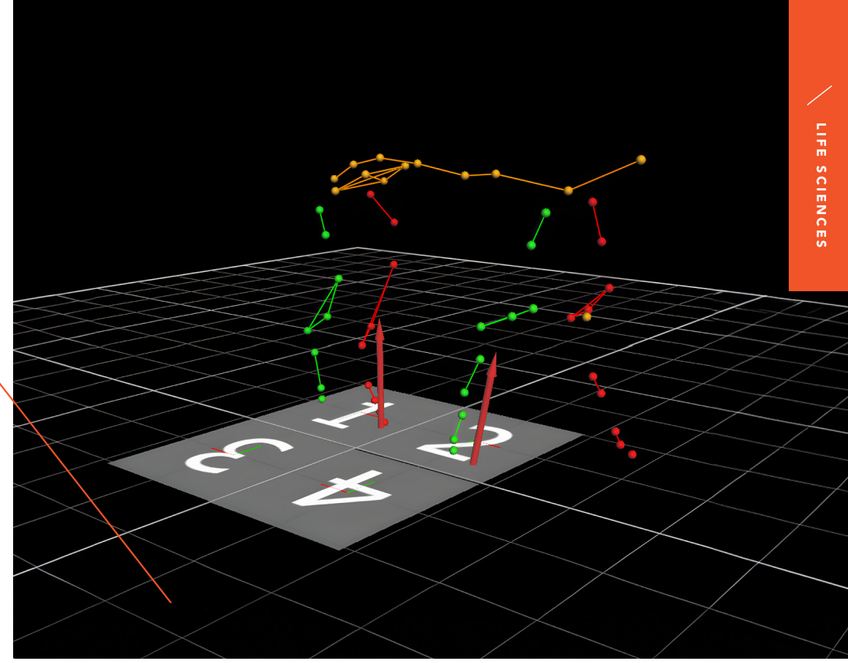
3AM STARTS, UNCOOPERATIVE QUADRUPEDS AND A 100°F BARN

LESSONS FROM ANALYZING SHEEP GAIT IN THE TEXAN SUMMER



Aaron Henry, PhD researcher at Texas A&M University's Department of Multidisciplinary Engineering

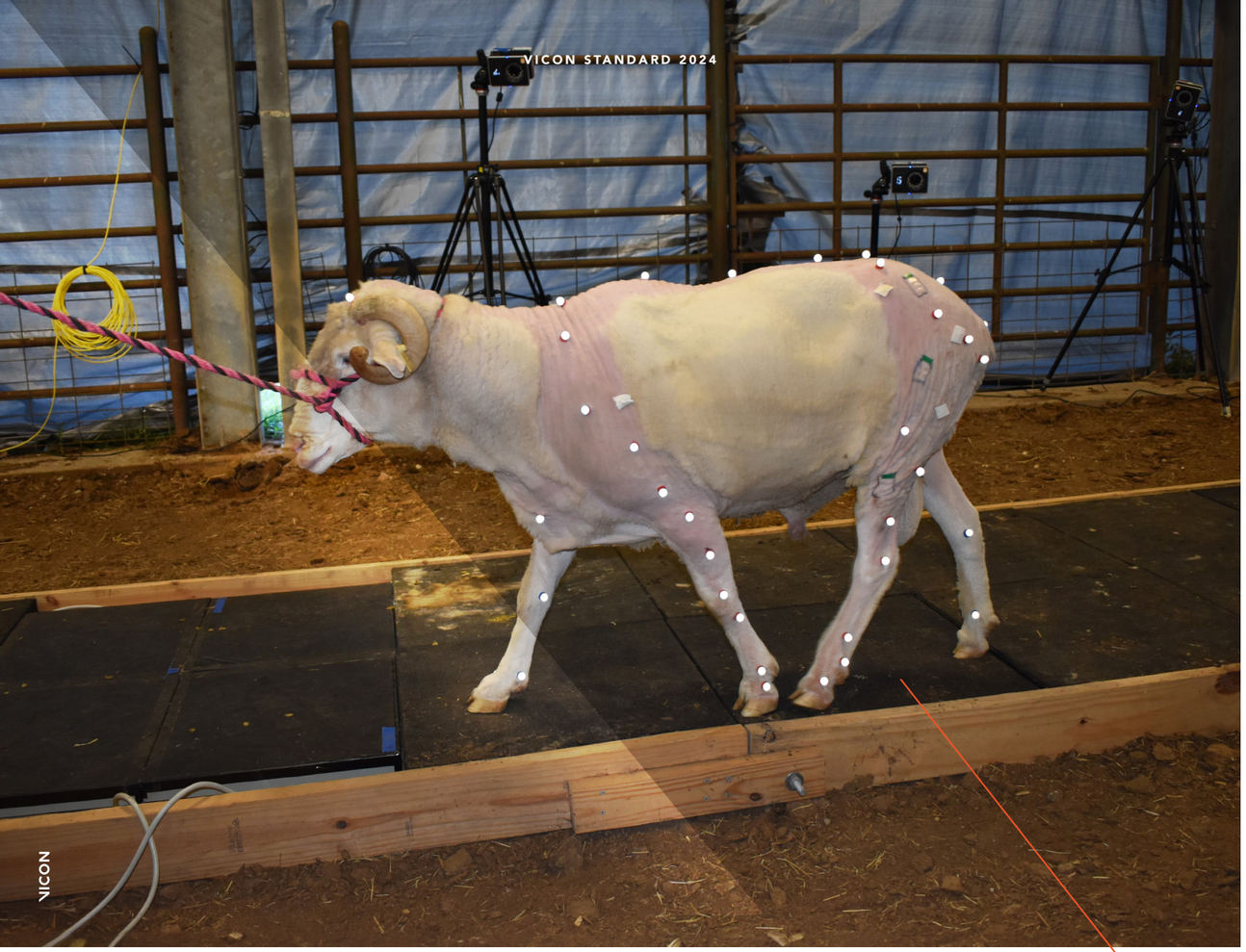
"One thing that we've learned but never actually implemented is to not do it in the middle of summer in Texas," jokes Aaron Henry, a PhD researcher at Texas A&M University's Department of Multidisciplinary Engineering. He's describing a 2021 motion capture project he co-led, under the supervision of Dr. Andrew Robbins and Dr. Michael Moreno, that saw a team of researchers and veterinarians analyze the gait of 20 often uncooperative sheep in a barn in 100-degree Texas heat.

While the project will sound like a worst-case scenario for many movement analysis practitioners, the research team hopes that it will advance the study of hypophosphatasia (HPP), an extremely rare metabolic bone disease that causes abnormal development of the skeleton and teeth. They also hope to develop a model similar to Plug-in Gait, which can be used for future quadruped studies.

"Dr. Gaddy and Dr. Suva, who are the principal investigators studying HPP, have a granddaughter who has hypophosphatasia, so they're invested in this project from a personal standpoint," explains Henry. "The disease is super rare and presents itself with extreme variability, so it's difficult to study or even to put a cohort together. Even when you do have a cohort, the subjects have varying presentations of the disease."

"In people with the same gene mutation, the symptoms can range from females who will lose their teeth post-menopause to, if you have the most severe form of it, something incredibly tragic like kids who can't breathe without a respirator and can barely walk," explains Jordan Ankersen, a Clinical Research Engineer and recent PhD graduate in Biomedical Engineering at Texas A&M, who co-led the project with Henry.

While the rarity and variability of hypophosphatasia make studying it in humans difficult, the condition can also be modeled in animals, offering alternative research routes.



Jordan Ankersen, Clinical Research Engineer and recent PhD graduate in Biomedical Engineering at Texas A&M

Dr. Gaddy and Dr. Suva previously looked into using small animals to study the disease but found that they didn't offer sufficient variety in the condition's presentation. Mice don't lose baby teeth, their bones form and remodel differently from humans and the mouse models did not show the altered gait of humans with HPP. As a result, large animal models, in particular sheep, who do have two sets of teeth, were the next step.

Despite the fact they're quadrupeds, sheep have musculoskeletal similarities to humans that make them ideal candidates for investigating gait biomechanics. "They allow you to do much more gross locomotion biomechanics, which is particularly helpful for a disease that affects the musculoskeletal system and impacts gait," says Henry.

Sheep with hypophosphatasia move, for example, "like Victoria's Secret models, they cross their legs when they walk," says Ankersen — something that corresponds with some of Dr. Gaddy and Dr. Suva's observations of human subjects with the condition.

"Another thing was that they wiggle like snakes when they walk — the bottoms of the sheep with the mutations sway back and forth," expands Ankersen.

A CHALLENGING ENVIRONMENT

In 2019, the research team did a pilot study that proved foundational to its efforts in 2021, leading them to a setup of 12 Vantage cameras, two Bonita video cameras, and one Vue camera, complemented by four force platforms. While the tools were high-tech, the setting was not.

"We learned a lot," says Ankersen. "We already knew we were going to 3D-print marker bases so that we could glue them to the sheep, because you don't have a great chance of sticking markers to them otherwise. And I had the idea to color-code the markers so we could help the veterinarians put them on, and we printed out sheets that matched the colors so they would be easier to count.

"We knew what cameras we needed and how we were going to place them. However, we were not prepared for the true magnitude of how much you have to change the capture environment.

"We didn't realize how much of a challenge the lighting was going to be because we were in an open-sided barn. It wasn't like we were in full sunlight, which would have meant we could set the cameras accordingly," Ankersen says. Instead, full sun would come into the barn at certain times of day, but there would be none at others.

"If you set the cameras one way in the morning, you'd have to recalibrate every 30 minutes, so we ended up having to put tarps up," Ankersen says. "But when you tarp the barn in, you essentially cut off the airflow and then you have 12 cameras, all of your switches and your computers running in this tarped-off barn in 100-degree weather, so keeping the cameras cool becomes a really non-trivial challenge! We ended up knowing that we had to get there at 3am, set everything up and start going at 5am so that we could finish by 11am."

Another problem came from moving the animals around in the tight space. "We knew that we had to have room for them to turn around. We thought

we had accounted for the side that the handler was going to walk on by putting a few extra cameras over there. But we learned we really had to have them switch sides because you still don't get great coverage when there's a handler walking. Especially because you can't predict which sheep are going to be lovely and which will be uncooperative."

As well as modifying their motion capture setup, the team learned tricks to change the sheep's behavior. "We learned to have a student sit under one of the tripods with a bag of food so that the sheep would walk to him," says Ankersen. "And we learned that since sheep are herd animals, they get a lot of comfort from companionship. So, we had to have another sheep in the capture area that we positioned too close to one of the cameras for my comfort."

KEEPING YOUR CAMERAS STABLE

Working on a large project with so many moving parts made user-friendliness a top priority for the team.

"Being able to monitor the camera temperature was super useful with heat being such an important factor," says Henry. "Then also, just general quality of life things, like being able to quickly reconstruct and make sure that we have all of our markers on a sheep is invaluable. When you've got over 50 markers on a sheep it's easy to miss one and not realize it, so being able to do a quick calibration and spot that he's missing one on his spine is really useful."

Ankersen concurs. "There would be times where a handler who's never worked with motion capture before would look at the screen and say, 'Oh, a dot moved on the screen, he lost a marker!'" she says.

Ankersen hopes that their work will prove useful beyond the sheep study, too. "We're trying to publish some more methods-based learning," she says, "because the data collection that we did in October 2021 was by no means perfect, but it went exponentially better than the collection we did in 2019 because we learned so much. We hope to give other researchers that same head start."

One of the outcomes of the project that could prove most important is the custom model Henry developed in collaboration with the veterinarians they had on site.

"Being able to apply it to other quadrupeds is the end goal, and really what would be ideal is having a Plug-in Gait quadruped model. You need to be careful with that because of the different morphologies that you would see in different quadrupeds, but a baseline that you could build off would be super helpful. I think it would help a lot of people that don't 'speak in squiggles', as it were, to get started in more animal studies, and do so correctly."