How to use blood samples to assess the risk of sand fly-borne diseases

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In the CLIMOS project, we are developing an early warning system for diseases transmitted by phlebotomine sand flies, tiny blood-sucking insects similar to mosquitoes. As climate changes allow sand flies to spread to new areas, they can carry pathogens that cause diseases in humans and animals. Sand flies are poor flyers, living and feeding in a limited area. To create an effective early warning system, it is essential to identify where sandflies are found and feed. Extensive field collection followed by the identification of sand flies in different locations gives us information about the distribution of sand flies, an important piece of the puzzle in the complex picture of risk assessment. But how can we assess the risk even more accurately?

That's where measuring antibodies in blood samples comes into play. Are you familiar with what antibodies are? I'm pretty sure you know the concept of vaccination - when your doctor administers a vaccine containing a portion of a pathogen. You don't become ill, but your immune system learns to recognise that invader molecules and starts producing special proteins - antibodies - that act like a guided missile, targeting exclusively that particular invader and helping to clear it out as it enters your body.

Antibodies are produced by the immune system against almost anything foreign in our body. But how can we use antibodies to estimate the risk sand flies pose to us? Can sandflies get inside our bodies? Certainly not, but when it bites us, it inserts its tiny, straw-like mouthpart under our skin to suck out our blood. But our body tries to stop the bleeding by forming a clot at the bite site. To keep the blood flowing, the sand fly injects a special liquid called saliva into the bite wound. This saliva prevents the blood from clotting, so the sand fly

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can continue feeding. Once it's full, the sand fly flies away, but it leaves a little bit of its saliva behind under our skin. Our immune system recognizes this sand fly saliva as something foreign to our body. In response, it starts producing antibodies that specifically target the sand fly's saliva.

By measuring the levels of antibodies to sand fly saliva in a person's blood, we can estimate how much they've been bitten by sandflies recently. The more times we get bitten by sandflies, the more of these antibodies will circulate in our blood. This gives us an idea of the risk of getting sick from diseases that sand flies can spread. The same goes for animals like dogs that sand flies feed on. Vets may test a dog's blood for antibodies to figure out if it's been bitten a lot and is at risk of contracting sand fly-borne diseases.

One of the goals of the CLIMOS project is to create such test that can measure these antibodies to sand fly saliva in blood samples. But to develop this test, we first need sand fly saliva proteins as targets to detect the antibodies.

The usual way to do this is by dissecting the salivary glands out of sand flies. However, this approach has a few challenges: (1) There aren't many labs that have the expertise to successfully breed sand flies in captivity. (2) Even fewer lab technicians have the delicate skills needed to dissect the tiny salivary glands out of these miniature insects. (3) It would be extremely tedious and time-consuming to dissect many sand flies to get enough saliva for all the tests.

Instead of relying on dissected sand fly saliva, our CLIMOS team is going to engineer bacteria to produce the key salivary proteins that antibodies bind onto. First, we'll analyse the mixture of proteins in sand fly saliva and identify the specific protein that most antibodies bind to. Then, we'll insert the genetic instructions for this protein into common bacteria that are easy to grow in the lab to mass-produce it for us. This approach allows us to bypass the challenges of breeding sandflies and dissecting their salivary glands. It provides us with a steady supply of the key ingredients needed for the CLIMOS antibody test to detect

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sand fly exposure at the individual level and thus assess disease risk.

The antibodies in a dog's blood are specific to the saliva of the particular sand fly species that bit it. We can use this to assess the risk more accurately because different sand fly species can carry and spread different pathogens. If we detect antibodies to a sand fly species known to carry a particular pathogen, we know the dog is at risk.

As dogs and humans are often bitten by the same sand fly species, the antibodies in a dog's blood can also indicate the risk of disease for people living in the same area. The more dogs we test, the better picture we get of the sand fly-born disease risk in a particular area.

By monitoring sand fly exposure in dogs across many locations, the CLIMOS project can map the disease risk more accurately. This will provide another key piece of the puzzle for an early warning system to alert people to sand fly-borne diseases in local communities.

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