision.

The photo library, comprised of pictures of nearly 3,000 individual whales, is invaluable as it provides the minimum age of many animals. The researchers are now systematically genotyping each animal — that is, obtaining an individual genetic 'fingerprint' — using the DNA extracted from sloughed skin samples.

Signs of success

While Burns braves unpredictable waters to collect the skin samples, his colleague Martin Elphinstone processes their telomeres in the quiet calm of the lab environment. Some early results are encouraging. According to Harrison, preliminary data from a handful of humpback whales hint that calves and adults can be distinguished on the basis of telomere length. "Ideally, in the long term, we'd like to refine it to a 5- to 10-year band width," says Harrison. Although unlikely to be as accurate as counting ear wax laminations, this range would be sufficient for population modelling, he says. "It's better to have the approximate age of a live whale than the exact age of a dead whale."

Support for these data comes from Per Palsbøll, a population geneticist at the University of California, Berkeley, who is also studying telomeres in humpback whales. His team has examined the DNA in skin biopsies taken from a dozen whales, ranging from calves to 20-year-old animals. "From this tiny data set, we see longer telomeres in younger animals," says Palsbøll. But he cautions that the method is far from reliable at this stage. "We have tremendous problems with reproducibility, even within samples from the same individuals," he says.

Telomere work is a complicated business. To assess how the rate of telomere loss changes over an individual's lifetime, Harrison's team is testing multiple samples from the same individual at different times in its life. To do this, the group will tap into a treasure-trove of skin biopsies collected over several decades by Scott Baker, a molecular ecologist at the University of Auckland in New Zealand.

Baker has samples from northern-hemisphere humpback whales, ranging from 2 to 30 years old, which were resampled 14 years after the initial biopsies were taken. Harrison is collaborating with Baker to measure telomere length in these animals to see how it varies within and between individuals. Baker also has a collection of biopsies taken ten years ago from southern right whales (*Eubalaena australis*) from the Auckland Islands; working with Nick Gales, of the Australian Antarctic Division of the Commonwealth Scientific and Industrial Research Organization, based in Canberra, he is resampling the same animals for telomere analysis.

M. EGAN/SCU

Daniel Burns (left) and his colleagues collect shed skin for DNA analysis by trailing after whales with a sieve.

Harrison's method for analysing whale telomeres from sloughed skin samples is unique in that it is non-invasive; getting DNA from skin biopsies involves taking a plug of tissue from near the dorsal fin using a biopsy dart from a cross-bow or modified rifle.

But using whale skin flakes is not without problems. The skin is often damaged and dying tissue. "Many samples don't provide enough high-quality DNA," says Elphinstone. It is also difficult to target specific whales using this technique, as skin flakes from different whales can get mixed up in the water. Harrison thinks this problem will be overcome once they have got a DNA fingerprint, or genotype, for every animal in their photo library to act as a reference.

If telomere analysis can be validated in humpbacks, it could be extended to other whale species. Gales is examining telomeres in animals ranging from those that are relatively short-lived, such as harbour porpoises that live less than 20 years, to bowhead whales that may live for more than 200 years. "We are looking at animals of quite different lifespans to see whether telomere length correlates with longevity," says Gales.

Gales is also analysing the method in a resident bottlenose dolphin population in Sarasota Bay, Florida — one of the world's best-studied dolphin populations. "The advantage is that a tremendous amount is known about their age and population structure, so telomeres can really be put to the test," says Gales, who is collaborating on the project with Randall Wells at the Mote Marine Laboratory in Sarasota.

The developments can't come soon enough for the Australian researchers. Japan plans to catch humpback whales in the Antarctic feeding grounds in the south-latitude summer of 2007–08, which includes the population that Harrison's team is studying. They dread the thought that one of their whales, many of which have been given nicknames, will turn up on a whaling vessel.

Even if whale researchers can develop a non-lethal method to age the creatures, whether it would immediately halt scientific whaling is questionable. Japan's Institute of Cetacean Research declined to comment when approached by *Nature*. Gales, who heads the Australian delegation for the scientific committee of the IWC, thinks it wouldn't. "But at least we can use it to apply more political pressure," he says. ■

P. HARRISON

Filter feeders: baleen whales don't have teeth, which are often used to age whales.

Carina Dennis is *Nature*'s Australasian correspondent.

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