

Lonesome George is probably the last giant tortoise of his type. But are scientists doing all they can to find him a partner, boost his sex drive and save his subspecies? Henry Nicholls finds out.

Poor Lonesome George. He may be famous, but he hasn't got a mate. All alone in the world and, sadly, singularly uninterested in sex, George the Galapagos giant tortoise looks set to be the last in his line.

Ever since George was discovered in 1971, there have been many attempts to get him to reproduce. Researchers have combed his island and the world's zoos in search of a female of the same subspecies. They have brought in a pair of female tortoises from a nearby island to act as playmates. They have even enlisted the help of a young, attractive female zoologist from Switzerland to help boost his sex drive.

So far they have met with little success: Lonesome George hasn't shown the slightest interest in the females in his pen. But his keepers do have other cards to play. George has yet to be introduced to what may prove his best possible partners — female tortoises from a distant island that seem to be more closely related to him than those currently in his pen. If researchers can get the two together and overcome George's low libido, he may yet reproduce.

The Galapagos Islands were once crawling

with giant tortoises. Before the first ship sailed into the Galapagos archipelago in 1535, it was home to at least 15 distinct populations of giant tortoise (Geochelone nigra), isolated from each other on different islands or on volcanoes separated by impassable lava flows. But heavy exploitation at the hands of eighteenth- and nineteenth-century whalers, along with competition from introduced animals such as goats, pigs and rats, caused four of these subspecies to go extinct and has earned the remaining 11 a place on the World Conservation Union's list of threatened species. Indeed, the G. nigra abingdoni subspecies from the island of Pinta was thought to be extinct until a snail biologist stumbled on a sole survivor: Lonesome George.

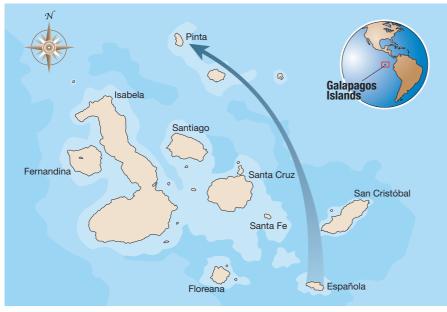
### **Lonely existence**

George lives at the Charles Darwin Research Station (CDRS) in Puerto Ayora, a bustling tourism-driven town on the Galapagos island of Santa Cruz. There he serves as a mascot for the archipelago's conservationbased research. The ultimate goal of his scientific custodians is to restore the Pinta ecosystem, which means repopulating the island with giant tortoises — ideally, with Lonesome George's descendants.

For many years, that was thought to be a lost cause. What could you do with a single male tortoise? "Conservation work in Galapagos reminds me of triage," says Linda Cayot, former head of protection at the CDRS. "The cases that look to be beyond saving are set aside to deal with those that are great emergencies but where the patient can be saved." For 20 years George was left alone, until, in 1992, Cayot put a few females from the nearby island of Isabela in his pen, hoping that romance would blossom. At the time it was reasonably assumed that tortoises from this island - the nearest geographical neighbour to Pinta that has tortoises on it - would be most closely related to him and so offer the greatest chance of a successful sexual liaison.

But the first examination of genetic similarities between the different tortoise populations in the Galapagos, done in the late 1990s, threw up something of a surprise<sup>1</sup>. DNA analyses conducted by Gisella Caccone, an evolutionary geneticist at Yale University in New Haven, Connecticut, and her colleagues showed that Lonesome George is more closely related to the subspecies on the island

# news feature



Distant relatives: genetic analyses suggest that George's ancestors travelled from Española to Pinta.

of Española (*G. nigra hoodensis*). This indicated that giant tortoises, which are ill-suited to life at sea, somehow survived an incredible 300-kilometre journey from Española in the south to Pinta in the north (see Map), probably by hitching a ride on strong currents.

More than a scientific curiosity, the result means that George has not been paired up with the most appropriate females. "Now that we see he has close genetic affinities to the Española and San Cristóbal subspecies, perhaps they would be a more appropriate source of a mate for this sole survivor," concluded the authors in their paper. But five years on, their recommendation has not been acted on.

## **Unnatural selection**

This is because the Darwin station abides by an ethical principle outlined by the Galapagos National Park Service — trying to ensure that biological processes continue as if humans had never set foot on the islands. Encouraging Lonesome George to mate with a tortoise from another island is an unnatural intervention that goes against this mandate, says Howard Snell, director of science at the CDRS.

This view is clearly at odds with Cayot's attempt at matchmaking between the Isabela females and George. But there has been no move to split them up, probably because there seems to be little need: in the dozen years that they have been together, Lonesome George has shown a spectacular indifference towards them.

Snell's attitude may sound odd, as it seems to favour the total extinction of a species over any 'unnatural' intervention that might preserve at least some of its genes. But Snell isn't completely opposed to cross-breeding. He is just biding his time, hoping to come across a better solution before having to resort to contaminating George's gene pool.

Snell's first priority is to find a living Pinta female for George. This, though unlikely, isn't impossible. In spite of their size, giant tortoises can be very hard to spot. Two tortoises live on the tiny island of Santa Fe, says Snell, which is visited by at least 50,000 tourists a year and serviced by three research teams. Yet these tortoises are seen only once every seven or eight years.

And there has been at least one tantalizing hint of tortoise life on Pinta. Back in 1981, nearly ten years after Lonesome George had left the island, Cayot and a colleague came across a lump of tortoise excrement near the top of Pinta's volcano. "As I stepped across a grassy area I looked down and saw a tortoise scat," she recalls. "I yelled out and we all stopped. We were all amazed." The scat hadn't completely decomposed, stirring hope that the faecal specimen had come from another individual still roaming the island.

But so far no one has seen it. Late last year, Peter Pritchard, founder of a privately funded conservation group in Florida called the Chelonian Research Institute, gathered together more than 20 park staff and conducted the most thorough search of the island to date. They had mixed success they found a total of 15 Pinta tortoises, although all of them were dead and only one of them was female.

### **Singles club**

There is a chance, however, that there may be Pinta females elsewhere, in private collections or zoos. Edward Louis, a conservation geneticist at Henry Doorly Zoo in Omaha, Nebraska, has collected DNA samples from more than 400 Galapagos tortoises from zoos worldwide. None looks particularly like a Pinta tortoise — although with only Lonesome George's genes to go on, it is impossible to be sure what actually constitutes a Pinta tortoise. Still, there are more than 100 zoo-based tortoises yet to sample, estimates Louis. "There are a lot of animals in South America out there that we haven't even looked at," he says. The Quito Zoo in Ecuador has "a tonne", he notes.

The CDRS itself is home to 23 adult giant tortoises of uncertain geographical origin, mostly garnered over the years from private collections. Some have been in captivity since the 1960s, and until 1976 (before the purist ideology came into effect) were allowed to mate freely with each other. Between them, they have produced a rabble of 36 offspring — most the result of undocumented pairings. Caccone and her colleagues have used DNA sequences to work out the geographical origins of the adult tortoises and the genetic origins of the progeny they produced<sup>2</sup>. Sadly for Lonesome George, none of them seems to be a Pinta tortoise.

## Long-distance romance

Most of the giant tortoises in captivity are not held in research stations or in zoos but are in private collections, says Snell. But these are much harder to track down. The permit requirements for owning a Galapagos tortoise are very, very confusing, and people are probably unsure whether their animals are legal, Snell explains. What's needed is a worldwide amnesty that would allow this population to be quantified and sampled, he adds. But such a move is logistically fraught.

As each day passes without news of a Pinta female, the CDRS comes ever closer to compromising its purist approach and turning its thoughts towards hybridization. If the work of geneticists is heeded, the Isabela females should be replaced by a closer relative from Española.

Although the Española population is itself under threat of extinction, things are far healthier than they are on Pinta. In the 1960s, the population fell to just 14 tortoises — 12 females and 2 males — that were brought into captivity at the CDRS. A third Española male was subsequently shipped in from the San Diego Zoo to be part of a captive breeding programme that aims, one day, to restore the Española subspecies to something like its former glory. Since 1975, CDRS scientists have returned more than 1,200 tortoises to Española. More than 15 years ago, nests once again began to appear on the island.

But things may not be as healthy as they seem. Michel Milinkovitch, head of the evolutionary genetics unit at the Free University of Brussels in Belgium, is part of a team that includes Snell and Caccone that has been looking at the genetics of this population<sup>3</sup>. Its analysis indicates that not all of the tortoises are contributing equally to the next generation. The male brought in from San Diego,

# news feature



Take your partner: most male tortoises are keen to acquaint themselves with the opposite sex (right), but Lonesome George (top left) is less accommodating.

known as Macho because of his sexual prowess, seems to have sired more than 60% of the offspring. Such a skewed genetic contribution "raises the spectre of severe inbreeding", warns Milinkovitch.

On the positive side, the sheer number of tortoises to have come out of the Española programme means that there should be plenty of sexually mature Española females to pair up with Lonesome George, says Milinkovitch. There are even four Española females already housed in George's neighbourhood at the CDRS, according to Caccone's survey of its tortoises of unknown origin.

### No sex, thanks...

Whoever his keepers decide to give George as a partner, there is still the problem that he seems to have no interest in sex. If this can't be solved, then all this talk of reproduction will remain purely academic.

Normally, a male giant tortoise will mate with almost anything. "It will mate with a dead tortoise; it will mate with a rounded rock on its trail if it's in the mood," says Pritchard. "But Lonesome George seems to be cut from a different cloth."

One possible explanation for George's indifference is that he hasn't been exposed to normal mating or courtship behaviour. Introducing a competitor male into his pen might goad him into activity, says Pritchard. "It might also be good if George was able to watch the active courtship and mating of Española tortoises in the adjacent enclosure," he says. If none of these measures succeeds in getting George going on his own, then artificial insemination might be the only option.

Sadly, nobody actually knows if Lonesome George can produce any sperm. In 1993, only a year after the Isabela females had joined him in his pen and while the disappointment at his inactivity was still freshly felt, German vet Gisela von Hegel came to the Galapagos, and showed the staff how zookeepers usually examine the sexual health of a male tortoise. If the animal's rear end is lifted off the ground, its muscles relax and its penis will drop out of the cloaca. This can then be manually stimulated.

Shortly afterwards a young and attractive female zoology student from Switzerland, Sveva Grigioni, arrived at the CDRS and volunteered her services. Initially things looked promising. "Sveva could get the other male tortoises in the exhibition pens to produce sperm within 15 minutes of approaching them," says Cayot. But with bashful George, things did not move so fast.

#### An intimate relationship

Grigioni spent several hours a day with him, gaining his confidence and a nickname — Lonesome George's girlfriend. "Day by day, he started to be more interested in the females with him," says Grigioni. But after nearly four months of daily visits to his enclosure, Grigioni's visa ran out and her work with George came to an abrupt end. "If I had had more time, perhaps I would have had more success," she says. "I am almost certain that I would have got sperm."

But she didn't. In the absence of a very patient researcher, there are other, harsher methods that might induce George to come up with the goods. Giving an animal a small electric shock does tend, in most instances, to generate a response, and is commonly used in many captive breeding programmes to obtain sperm<sup>4</sup>. But it is hardly a non-invasive procedure. Nobody knows how Lonesome George would react to having a probe inserted into his cloaca and an electrical current passed across his testes. So far, no one has been willing to find out. B. A. VIKANDER/CORBI

If Lonesome George is infertile, what other options are there for the survival of his subspecies? The answer, according to the tourist information panels that surround his enclosure, is the fantastic world of cloning. But if there is one thing that the scientists with an interest in Lonesome George all agree on, it's that there are other problems in the Galapagos on which money would be much better spent.

"Cloning George is ridiculous," says Caccone. "It was so difficult with Dolly the sheep and we knew so much about mammals. The task is daunting for reptiles." The effort required to clone each new species is huge, admits Dolly's cloner Ian Wilmut of the Roslin Institute near Edinburgh, UK, but cells from George and the other species of Galapagos tortoise should be stored anyway. "Who knows, they might be used for cloning at some distant time in the future," he says. As yet, cells have not been collected.

The one thing that Lonesome George has got going for him is time. He is probably less than 100 years old — young and sprightly for a giant tortoise — and might live for another 100 years. With patience and luck, George may yet be a father.

### Henry Nicholls is a freelance writer based in London.

- Caccone, A., Gibbs, J. P., Ketmaier, V., Suatoni, E. & Powell, J. R. Proc. Natl Acad. Sci. USA 96, 13223–13228 (1999).
- 2. Burns, C. E. et al. Anim. Conserv. 6, 329–337 (2003).
- Milinkovitch, M. C. et al. Proc. R. Soc. Lond. B 271, 341–345 (2004).
- Silva, A. R., Morato, R. G. & Silva, L. D. M. Anim. Reprod. Sci. 81, 159–175 (2004).

#### www.galapagospark.org/en/home.htm