Ponds located on the St. Olaf natural lands provide a good habitat for native turtle species.

Turtle diaries

BIOLOGIST STEVE FREEDBERG SHARES HIS PASSION FOR TURTLES WITH ST. OLAF STUDENT RESEARCHERS WHO DIG INTO GENETICS, EVOLUTIONARY BIOLOGY, ECOLOGY AND BIOINFORMATICS.

By Patricia Grotts Kelly ’77

PHOTOGRAPHED BY TOM ROSTER

IN A DREARY FEBRUARY morning, Steve Freedberg unlocks a makeshift plywood door to reveal a bit of spring inside the Science Center. "It's warm in here," he says, "and a little stinky."

The room is lit up like a sunny day. All is quiet, except for an occasional *splash-plop… splash-plop*. Shelves lining the walls are packed tight with open plastic bins, which Freedberg calls "tanks." Each tank holds a dogpaddling turtle.

Thanks to Freedberg, who joined the biology faculty in 2006, St. Olaf is home to one of the most impressive and diverse collections of rare turtles at any American college or university — and St. Olaf students have the unique opportunity to work with them hands-on.

Freedberg's turtle collection comprises 65 specimens from South America, New Guinea, China, Africa, Australia, Belize and other exotic climes. It was willed to him by his teacher, Indiana University research associate Michael Ewert, who died two years ago. Freedberg earned his Ph.D. in ecology and evolutionary biology at Indiana in 2003 and came to St. Olaf after three years of postgraduate work at the University of Virginia. He arrived on campus on a sweltering, 100-degree day, the back of his truck packed with turtles keeping cool with chilled water bottles and ice packs.

They settled right in to their cramped closet digs, which Freedberg says work just fine because light, warmth, food and water are all that turtles really need. Turtles in nature spend much of their time crammed into rocky crevices or wedged between other turtles sunning on a log, not moving for hours.

"When I was a graduate student, I tried to set up more natural environments for them," he says, "but it didn't make a difference at all. They didn't seem to really care." This fall, the turtles will move to a more spacious abode in the new Regents Hall of Natural and Mathematical Sciences, complete with their very own sink.

"We know these guys are happy because they do all the stuff that psychologically well-adapted turtles do," he says. "They have great appetites, they never get sick, and they will breed."

Freedberg says it's important to set up 65 separate tanks. "Turtles housed together tend to either scuffle or mate," he explains. So far, he has not encouraged breeding, but if his students are interested in studying development, he says he'll create the right conditions for turtle love: extra nutrients and a gradual temperature change to mimic that of mating season. "I'll play Barry White," he says with a grin.

A TURTLE GUY

Steve Freedberg grew up in eastern Pennsylvania along the Delaware River. His fascination with turtles began at summer camp in the Poconos: "Some kids were down in the muck, and I saw one of them pull out a little snapping turtle and carry it back to his tent. I was instantly hooked."

Freedberg smuggled turtles home from camp, searched the swamps at home for more, and read everything he could find on turtles, which broadened his interests to include other species. "When I came across an interesting snake, I would hold on to it for a while," he says, "but I really was just a turtle guy."

When Freedberg attended the University of Florida at Gainesville, he majored in political science because it seemed "reasonable." At the same time, he set up several turtle tanks in his apartment, including a 55-gallon aquarium where he kept seven or eight turtles at a time. "Florida has some really cool turtles," he says. "Whatever critters I would catch, I would keep in there for a year and then let them go. I kept rotating them."

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It wasn't until his third year at Florida that Freedberg had his "epiphany:" "I took a biology course for nonmajors on ecology and evolutionary biology just for a science credit," he says. "I really got into it. The name of the biology building was Carr Hall, and I didn't think anything of it until one day I noticed a plaque that described how the building was named after turtle biologist Archie Carr. As a kid, I carried his books everywhere I went. I stood and stared at the plaque for about five minutes. Here was a guy who made a career out of something I was passionate about, and he did it well enough to have a building at a major university named after him. Right then I walked to the academic advising office and asked: 'What do I need to do to become a biologist?'"

▲ Favorites from Freedberg's 65-turtle collection include, left, an African forest-dwelling *Kinixys homeana* (hingeback tortoise) and, right, an *Emydura subglobosa* (red-bellied short-necked turtle) from Australia.

While Freedberg's fellow political science majors took electives like volleyball, Freedberg took organic chemistry, physics, calculus, and all the prerequisites for graduate school in biology. He chose to attend Indiana University because of its superb turtle collection, but he says he ended up benefiting most from it being one of the nation's top schools in evolutionary biology.

"At grad school I quickly realized that I wasn't going to get very far just focusing on one organism. I loved turtles, and they were my thing," Freedberg explains, "but if I wanted to get into any broad-reaching questions in biology, I had to branch out. If I got to work with turtles, it would be an added bonus."

TURTLE SMARTS

"Turtles are definitely inquisitive," says Freedberg. "I've dissected a turtle brain, and it's pretty tiny. But after years of interacting with them, It's clear that there's something more than instinct going on up there."

Freedberg shows off a few of his favorites: an *Emydura subglobosa* from Australia, with a pretty orange belly; a docile twistneck turtle that lays one giant egg instead of the usual 15 small ones; a hissing snakeneck turtle from Papua New Guinea; a giant musk turtle with whiskers, known to skim down muddy hillsides in Belize; and a shy Asian box turtle that Freedberg says is quite the gourmet.

Freedberg holds each turtle by the shell, careful to avoid their sharp, snapping beaks, and strokes under their chins. "You might not think of turtles as having personalities, but they do," he says. "When I was about 12, I had a wood turtle who was extremely charismatic," he says with a smile. "Wood turtles in particular have been known to perform about as well as rats in finding their way out of mazes. Turtles are actually brighter animals than most people give them credit for."

Freedberg's students love the turtles, which they use to study many areas of biology, including genetics, ecology, egg development, evolutionary biology and bioinformatics, which is the collection and analysis of biological information using computers.

At St. Olaf, Freedberg relishes his combination of turtle fieldwork, bioinformatics, and teaching. "The students here are great," he says. "They're so involved and dynamic. They are eager to contribute to real science and the turtles offer another opportunity to do so."

His research focuses on three areas: sex-ratio evolution, sex-determination evolution, and gene introgression (the incorporation of genes from one species into the gene pool of another through hybridization) in animals. For most turtle

species, sex is not determined by genes but by the temperature at which the egg is incubated — specifically, the temperature during the middle 20 days of a 70-day incubation period. Warmer temperatures produce females; cooler temperatures produce males. Variations can occur even within individual nests, with females hatching from the sun-warmed eggs at the top of a nest and males emerging from cooler areas at the bottom.

"One question that interests me is: How do they maintain a balance of males and females, given that they are at the mercy of the environment?" he says. "If you have a string of really hot or really cold years, you figure the population is going to be in trouble."

Freedberg says that most turtle populations today have more females than males. One explanation has been that they are already experiencing the effect of global warming and can't adapt quickly enough to overcome it. Sea turtle conservation groups have been particularly interested in Freedberg's research because of differing opinions about the efficacy of captive breeding programs, and about whether it is prudent to interfere with what may be natural controls for population genetics.

The earliest known turtles coexisted with the dinosaurs, 215 million years ago.

"With captive breeding programs, you can manipulate the sex ratio in any way you want," he explains. "Some argue that it's best to produce a lot more females than males because females are the reproductive unit, and the more eggs they can produce, the faster the population will recover. Some folks argue the flip side: that the natural situation should be produced, one male and one female, because if you get a small number of males, you could lose a lot of your genetic variation, which is really important. If only a couple of males are fertilizing many clutches, you get inbreeding."

Freedberg says his interest in sex ratios led logically to more theoretical and mathematical modeling work. And from there, he began focusing on genetics and bioinformatics.

"With temperature-dependent sex determination, you should get huge fluctuations over time. Sometimes you might get just a couple of males and a bunch of females, or vice versa. And when that happens, you basically bottleneck all of your genetic information into a couple of individuals and you predict that you'll lose a lot of rare genes; they just disappear," he says. "But that shouldn't happen with genetic sex determination because they should always produce an even number of males and females. At least that's what the theory predicts, but no one else has ever studied it."

Bioinformatics plays a big part in all of Freedberg's research, and he teaches a course in it as well. Simply put, he writes computer programs that serve as research tools. "It's called simulation modeling," he says. "Basically, it's a series of equations, computational models where you can simulate thousands of individuals in a population and study what might be going on over millions of generations. We input actual data from natural populations and hopefully it gives us a fairly accurate facsimile of what's going on in nature."

WILD TURTLE HEAVEN

Fifteen miles south of Wabasha, Minnesota, is a turtle biologist's dream known as Weaver Bottoms: a wetland with about 2,000 acres of elevated sand prairie, bordered by the Mississippi River on one side and marshland on the other, and home to eight species of freshwater turtles.

Last summer, St. Olaf seniors Elizabeth Leslie and Eric Scholten worked with Freedberg at Weaver Bottoms. Leslie, a chemistry major, had taken Freedberg's bioinformatics course and was looking to get some fieldwork experience. "I liked that the project had to do with genetics and molecular work," she says, "and we definitely got our hands dirty!"

The Weaver Bottoms populations are perfect for Freedberg's genetic research. The eight species of turtles eat the same things and nest in the same areas yet differ in whether they have genetic or temperature-dependent sex determination. Freedberg and his students are using the DNA data they collected from about 400 turtles to see if there is a difference in genetic diversity between the group with genetic sex determination and the group with temperature-dependent sex determination.

Freedberg came upon Weaver Bottoms by "serendipity" in 2007 when he attended the Arizona Powdermill conference, a sort of freshwater-turtle biologists' think tank. He was assigned to share a room with Michael Pappas, a Rochester, Minnesota, restaurateur and well-respected, largely self-taught turtle expert, who has been studying the turtles of Weaver Bottoms for 30 years. Pappas invited Freedberg and his students to work at the site and stay at his family cabin last summer and a great partnership was formed. Pappas shares his knowledge of natural behavior; Freedberg shares his knowledge of genetics and the assistance of his student researchers.

"Mike [Pappas] is fantastic," says Freedberg. "He has a huge, encyclopedic knowledge of the natural history of the turtles of the area. He knows where they are and how they behave, which helps us to collect them."

Pappas is equally impressed with Freedberg: "He's a really brilliant guy. You think he might be afraid to get his hands dirty. No, not at all." Pappas says Freedberg got an education in how to catch some of the more challenging Weaver Bottoms turtles: huge softshell turtles, measuring a foot and a half across, and 30- to 40-pound snapping turtles. "His students got right into it, and Steve did, too. He's fearless," adds Pappas.

On a typical day at Weaver Bottoms, the researchers would

▶ St. Olaf biology major Casey Baustian '08 holds a baby *Graptemys ouachitensis* (map turtle) in the research lab adjacent to the "turtle room" in the Science Center. The turtles will move this fall into a new home in the Regents Hall of Natural and Mathematical Sciences.

Studying evolutionary processes in turtles applies to many areas of biology, like the dynamics of diseases or the conservation of natural populations of organisms.

On a good day, the students would collect 50 to 60 turtles, measure them, take a tissue sample from their tails, mark them, and send them on their way.

set and check traps by boat; drive a stretch of two miles to look for turtles crossing the road; and hike through fields looking for nesting turtles. Once a nest was found, they would insert a data logger to keep track of temperature and other information, record the location, and cover the nest with a wire cage to protect it from predators. Their work with turtle nests benefited Pappas, who is studying the effect of agriculture on hatchlings. Baby turtles that emerge from their nests into a corn or soybean field become disoriented by the tall crops and can't follow their natural signals to water.

On a good day, Freedberg and his students would collect 50 to 60 turtles, take them back to Pappas's cabin to measure them, take a tissue sample from their tails (a tiny snippet of skin that the turtle doesn't feel), mark them, and send them on their way.

Sometimes the turtles had to spend the night on the porch

◂ As part of their summer research, St. Olaf seniors Eric Scholten and Elizabeth Leslie worked with Freedberg collecting DNA and tissue samples from turtles at Weaver Bottoms, near Wabasha, Minnesota.

of the cabin, and they took that opportunity to outsmart their captors. "We were shocked at their ability to get out of a container that was seemingly too tall for them," says Freedberg. "They would stack up on each other, and the top one would get out. Some of them actually escaped. We'd end up chasing the turtle as it was running back down to the water!"

Leslie processed the 400 DNA samples from the field turtles and has now taken the genetic-diversity project a step further by studying how genetic mutations occur in general.

"Turtles are a good model organism because they have such slow molecular evolution," she says. The Minnesota turtles from Weaver Bottoms and the exotic turtles from St. Olaf are both contributing data for her study.

"It's a really cool project," says Freedberg. "A couple of genetic markers are the same in really diverse turtle species, so we can study how certain types of genetic mutations occur in a whole bunch of turtle lineages. Again, the idea has broader applicability — not just studying for the sake of learning about turtles but studying to learn about how mutations occur and how different parts of the genome evolve."

Leslie has been accepted into a graduate program in genetics at the University of Iowa. Her particular interest isn't turtles; it's biomedical genetics.

"I want to study human disease, particularly identifying the genes that cause disease, because it is like putting together a puzzle," she says. "By finding out what happens when things go wrong, we can deduce how things actually work. And by understanding how disease happens we can open avenues for the development of therapies."

"The best thing about turtles is that they open students up to a lot of areas of biology," says Freedberg. "I like to think that studying evolutionary processes in turtles applies to lots of things, like the dynamics of diseases or the conservation of natural populations of organisms or even managing populations of things we eat and utilize. The more we understand how biological systems operate over long periods of time, the more beneficial it can be to people studying other systems. They're all interrelated."

Freedberg leans down to pick up a turtle that is staring fixedly at him, alert to his every move: "This guy's spunky," he says, as the turtle snaps at Freedberg's fingers. "He is really rare — a Chinese bighead turtle." Freedberg puts him back in his bin, and the turtle tries to bite again and inadvertently tumbles over on its back, legs waving in air. "We'll get you up, buddy," says Freedberg, helping the turtle to its feet. "He just wants to eat."

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Turtle Trivia

- Turtles are reptiles and belong to the order *Testudines.*
- The earliest known turtles coexisted with the dinosaurs, 215 million years ago.
- About 300 species of turtles are alive today. About 50 species live in North America.
- More than 40 species of turtles, including most sea turtles and many types of tortoises, are endangered.
- Like all reptiles, turtles are poikilothermic, which means their body temperature corresponds to the surrounding environment.
- Turtles breathe air, but they also can breathe subcutaneously under water for limited periods of time.
- The carapace (upper shell) of the tortoise or turtle is living tissue and comprises about 50 bones.
- Turtles have a rigid beak and no teeth.
- Turtles can live 150 years or more. Researchers recently discovered that a turtle's organs do not age (become less efficient) over time.
- Most turtles are omnivores and scavengers.
- Turtles vary in size from the bog turtle of about 4 inches long to the leatherback sea turtle, which can grow to 4- to 8-foot lengths.
- There are seven types of turtle: mud and musk turtles; pond and marsh turtles; sea turtles; side-necked turtles; snapping turtles; soft-shelled turtles, and tortoises.
- Turtles live on all continents except the poles.