Genetic tracking of a protected whale

Unusual circumstances have allowed us to trace the life of an individual whale, from its conception in the North Atlantic in 1964 to its sale as raw meat in Osaka, Japan, in 1993. We documented genetic polymorphisms in nuclear and mitochondrial genes of the Osaka meat and showed that they exactly matched those of a naturally occurring blue/fin whale hybrid harpooned near Iceland in 1989. This genetic match allowed us to trace the pathway of this individual from ocean to market. Such techniques can be useful in genetic monitoring programmes developed for the future regulation of the whaling industry and for international management of whale stocks.

Sample JE93-4 was purchased during a survey of Japanese whale markets in 1993. Mitochondrial control-region sequence analysis showed it to be a blue whale — or at least that its mother was a blue whale, because mitochondrial DNA is inherited maternally[1]. A check of genetic databases showed that its sequence is identical to that of whale #26 (GenBank accession no. MIBMCG), a blue/fin hybrid harpooned near Iceland in 1989 (ref. 2). This match suggested the possibility that we had discovered products from #26.

To confirm this, we returned to Japan to amplify a diagnostic nuclear locus from sample JE93-4. DNA sequences of actin introns clearly distinguish fin (*Balaenoptera physalus*) and blue (*B. musculus*) whales[3], and we found that JE93-4 possessed an allele characteristic of each species. Furthermore, analyses of actin alleles from #26 (from an archived sample) showed identical matches to the two alleles from JE93-4 (Fig. 1). Actin introns and the mitochondrial control region are polymorphic in both blue and fin whales[3]. This exact match of mitochondrial sequences[1] plus actin alleles of each parental type (Fig. 1) occurs with no other known whale, and indicates that product JE93-4 was derived from whale #26.

Alternative explanations consistent with these data, involving numerous blue/fin whale hybrids in the North Atlantic, seem much less likely. In principle, the inclusion of additional independent genetic markers (from other variable nuclear loci) could allow courtroom-level confidence intervals to be determined in the individual identification of whale products.

The identification of meat from whale #26 and the investigation of trade documents allow us to reconstruct its life. Whale #26, a male, had a blue-whale mother and a fin-whale father. Ear-plug growth layers indicate that #26 was born in 1965 (ref. 2). He was protected when the International Whaling Commission (IWC) adopted a global moratorium on commercial whaling in 1986 (ref. 4). In June 1989, #26 was 21.5 metres long but was sterile, with testes weighing only 2 kilograms (ref. 2). He was killed near Hvalfjördur, Iceland, on 29 June 1989 under a four-year scientific whaling permit issued by Iceland[5], but the whereabouts of meat from #26 has been unknown until now. Icelandic tariff records[6] show that the last documented export to Japan of frozen whale meat (1,074.6 tonnes) occurred in 1990, and no exports occurred in 1989. In 1993, our agents purchased 69 grams of #26 meat for ¥538 (about US$5) in a department store in Osaka[1].

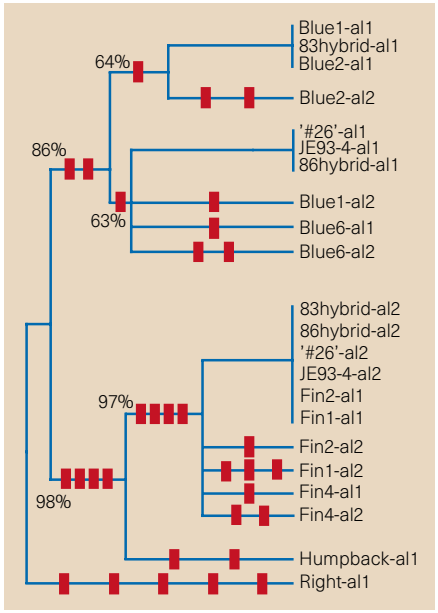

Figure 1 Maximum-parsimony reconstruction of relationships between 309-base-pair actin intron fragments from reference samples, known blue/fin whale hybrids, and whalemeat sample JE93-4, purchased in Japan in 1993. Alleles from each individual are identified by the suffix -al1 or -al2. Numbers adjacent to nodes indicate bootstrap percentages (500 replicates). All sequence differences between alleles are shown: tick marks along branches indicate mutation events. Actin intron fragments were amplified[3] using a portable MJ Research minicycler and primers ActIV (5′-CACATTGAGCCTATCTGATT-3′) and ActVIII (5′-CTGTGCTTGATTTCTCATAG-3′). Gel-purified actin fragments from some reference samples and from the Icelandic fin/blue hybrid whales collected in 1983, 1986 and 1989 were first extracted using silica membrane columns (Qiagen), then cloned directly into a phagemid vector (Invitrogen). Inserts were sequenced using ABI Prism reagents (Perkin–Elmer). Sequences were analysed using PAUP 3.1.1 (ref. 11) for parsimony-based phylogeny reconstruction and character change analysis. Sample sources and GenBank accession numbers are: Blue1, NMFSZ5794 (AF095354, AF095361); Blue2, PBRCBlue2 (AF095356, AF095357); Blue6, ULundBlue6 (AF095362, AF095363); Fin1, NMFSZ2821 (AF095369, AF095371); Fin2, PBRCFin2 (AF095368, AF095370); Fin4, ULundFin4 (AF095372, AF095373); Humpback, PBRCHback (AF095374); Right, ULundRight (AF095375); 83hybrid, ULundW1983 (AF095355, AF095364,); 86hybrid, ULundW1986 (AF095360, AF095365); '#26', ULundW1989 (AF095358, AF095366); JE93-4, Japan1993b-4 (AF095359, AF095367); NMFS, Southwest Fisheries Science Center, National Marine Fisheries Service, La Jolla; PBRC, Pacific Biomedical Research Center, University of Hawaii; ULund, University of Lund, Sweden.

In 1997, Norway announced that it would institute a genetic database to document individual minke whales killed in its commercial hunt in the North Atlantic[9], and

a genetic monitoring programme was part of the recent proposal to break the IWC stalemate by allowing coastal whaling by Japan and Norway[7]. Such programmes would allow the documentation of traces from hunt to market by comparing genetic profiles at the market to those of registered animals. Our results indicate that the use of several genetic markers now available for whales, including mitochondrial[1], microsatellite[10] and intron sequences[3], will allow individual whales to be tracked through international commercial channels.

The IWC and CITES policies are unprecedented efforts to control marine resources at a global level, but our results indicate that the control of whaling and trade in whale products would benefit from a comprehensive genetic monitoring programme. We have shown that loopholes in the current regulatory network are large enough for protected whales to slip through. More important, the use of similar genetic tools will allow new management efforts to focus on the individual, rather than the species or stock, allowing particular whales to be tracked from fishery to market, and to distinguish individual 'legal' whales from all the others.

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Motion of vortices in superconductors

A dissipation-free current can be achieved in a superconductor only when tiny magnetic vortices, which penetrate the superconductor when a magnetic field is applied, are pinned down against current-induced force. To investigate the mechanism by which such vortex pinning occurs, we have made real-time observations[1] of the onset of vortex motion in high-temperature

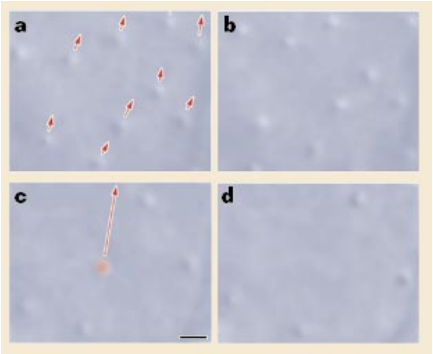

Figure 1 Lorentz micrographs showing the motion of vortices in a Bi-2212 film when the force is exerted on vortices by changing magnetic field. The motion is completely different above and below T_t. **a, b,** Micrographs, taken 1 second apart, showing slow migration at a temperature (T) of 20 K and a magnetic field (H) of 0.1 mT; $dH/dt = 0.0008$ mT s^{-1}. **c, d,** Micrographs before (**c**) and after (**d**) sudden hopping (T, 30 K; H, 0.05 mT). Scale bar, 2 μm; see Supplementary Information.

$Bi_2Sr_2CaCu_2O_{8+\delta}$ (Bi-2212) superconductors.

We created vortices in a Bi-2212 thin film by cleaving a single $Bi_2Sr_2CaCu_2O_{8+\delta}$ crystal[2], applying the magnetic field above the critical temperature (85 K) and cooling the sample below it. We moved the vortices by slightly changing the magnetic field and observed their motion through a 300-kilovolt field-emission electron microscope with a video system by improving Lorentz microscopy[1], such that the film thickness, the tilt angle of the film, and the intensity of incident electrons were as large as possible.

We investigated vortex motion for magnetic fields of between 0 and 4.5 mT and for temperatures of between 7 and 50 K, where individual vortices could be observed dynamically. The behaviour of the vortex was completely different above and below the transition temperature, T_t, which ranged from 17 to 25 K depending on the sample.

Below T_t, all vortices migrated slowly and maintained their relative positions. Analysis of two video frames (Fig. 1a, b) indicates that the speed of the vortices was 1.5 μm s^{-1} at 20 K, but they slowed down rapidly as the temperature decreased. This could explain the strong pinning in Bi-2212 at low temperatures.

Above T_t, vortices moved in different forms of plastic flow[3], depending on the strength of the magnetic field. At less than 0.1 mT, vortices trapped at preferential points suddenly hopped one by one, and the vacant sites were soon replaced by new vortices (Fig. 1c, d). The hopping was so fast that the vortex looked as if it was blinking on and off.

A characteristic of high-temperature superconductors is that vortices can be pinned at extremely small, densely distributed oxygen defects, and we believe that this might cause the migration. Because a single

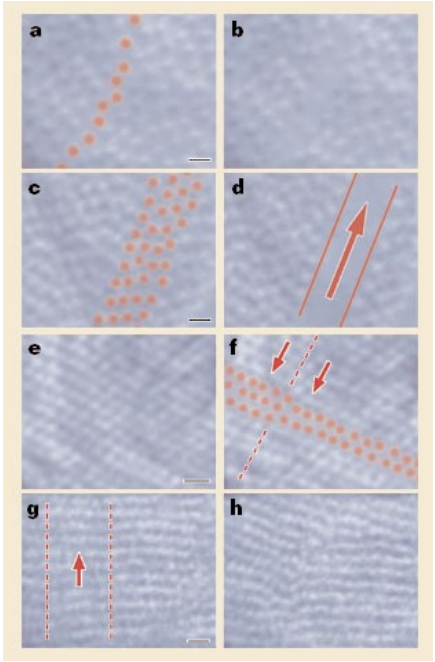

Figure 2 Lorentz micrographs showing plastic flow of vortices at the depinning threshold, $T > T_t$. The flows vary with H and T. **a, b,** 'Red' vortices forming filamentary flow at a temperature of 40 K and a magnetic field of 0.6 mT shown before (**a**) and during (**b**) the flows. **c, d,** River flow consisting of intermittently flowing 'red' vortices (T, 30 K; H, 1 mT) before (**c**) and during (**d**) the flow. **e, f,** Before (**e**) and after (**f**) production of a dislocation caused by a slip between two domains (T, 50 K; H, 2 mT). **g, h,** Before (**g**) and after (**h**) rearrangement of vortices during slips between domains (T, 50 K; H, 2 mT). Scale bar, 2 μm; see Supplementary Information.

vortex penetrating a film 200 nm thick, for example, may be collectively pinned by more than 100 oxygen defects, vortices would appear to move smoothly when thermally activated. Even above 0.1 mT, vortices continued to migrate at temperatures below T_t. Above T_t, however, larger and sparser defects, which have not yet been identified, became dominant and replaced the oxygen defects. In this case, as the interactions between vortices increased with the magnetic field, vortices moved in different types of plastic flow.

At magnetic fields of between 0.1 and 0.5 mT, many vortices were pushed by other hopping vortices, but moved only short distances before being interrupted by surrounding vortices. Generally, many vortices seemed to be moving randomly.

Between 0.5 and 0.7 mT, vortices became more closely packed and tended to move simultaneously along a filament (Fig. 2a, b). Such filamentary flow was predicted to occur at the depinning threshold in a two-dimensional film[4–6] where vortices favour a uniform distribution and the vortex lattice becomes easy to shear. Filamentary flow appeared only for narrow ranges of magnetic field and temperature because