

# The animals that sniff out TB, cancer and landmines

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In a small, hot room in a compound located in Tanzania's lush southern highlands are three white-clad technicians, a glass-and-metal chamber and a large brown rat named Charles.

After being gently dropped into the chamber, Charles aims his long snout towards the first of a series of ten sliding metal plates in the chamber's base. A technician swiftly opens it, revealing a small hole. Charles sniffs at it... and moves on. The hole is re-closed, and there's a clink of metal as the next plate is yanked back. This time, Charles is gripped. He sniffs hard, scratching at the metal, the five claws on each paw splayed with the pressure. The technician calls out "Two!"

Over by the window, her colleague is holding a chart, which he keeps raised so the others cannot see it. He inserts a tick. I glance over. The chart is a grid of small boxes, ten across by ten down, each marked with an alphanumeric code. Two of the boxes in each line are shaded grey. The tick has been placed in one that is white. It's highly possible that Charles has just saved someone's life.

Charles is an African giant pouched rat, a species endemic to sub-Saharan Africa. He's also a pioneer, one of 30 of his species that live and work here in Morogoro, a few hundred kilometres west of Tanzania's largest city, Dar es Salaam, on a programme to sniff out tuberculosis (TB).

TB is a disease that can destroy the lungs. About 9 million new cases are

diagnosed worldwide every year, one-quarter of them in Africa. Africa also has the highest TB death rate per head of population. Antibiotics can cure TB, but it's fatal if untreated, and many patients are never diagnosed. This is partly because the 125-year-old microscope-based test used across Tanzania (and in many other cash-strapped countries) picks up only about 60 per cent of cases, a figure that drops as low as 20 per cent for people also infected with HIV.

This is where Charles the rat comes in. Charles and his rat colleagues sniff cough-and-spit samples provided by suspected TB patients. The [rats](#) aren't infallible, but they do detect about 70 per cent of cases, and it doesn't matter to them if a patient has HIV – which matters a great deal in Tanzania, where about four in every ten people with TB are HIV positive.

This particular morning Charles has sniffed 100 samples, missing one that has been identified as positive by the public clinic – shaded grey on the chart – but identifying 12 new suspected cases, which will now go for secondary checking.

The next rat brought into the testing room, a sleeker, bigger-eared, three-and-a-half-year-old named Vladić (after a Bosnian Croat footballer; many of the rats are named after footballers), is even speedier than Charles. There's a rapid clatter of metal plates being pulled back and replaced. The two technicians manning the chamber call out numbers: "Three!... Nine!" Ticks rapidly accumulate on a fresh copy of the same chart. About 15 minutes later, Vladić has correctly identified eight out of 10 clinic-positives, and also 15 new suspects.

Fidelis John, the training supervisor, is looking on. Unlike the standard lab rat, *Rattus norvegicus*, the African giant pouched rat (*Cricetomys gambianus*) is not a species that has been bred over many generations to cooperate well with people. Is it very hard to train them to perform like

this? "It's not easy..." he says, smiling. "But it's possible. When a rat doesn't perform well, it is usually the trainer who is to blame."

Around the world, other animals – mostly dogs – are being used experimentally to screen human samples for disease; the TB-sniffing rats of Tanzania are the only animal disease-detectives in routine use. When medics first hear about the programme, they are often sceptical about the idea of using rats rather than machines, says Christophe Cox, CEO of Apopo, the Belgian-based organisation behind the project. But then they are shown the case detection data. The rats are saving lives every day, and, argue some advocates, the time has now come for dogs to do the same.

The first 'Lancet letter' came in 1989. Writing, as the name suggests, in the Lancet medical journal, a pair of dermatologists reported the case of a patient whose dog constantly sniffed at a mole on her leg, on one occasion even trying to bite it off. The woman sought medical advice. Tests showed it was a malignant melanoma, almost two millimetres thick. It was removed, and she remained well.

The second Lancet letter (as they're known in the dog-cancer-detection community) was published in 2001. John Church, a British doctor, and his colleague reported the case of 66-year-old man whose pet labrador, Parker, kept pushing his nose against the man's leg, sniffing at a rough patch of skin that had been diagnosed as eczema. The man went back to his doctor. The 'eczema' was found to be a basal cell carcinoma, which was swiftly removed.

"This is how it started," Church told the inaugural international conference on medical biodetection, held in Cambridge, UK, in September 2015. "It was all anecdotal."

At least, that was how interest in using dogs to sniff out cancer began.

But the idea of smelling breath, urine and stools to diagnose disease goes back millennia. In the time of Hippocrates, around 400 BCE, it was reportedly common for patients to cough and spit on hot coals to generate a smell that the physician would sniff to aid diagnosis.

Methods for disease diagnosis have clearly come a long way. But the Lancet letters got some, including John Church, thinking: might animal noses be quicker, or more accurate, and/or cheaper – and so able to be used more widely – than some high-tech cancer-screening techniques? If dogs really could sniff out cancer, what other diseases might they smell? And might the noses of other animals be useful too?

Over the past decade, there have been projects investigating the use of bees to sniff out cancer, for example, but that research hasn't advanced very far. The overwhelming focus in the field now is on dogs – and the African rats.

When someone with TB coughs, he or she exhales compounds produced by the bacterial pathogen *Mycobacterium tuberculosis*. If the TB is advanced enough, the smell of these compounds can even be detected by people. In 2002, when research to investigate the potential of using dogs in cancer diagnosis was in its embryonic stage, a former product designer from Belgium called Bart Weetjens began wondering about African giant pouched rats and TB.

Weetjens already knew that TB has a distinctive smell. "There is a lyric of a Van Morrison song: 'I can smell your TB sheets' – your bedsheets." Also, "in my native language, Dutch, the name for TB traditionally is *tering*, which etymologically refers to the smell of tar." Weetjens also knew that these rats are superlative sniffers. More than that, he understood how to breed them and how to train them, and his track record of using this species to save lives, albeit in a very different setting, was well-established.

As a boy, growing up in Antwerp, Belgium, Weetjens had kept pet rats. "Not only rats – I was very fond of all kinds of rodents. Hamsters and mice, and then rats. I tried gerbils and squirrels as well." He bred them in his bedroom. "I learned that they smell very well, but I was not occupied with that. I was simply breeding these animals to give offspring to the pet shops. It was a way to get pocket money. I gave up all rat breeding in my bedroom when I was 14."

After graduating and starting work as a product designer, Weetjens found himself increasingly preoccupied with the problem of landmines. "I saw a documentary about Cambodia, and also Princess Diana in Angola visiting mine-extraction operations. These two things triggered in me the magnitude of the problem." He began to consider landmine-detection systems: in theory, what kind of engineering solution would work best? Then he met a Dutch researcher who had come across stalled plans to try to use cockroaches to detect TNT exuding from buried landmines. "I thought, yes – this was the way forward: using local resources, a solution based on what was available in the context. This was for me an a-ha moment."

Except that Weetjens didn't think cockroaches. He thought rodents. In 1997, at a time when the local military academy was working on a landmine-detecting robot, he secured his first research grant, from the Belgian Development Cooperation, a government agency. "The secretary for the Development Cooperation had been a director of Doctors Without Borders. He knew the African realities much better than the army folks, actually. He immediately said to one of the professors in our team: 'This is a stupid idea, let's do it!'"

There were all kinds of questions to address, not least: which species to use? Ideally, he wanted an animal endemic to sub-Saharan Africa – which at the time was the region most affected by landmines – that wasn't that susceptible to disease, that relied heavily on scent (because

they'd have to sniff TNT in tiny concentrations in the air), that was long-lived and that could be trained. Professor Ron Verhagen, Head of Evolutionary Biology at the University of Antwerp, who had worked in Morogoro for many years, had a suggestion. "He said, well, I might have a suitable animal for you: the African giant pouched rat, which he had seen at some point in a village on a leash."

There were some early setbacks. At first, the rats didn't breed well in captivity, and it took a while to work out how best to train them. But the landmine programme, which operates from a base on the campus of Sokoine University of Agriculture, has become hugely successful.

At an average weight of about 1 kg, the rats are too light to set off mines. They can scurry across and search 200 square metres of ground in 20 minutes, compared with 50 square metres per day for a person using a metal detector. Apopo, the organisation Weetjens founded, dispatches trained rats to areas of land known or suspected to be mined (but not too heavily) and which cannot be farmed or lived upon because of the risk of setting off a mine.

Lightly mined regions can also be disproportionately dangerous, because local people are more likely to take the risk of venturing into them. James Pursey, who manages communications for Apopo, tells a story: "I was just in Angola. There's an area next to a school where a landmine once went off. I was talking to the headmaster and he said if the boys kicked a football into this area they would draw straws as to who would go and get it. None of the boys had been hurt. But when the rats searched the area, they found another landmine."

The Apopo rats are trained on a practice field a short drive from the headquarters. Early one morning, I catch the 'rat bus' – a truck that transports the rats from their kennel – along a bumpy road to a kiosk at the end of a red dirt track. Here, trainers are gathering to collect their

blue coats, with the Apopo logo, and water bottles, peanuts and bananas for the rats. When they have everything they need, they head down to the field.

Here, 1,500 deactivated landmines are buried up to 30 centimetres below grass and shrubs. The field is taped off into rectangles, varying in area from 5 x 3 metres up to 10 x 20 metres. The trainers work in pairs, each pair working a different rectangle. They start at one end. The trainee rat wears a harness with a large-bore spring attached to it. A cord runs from one trainer's boot, through this spring to one of the other trainer's boots. The rat runs freely along the cord, sniffing the ground as it goes. Tapes are also attached to each end of the spring, and the trainers hold the tapes. They use them to gently tug the rat back into position, or into action if it stops moving for too long.

Once the rat has sniffed the first half-metre's width of the rectangle, the trainers take a half-metre step to the side, and the rat sets off again. The trainers know where the landmines are. When a rat stops and sniffs and scratches in the right location, one squeezes a clicker (the kind routinely used in training dogs and dolphins) and the animal darts over for a nibble of banana or a nut.

In a real (rather than practice) field, paths for the trainers are first cleared by metal detectors. If a rat sounds the alarm, a trainer puts down a marker, and when the zone has been fully checked, someone with a metal detector goes to the spot to confirm the rat alert.

To graduate as a landmine-sniffer, a rat has to find 100 per cent of landmines in a test field in a single sweep. Abdullah Mchomvu heads the landmine-training team. He's out here this morning, supervising the session. "You have to be patient," he says. "Some learn quickly and others more slowly – but all in all they normally reach the goal."



Apopo rat teams have now worked in Angola, Mozambique, Cambodia, Thailand, Vietnam and Laos, not just on landmines but also on old ammunition, mortars and grenades. The Apopo team working in Mozambique, for example, has destroyed 13,294 landmines and returned over 11 million square metres of land to communities. This programme played a big role in the country's ability to declare, in September 2015, that it was landmine-free.

Mchomvu started working with the rats in 2002. He has 24 trainers under his supervision. It's satisfying work, he says. "To train the rats to detect landmines means we save the lives of people. To work at serving other people – I like it."

In another small, hot room at the Apopo TB centre, which is down the road from the landmine HQ, a skittish four-month-old rat digs his nose into one of three holes in a scaled-down version of the full TB testing chamber. He scrapes so hard at the hole it looks as though he's trying to disappear down it. Then he hears a click, and he quickly turns to gape his jaw at a small opening in the side of the chamber for his reward: a syringe-delivered slug of mashed banana, avocado and pellets.

The initial stage of training for rats, whether they're destined to smell landmines or TB, is socialisation, Fidelis John explains. Baby rats are first taken from their mothers when they're about five weeks old. They're handled every day for gradually extended periods, building up to their being carried around for the day on a trainer's person. The next stage is clicker training: they learn that the sound of the clicker means food. Rats in the landmine stream then learn to associate the scent of TNT with reward. The rats who will work on TB are given a TB-positive sample, explains John. "Once the rat sniffs the hole, I click. So the rat understands that if they smell this and the clicker comes, it means they get food. So then they understand: 'If I smell TB, I get food'."



It takes about nine months to train a rat. When an animal is thought to be ready, it's presented with 30 samples, eight of which are TB positive. To graduate, it has to detect seven out of eight positive samples with no false indications or eight out of eight with up to one false indication.

Training then continues on the job. The public clinics taking part in the programme send in half of all the cough-and-spit samples given by suspected patients, along with the results of their microscopy tests, which look for the presence of *Mycobacterium tuberculosis*. The testing rats sniff at least ten sets of ten samples every weekday. Two clinic-positive samples in each set of ten act as training reinforcers: when a rat correctly identifies one of these, it hears a click and receives a slug of mashed food. (The clinic's positive results generally are positive, Apopo says; it's the large proportion of missed cases that constitutes the main problem with the standard microscopy technique.)

At least two (if not more) rats sniff all the clinic-negative samples sent in. Any sample indicated as positive by any of the rats then goes for checking with a more sophisticated, more accurate – and more expensive – microscope technique than the one used in the clinics. In another lab in the complex are the five LED microscopes generally used for this final diagnosis. On the day of my visit, two technicians are at work. One shows me what the TB bacteria look like through his microscope: tiny, bright fluorescing stripes. It's only if the LED microscope check confirms the rat indication that a positive result is sent back to the clinic.

Better ways of detecting TB are badly needed in southern Africa, the epicentre of the TB epidemic, says Helen McShane, a professor of vaccinology at the University of Oxford and a specialist on TB in Africa. "Anything that is quicker, or more sensitive – or both – at picking up TB than current methods is to be welcomed. Particularly something like this, which is not resource-intensive."

Still, any new method for diagnosing TB needs to be both highly sensitive (to pick up all the cases) and highly specific (to avoid identifying too many samples as TB positive when they're not), argues McShane.

GeneXpert, a highly accurate DNA-based technique, which is supported by the World Health Organization, performs strongly on both variables. And in an ideal world, most clinics would use LED microscopy or GeneXpert. But these techniques are expensive and slow. A rat, which costs \$6,500 to train, can rattle through 100 samples in 20 minutes. A GeneXpert device, which costs \$17,000, takes around two hours to analyse a single sample. It costs about \$1 to screen a sample using a rat, compared with \$10 for GeneXpert. GeneXpert requires a stable electricity supply and controlled temperatures; the rats require food and water and play cages.

It's not possible to get both very high sensitivity and specificity with the rats, says Christophe Cox. Sensitivity can be improved (by using more rats to sniff each sample), but then specificity worsens, and vice versa. Still, he's convinced the rats have a crucial role to play in diagnosis. For developing countries, he argues, the rats are a fast and affordable life-saving triage tool.

The walls of Dr Claire Guest's office are covered with framed pictures of dogs. Behind her head is a portrait of a golden labrador, a dog that right now is curled up on my feet, called Daisy. Daisy has a special place in Guest's heart. In 2009, when she was working on a study to investigate whether a group of dogs, including Daisy, her pet, could reliably sniff breast cancer in human samples, Daisy started "acting weird."

"She kept staring at me. One day, I opened the boot of my car to get her out and she kept jumping at me. It was strange, because she's a very gentle dog. She bashed at me a few times, and I pushed her off. I felt

where she'd bashed me..." – Guest touches her chest – "...and I thought: there's a bit of a lump there." She went to her GP, who said it was probably a cyst. A specialist took a sample, and the results were fine. But the specialist also sent her for a mammogram, and he was concerned about the resulting image. "I ended up in hospital and had an ultrasound-guided core biopsy. I went for my results and was told not only had they found cancer, but it was so deep that by the time I'd felt it as a lump, it would have been very far advanced."

At that time, Guest was about a year into her role as Chief Executive of Medical Detection Dogs (MDD), a charity that she set up with support from Dr John Church, coauthor of the second Lancet letter. MDD, which operates out of a few buildings not far from Milton Keynes in the UK, has two aims. The first is to train 'medical assistance' dogs: dogs whose noses can save the lives of their owners. MDD has trained dogs to protect people with all kinds of problems – dogs that can sniff a dangerously low blood sugar level in someone with diabetes, for example, and sound the alarm; a dog that can detect the presence in the air of extremely low levels of peanut proteins, and warn its severely allergic owner; even a dog that can warn its owner, who has a disease called postural tachycardia syndrome, when she's about to fall unconscious, so she can get into a safe position.

Just before her cancer diagnosis, Guest, who originally worked as a psychologist and dog trainer, was pursuing this medical assistance training as well as work on cancer detection. The assistance dog training was going well. "I nearly left the cancer stuff alone. I thought medical assistance was where we could really make a difference. But then Daisy did what she did, and I thought: this is something we just have to get to the bottom of. This isn't something I could just walk away from."

Guest is now regarded as one of the world's leading dog-cancer-detection researchers. She helped to organise the 2015 Cambridge biodetection

conference, and she and her team, who train the dogs and run trials in collaboration with medical teams who provide the samples, have published a series of papers demonstrating that, yes, dogs can sniff cancer, and that more sophisticated training protocols dramatically improve their accuracy. Other teams have published work reaching the same conclusion. Dogs can smell bladder cancer, colorectal cancer, ovarian cancer and [prostate cancer](#). One Italian study, for example, that involved two dogs and 900 urine samples found that the dogs could accurately identify those from men with prostate cancer about 98 per cent of the time.

Guest's team is now working on two major studies, one on breast cancer, in conjunction with the Buckinghamshire Healthcare NHS Trust, and another, with Milton Keynes University Hospital, which will try to replicate the Italian prostate cancer study. The indications so far are that dogs could do much better than the prostate-specific antigen (PSA) test currently used to detect prostate cancer (only about one-third of the men flagged up by the PSA test actually have prostate cancer, resulting in many unnecessary biopsies, and it misses about one-fifth of men with prostate cancer). The study will also investigate whether dogs might be able to detect prostate cancer at an earlier stage than the PSA test.

No one knows exactly what the dogs are smelling, but there is work showing that metabolic changes brought about by cancer cause the pattern of so-called 'volatile organic chemicals' produced by affected cells to alter – and dogs seem to be able to detect patterns characteristic of specific cancers.

Ten years ago, Guest says, there was "massive scepticism" among medics about dog cancer detection. To some extent, she says, it continues, partly because no one has identified exactly which compounds the dogs are sniffing, but the steady drip of quality research taking the concept beyond anecdote does seem to be changing minds. (At the Cambridge

conference, Professor Mel Greaves of the Institute of Cancer Research in London stood up and said: "I came as something of a sceptic. But what I've seen about it looks incredibly interesting and promising...")

"This has been a difficult journey," Guest says. "The past ten years have been about careful persuasion of the medical profession and scientists." Now, though, she says she is contacted almost weekly by groups hoping her dogs might become involved in studies to investigate whether there are odours associated with the early stages of a wide range of diseases, including Parkinson's disease (a collaboration on this with the University of Manchester is now under way). "It's an exciting time," Guest says. And it's time, she thinks, to 'come out' and say something she's secretly believed from the start.

When she first embarked on the dog research, to engage sceptics, she said it was purely proof-of-principle work, aimed at investigating whether human cancers really do have distinctive odours. The ultimate aim, she told anyone who asked, was to use the results to develop electronic-nose cancer detectors – to take dogs out of the equation. She does still believe that one day, yes, life-saving e-noses should result from all this research. But in her office, one of her four dogs gently snoring, Daisy still curled up on my feet, she says: "What about the people who are dying now? If there's something at this moment, while we're developing an e-nose, that can save somebody now – why don't we use it?"

Dr Georgies Mgone, head of Apopo's TB programme in Tanzania, explains that, unlike with the cancer-sniffing dogs, it is known what the rats are detecting. He did the studies himself for his PhD.

His series of careful studies revealed that they respond to a combination of six volatile organic compounds produced by the *Mycobacterium tuberculosis* pathogen. And the rats can detect this combination even at

very low levels, which is why they don't have a problem identifying TB in someone with HIV.

Because of their weakened immune systems, people with HIV develop TB when infected with far fewer bacteria than are needed to make a healthy person sick. Relatively few bacteria in the cough-and-spit sample make it less likely that a technician using a microscope will spot them. But they still produce a smell that can be detected by the rats.

It may even be the case that the rats can sniff out the bacteria at levels so low as to be undetectable even by sophisticated laboratory techniques, Mgode suggests. "You will get a [clinic] sample that is indicated by 11 rats, but you can't confirm it's TB [with the LED microscopy]. To me..." – he taps his chest – "...to me, I know this is TB. But since we don't have a conventional method which is approved, we don't report this patient."

If the rats can detect TB at an earlier stage of infection than any other method, this could be a huge benefit, since a patient who is treated earlier is less likely to transmit his or her infection to other people. Mgode is planning to do more research to investigate this. But the biggest aims right now with the TB programme are more practical: getting results to patients faster, and expansion.

The programme started with four clinics in 2007. Now, 21 clinics in Dar es Salaam, about a third of the total, send samples via motorbikes and a bus to the rats. The rats also get samples from one clinic on the coast and three in Morogoro. A smaller sister programme, launched in Maputo, Mozambique, in 2013, using nine rats trained here in Morogoro, receives all suspected TB samples gathered there.

In 2015, the TB rats screened over 40,000 samples. In total, since the programme began, they have screened 342,341 samples and identified 9,127 patients who'd been told by the clinics that they didn't have TB.

Overall, the rats have hiked the TB case detection rate in the populations they're screening by around 40 per cent. Apopo is now in talks to start a new rat TB programme in Addis Ababa, Ethiopia. If they can gather impressive detection data from a third centre, this may help to convince remaining sceptics about the usefulness of the rats, hopes Christophe Cox.

The other main goal right now is to switch the Tanzanian TB testing from Morogoro to Dar es Salaam. That should allow the rat results to be returned to the clinics in time for them to be given to patients at the same time as the standard microscopy results, instead of many days later – a delay that currently means just under a third of patients diagnosed thanks to the rats don't actually receive their positive results. This should mean, Mgone says, that the programme will save even more lives.

In Dar es Salaam I meet Claudi, a boy whose TB was picked up by Apopo. He's waiting for me on the crumbling concrete porch of his home in Tandale, a slum suburb, in a yellow short-sleeved school shirt and grey shorts. It's in suburbs like this, where many people often share a house and nutrition is poor, that TB can spread relatively easily, explains Scholastica Myemba, who works for Apopo and is completing a Master's in public health at the University of Dar es Salaam.

Myemba supervises Apopo's team of outreach volunteers – people who track down patients identified thanks to the rats and make sure they take their medication. She translates as Claudi's grandmother explains what happened. Claudi is eight now. He was six when he got sick. "He was not in a good condition," his grandmother says. "He was coughing and coughing and not feeling good."

When she took him to the TB clinic at nearby Tandale Hospital, the standard microscope test for TB came back negative. Claudi continued to suffer. But then, just over a week after the negative result, the family



were contacted by an Apopo volunteer, who explained that Claudi's sample had been checked again, this time by rats. The rats had flagged Claudi's sample for further testing – and this test confirmed he had TB.

Claudi was prescribed antibiotics by the doctors to treat his TB. The volunteer came every day to make sure Claudi took his pills for the full six-month course of treatment. Now, he is healthy and able to work hard at school.

Still, Mgode explains, the benefits of a correct diagnosis go beyond access to life-saving drugs. For some patients, there is also the stigma of being suspected of having HIV.

Not long ago, with a group of Apopo donors, Mgode met a man in Morogoro whose TB had been detected by the rats. "Afterwards I asked him in Swahili: 'When you went to hospital and were diagnosed negative, how did you feel?' He said: 'Ah, my colleagues were asking me: "Man, if not TB, what else?"' That is the problem. He was feeling like even his friends were thinking he had HIV. So when he got the rats' result showing TB, he was so happy."

Weetjens and Mgode both talk about how difficult it is to get funding for the rat programme. Much of what Apopo does get consists of an assortment of relatively small donations from various governments and businesses along with proceeds from an initiative that allows individuals to 'adopt' a rat. The pace and scope of the dog research also suffers from a lack of funding, says Claire Guest.

For Guest, the success of the Apopo rat programme is "inspirational". When it comes to the dogs, the next three years will be critical, she says. If the prostate cancer and breast cancer studies go well, then she hopes dogs will join the rats as fully fledged disease detectors. She also hopes, like Mgode with his rats, that further work may demonstrate that [dogs](#)

can detect disease at an earlier stage than many current techniques.

As for the rats, no matter what happens in the future in terms of hopes for expansion – and funding – "already the technique is saving a lot of people," Mgone says. "Already, the impact is huge."

**More information:** Hywel Williams et al. SNIFFER DOGS IN THE MELANOMA CLINIC?, *The Lancet* (1989). [DOI: 10.1016/S0140-6736\(89\)92257-5](https://doi.org/10.1016/S0140-6736(89)92257-5)

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