

NCPARC RIMM Meeting
UNC-Greensboro
October 10, 2007

Chair

Mike Dorcas (Davidson College)

Attendees

Kim Coffey (Mecklenburg Parks and Recreation), Bob Cherry (NPS-BRP), Heather Newton (Appalachian State University), Jeff Hall (NCWRC), Kendrick Weeks (NCWRC), Gabrielle Graeter (NCWRC), Jeff Humphries (NCWRC), Lori Williams (NCWRC), Ann Somers (UNC-Greensboro), Dave Woodward (NC State University) John Groves (NC Zoo), Sathya Chinnadurai (NC State University, NC Zoo), Veronica Barnes (NC D.O.T.), Brian Ball (Ft. Bragg), Steve Price (Davidson College, Wake Forest University)

The meeting began at 1:12 pm.

Mike: Welcome to everyone here today. Let's start with introductions (made around the room). The agenda for today was sent out in an e-mail. We will try to get through those items quickly so we can discuss chytrid, what we should be doing in NC, what direction we should take, etc. We won't have a plan by today of how to resolve chytrid problems in the state, but we will hopefully have some direction and consensus on what we should do. The first item on the agenda has been addressed—Lori volunteered to take the minutes. I have a quick update on the Carolina Herp Atlas. As of yesterday, we have 6177 records of 129 species, 1166 photos submitted, and 283 registered users. For NC records only, we have 5044 with around 2000 submitted since March. Notable records include: 2 for mole salamanders in the piedmont, 3 for green salamanders, 9 for spotted turtles, 2 for diamondback rattlesnakes at Camp Lejeune (Jeff Hall mentioned that at least one of the diamondback rattlesnakes at Camp Lejeune was killed), 57 for timber rattlesnake from 18 counties, 3 for bog turtles, 1 NC and 6 SC records for Southern hognose snake, 10 for pigmy rattlesnake, 2 for pine snake, and 667 for Eastern box turtles. Related to the proposed box turtle monitoring project (an initiative of the Box Turtle Working Group), the Atlas is accomplishing the Tier 1 goal of record collection. Also, we've acquired several other databases such as SREL's, NPS's, etc. that will be incorporated into the Atlas. We're also working on the ability to click on a county and see a list of all herps from that county. Everyone here should be submitting records.

Jeff Hall: Is it still OK to send Steve an Excel spreadsheet with records?

Mike: Absolutely. Send us your spreadsheets.

John: What if we don't have coordinates?

Mike: We'd like for them to have coordinates, but we can find a general area on the map. My homepage at Davidson has a "UTM Finder" tool that we can use, too.

Mike: Back to the agenda, for now, we've decided to table the idea of a bibliography for lack of time right now. If anyone wants to tackle this, then the opportunity is there. We'd still like to have this.

Gabrielle: We need an intern for this project.

Jeff Hall: We have offers for an intern through the WRC, so maybe one of those people might be interested. I'll pursue this further.

Jeff Hall: Regarding CASP, in 2006 we had 61 random routes in the state with 3 nonrandom routes. In January or February of 2007, Kendrick and Lori requested from NAAMP additional random routes. We got an additional 74 routes and added another nonrandom route. In 2007 103 of these routes were assigned to observers, but only 59 of these people passed the quiz, 35 haven't even tried the quiz, and 11 have yet to pass. For the online data submissions, 36 routes were submitted, but we had 45 routes with physical datasheets, so there is a big disparity in what actually happened vs. what we expected. I anticipate receiving more datasheets, and these results were still better than we had in 2006 where only 23 routes were run. In the next few weeks I'll be asking for all the data to come in and for people to pass the quiz. There is a lot of work to do to fill all these routes. We have 63 still to be groundtruthed, mostly in the coastal plain. Data analysis won't start until this fall.

Gabrielle: What have other states done trying to get this going? What is the proportion of volunteers actually running routes and passing quizzes? What lessons have other states learned?

Mike: The problem in the past has been the quiz, perhaps that people are intimidated by it. Breaking the quiz into regions has made it easier, however.

Jeff Hall: And some of the species on the quiz have been combined that sound similar such as gray treefrog, leopard frogs, American and Southern toads, etc. We still want to do more training workshops across the state; we're working with the EO working group on this. Lori did two workshops in the mountains, Steve did a few, I did a few, Kendrick helped...they were well attended, but people are running into the quiz.

John: There are several people at the Zoo interested in participating.

Gabrielle: What about trying to match people who are running routes with people who are having problems with the quiz or who might be slightly interested, almost a mentor-type situation?

Jeff Hall: Yes, I've done that with a couple of people, and I think Kendrick did the same. I'll try to send out an e-mail about this to current volunteers to encourage mentoring.

Mike: Next on the agenda, we have an update on WRC herp projects.

Lori: I have an update on mountain region amphibian projects. This past field season we documented two more common mudpuppy sites in Ashe County and over four new sites for red-legged salamander and Cheoah Bald salamander in Graham County. We verified the continued presence of Junaluska salamander at one site in Graham County. Over the spring and summer we recorded observations of over 50 hellbenders from 11 counties, based on our surveys as well as viable reports from the general public. For hellbender surveys I had a lot of help from over 25 different volunteers and project collaborators including other WRC Wildlife Diversity staff from all regions of the state, staff from the NC Zoo, NC State Parks, NC Heritage Program, Appalachian State University, Western Carolina University, etc. Other results from the field season include groundtruthing of 3 CASP routes and establishing coverboard surveys at Richmond Hill Park in Asheville and Talula Bog in Graham County. Green salamander surveys for the fall have begun, and so far we've found two new sites, including one in central Henderson County that significantly extends the known range in that area.

John: Speaking of hellbenders, the Zoo now has over \$18,000 in the grant program for hellbender work, several thousand dollars in the "Snot Otter" button sales, and we are now selling hellbender t-shirts as well.

Mike: (To Ann) Can you update us on the box turtle group?

Ann: The Box Turtle Research Group has had two meetings so far. The group is made of people from state parks, several universities such as Duke, Elon, UNC-G, and Davidson, the WRC, the Museum, the Zoo, etc. Our goal is to start a citizen-science research effort in several of the state parks, community parks, and other areas. Kim Burge has already started a similar program. We hope to have a pilot program for the state in 2008. We have no budget, so we are looking for money. Gabrielle and I will be working to establish the marking system to be used. This effort has the potential to outlive us; some of the turtles marked at Patuxent decades ago are still alive, for example. We hope to submit a NSF grant pre-proposal and application in March with the full proposal in June. The idea has received a lot of interest; 13 people attended our last meeting. Staff from the Con/Ed Division of the WRC have also been included in the group, so we are growing. The booklet Catherine Matthews and I wrote, *The Box Turtle Connection*, outlines a format for standardized data collection. This takes us a long way to being able to compare data across the range. Sandy Barnett, who some of us met at the 2nd Annual Box Turtle Conservation meeting held at the NC Zoo, has a PowerPoint show and program already developed that we might could use as a training tool. Kim Burge with WRC Con/Ed has approached us to use it already. This whole project is a major effort. We will be tying up a lot of time and energy for the foreseeable future, but a lot of preliminary work has already been done. Plus, most of us know each other well and have already been working collaboratively on various projects. John Groves and I will be presenting at the 3rd Annual Box Turtle Conservation meeting at Patuxent in early November and at Haw River State Park at NC State Parks' annual meeting.

John: Is the Patuxent meeting posted on the NCPARC website? (Jeff Hall and Mike indicated they are unsure of this.) We will be giving a 30-minute presentation, and all are invited to show this is a collaborative effort. I'll announce the meeting to people at the Zoo.

Mike: I'll make sure it gets posted on the website.

Dave: Ann, what is the goal of this work?

Ann: The part of this project that is the most outreaching is Tier 1, the citizen-science part. This spring we'll bring in people interested in receiving training in this, "Friends of the Park" groups, etc. Tier 1 is the first level of reporting, what the Atlas is doing for us now. Tier 2 is more intensive, for example, Kim Burge's project, radio-tracking, etc. Tier 3 is the most intensive research like at Davidson College. Hopefully out of the Tier 2 and 3 work, we'll have assessments of relative abundance. We don't really know what is going on with populations at this point. The Zoo work would be Tier 3. Tier 2 would involve the greater public, press, "celebration days", essay contests, art contests, etc. Jerry Reynolds is involved in our group; he had the idea of involving neighborhoods. The big question is what will come from all of this. Expected outcomes are largely educational, but we still should have a larger body of knowledge than we do now. We don't know if anything will be publishable at this time.

Mike: We should be able to publish some things, but it depends on the research questions being asked.

Ann: Some questions are why are turtles where they are? Why are they not where they're not? At the 2nd turtle conference at the NC Zoo, a regional research group was born, but it wasn't led effectively, and nothing really came out of it region-wide.

John: The focus of these annual conservation meetings are expanding to all box turtles, not just Eastern box turtles.

Mike: For time's sake, let's move on and discuss the focus of the next meeting. Jeff Hall e-mailed me an idea. Something that is needed is a centralized database where you can go and find every record of every herp ever found in NC. Think about this for next time. There are lots of issues involved, logistically, financially, politically, etc.

Mike: The rest of our time today we'll spend on chytrid in NC. Little research has been done in the past, but there is research going on now. Dr. Sathya Chinnadurai, resident vet at the Zoo and NCSU is collaborating with Dr. Mike Levy, Dan Dombrowski, and Dave Cooper on some chytrid work. I also have notes that Betsie Rothermel at Austin Peay University e-mailed me regarding chytrid and what direction we might want to take; she is currently conducting chytrid research in the southeast.

Sathya: Chytrid fungus, a skin disease, has been best documented in Australia where there have been mass mortality events. In recent time, the US has had similar mass

outbreaks. What is the risk for amphibians in NC? There are a few anecdotal reports, but no large-scale surveys are being done. A recent article in the *Journal of Wildlife Diseases*, indicates a large gap in knowledge in the southeast, particularly with regards to salamanders. The project we are working on has two parts and is coordinated through the museum. The project includes field surveys, swabbing animals, testing protocols, etc. Historically, testing was based on histology with live or dead animals. Now we have molecular diagnostics. We can do live sampling, noninvasive, and use PCR techniques. We have 27-30 field sites that have been tested in NC. Dave and Dan have done some more. We are looking at all amphibians, trying to get an idea of what is out there and where. Out of the 175-200 samples we've collected, we detected 3 positive occurrences of chytrid. Testing includes one-time skin swabs of apparently healthy amphibians, which are also easier to find rather than sick or dead animals. There are a few species of salamanders that need to be looked at more closely because there is differential susceptibility among species. We know that bullfrogs carry it, but they don't really show the disease. What species have we seen with the fungus? Slimy salamanders and Blue Ridge dusky salamanders, for example. In the lab we administered a high dose of the infection to these species. No duskies showed the disease, but the slimy salamanders went downhill quickly and then died. What does this mean? We can't extrapolate those answers yet. We also tried lower doses with the slimy salamanders, but they did not show signs of the disease. Positive animals from the first part of the testing to the second part all showed positive whether or not their health declined, so PCR won't tell us that. What are some new directions we need to take based on funds, time, etc.? Do we need more regimented surveys? Standardize the amount of effort? Are we undersampling or oversampling populations? Do we target hot spots where it has shown up in multiple areas? Do we do more experimental infections? Do we continue to look at effects on individual species? These are all questions we face. Dan wants to start looking at museum collections for histology or molecular diagnostics. The history of chytrid goes back 30-40 years or more. Examining preserved specimens might help us develop a timeline of the fungus and estimate when it showed up in NC.

Jeff Hall: So you can detect it in preserved specimens?

Sathya: It is harder to detect, but you can do it. It's possible to do PCR, but it is less sensitive.

Mike: One complication with this...most museum specimens are often crammed into jars with many other animals or species.

Jeff Hall: What about physical inspections to see evidence of chytrid?

Sathya: I haven't seen that yet. It might be hard to see those features in preserved specimens. You could do a toe clipping or tissue clipping and preserve it for those tests.

Mike: Also, using the same swab all over the body...what if you don't swab in the area it occurs?

Jeff Hall: Wouldn't it be the same if you collect a part of the tissue that isn't infected?

Brian: So, you said that slimy salamanders were carriers but only died when given high doses?

Sathya: Correct. Stress in the lab or in the environment could increase the chance of outbreak, too.

Ann: What causes outbreaks?

Sathya: We don't really know. Other diseases? Stress? Environmental factors? There is still so much we don't know.

Mike: Joe Mendelson, with his work in Latin America, has seen waves of frog die-offs in the high, mountain environments. You can go one year and see certain species, and the next year half or more of those species are gone, or you find dead frogs everywhere. You can track the progression of the fungus through mountain habitats down through South America.

Ann: We don't know why?

Mike: It's easy to assume its something people have done. Climate change is an easy one to blame, but we really don't know.

Jeff Hall: Is there any way to get rid of it?

Mike: You can clean individual frogs, but then what do you do?

Jeff Humphries: Hellbenders in Missouri and Arkansas have shown lesions from chytrid, and there are documented population declines in those states. Jeff Biggler is currently testing hellbenders for chytrid and has found a 50% infection rate. The St. Louis Zoo has hellbenders from the Davidson River. One day all the NC hellbenders got chytrid and died. They were in the same water as the Missouri hellbenders. We swabbed several in the Davidson, and chytrid did not show up. Jeff Biggler is also swabbing other animals like turtles, hatchery fish, etc. A lot of fish have it.

Sathya: The fungus attaches to keratin, so many other organisms have it already and carry it and spread it around.

Mike: I wish you all could have seen Joe Mendelson's talk at Davidson recently about chytrid. This is a conservation issue like none before. It is not as easy as saying, "Don't build a Wal-Mart in that wetland." This is a global problem. We don't know what to do. We haven't seen, yet, a die-off wave go through NC like there is in South America, but we can assume chytrid is probably already here, in many areas, and it has probably been here a while.

Sathya: Also, we know chytrid is killed at 97 degrees or higher.

Jeff Humphries: Some of the hellbenders were kept in water in the high 90s. The fungus was killed off, and the hellbenders survived.

Jeff Hall: Did Mendelson document that chytrid was without question the main factor (in the die-offs in South America)? Some studies suspect chytrid, but they can't say for sure.

Mike: Mendelson is testing and setting up labs, but that was the point Betsie had, that other diseases such as ranovirus, etc. may actually be to blame.

John: The Panama golden frog went extinct because of chytrid.

Ann: What about immune disorders? Do they increase the susceptibility of chytrid?

Mike: That is the thinking behind human-caused reasons—have we done something to hurt immunity?

Kendrick: Has global travel or humans moving around so much been a vector?

Mike: Animals move, too. If it can attach to keratin, then we may be talking about reptiles, birds, and mammals as vectors.

Dave: Is this airborne? Soil? Water?

Mike: It can be in soil and water.

Sathya: There are chytrid fungi in the same class that are nonpathogenic, but this particular strain is bad. It needs to reproduce on keratin.

Jeff Hall: Has anyone done genetic testing on chytrid to see where it originated?

Sathya: Some of the oldest organisms came out of Africa from testing museum specimens.

Bob: Back to Jeff's question, if you find a dead amphibian, and it happens to test positive for chytrid, we can't say chytrid was the cause of death?

Mike: That is a good question.

Bob: So if we find dead amphibians, what do we do with them? Send them somewhere to be tested?

Mike: Betsie will be presenting her work on chytrid at the upcoming workshop in Arizona. She recommends if we find dead amphibians, it is probably worth getting them checked. It is relatively easy to run PCR.

Gabrielle: Do we send them to the vet school?

Mike: We can run samples for a few bucks at Davidson. We have the lab materials already. We may or may not have to pay for someone's time.

Jeff Hall: Is it realistic to expect ~\$20 per test?

Bob: How do we send them, preserve them, etc.? What is the process for sending them off to be tested?

Mike: Mendelson has prepared a video for reference on that topic. I'll put a link to that on the NCPARC website. Part of it is common sense—don't contaminate animals, use gloves, etc.

John: Where else do we know in the US has chytrid? At what altitudes?

Sathya: The western states have tested positive.

John: Are our NC mountains high enough?

Mike: A species from the Rockies was one of the first to start disappearing, in Rocky Mtn. National Park.

Bob: One more thing, do we need to worry about sanitizing our boots, nets, etc.?

Sathya: We do just so we don't spread it worse.

Bob: So you recommend it?

Sathya: Yes.

Jeff Hall: We don't have a WRC protocol yet.

Mike: At Davidson, we just took our first chytrid samples. We bleach our boots but inconsistently. It adds another level of complexity to our field work.

Jeff Hall: I'll be going to the chytrid conference and hope to get more guidelines. This will also be a topic at the SEPARC habitat guidelines meeting.

Gabrielle: There has been recent discussion that you can use Lysol instead of bleach; it doesn't break down your boots.

Sathya: We use diluted bleach.

Jeff Hall: At one time, there was the thought that biologists were spreading it?

Mike: But in reality, there are so many more general people out there, they are probably just as responsible.

(break at 2:37 pm)

Mike: There are questions we should discuss. What should we do as the RIMM working group about chytrid in NC, if anything? In relation to the issues we've discussed, and Joe's and Betsie's comments, should we look at species? Locations? Historical framework? What about in light of programs already going on, like CASP? Should we go to some of those sites and swab some frogs, monitor for chytrid at the same time as populations?

Jeff Hall: One thing Dan mentioned along those lines...the sampling technique is so noninvasive, we could collect specimens even if we don't have them tested right away. The specimens will keep even if we don't have money for testing just yet.

Mike: Right. It is relatively easy to do. So, we starting doing this and used plastic swabs. We store them dry and then put them in the freezer, one swab per vial, labeled. We have individually marked animals, so it would be interesting to sample the same animal repeatedly over time in the same habitat. It appears some groups are higher risk than others? Frogs vs. salamanders? We need to get the word out to all herp folks in the field, WRC biologists, etc. If we see dead amphibians, collect them and freeze them. Report any die-offs or dead amphibians, whether we can say for sure chytrid caused it or not.

Ann: Is there some place to deposit these? Do you want them? (to Sathya)

Sathya: Anyone who has a PCR machine can do the testing.

Mike: We're not looking to become the state lab for PCR, but we can do certain things in our lab, so it is a possibility.

Jeff Hall: That is exactly what everyone is asking.

Kim: That could be a lot of animals, particularly in one area if there is a large die-off.

Ann: Can we all just use our own freezers for now, until we find out more information or have a central repository?

(discussion among various group members....What does this mean if we find it? What do we do? We can document all we want...no way to stop it that we know of...bottom line, we don't know what to do...reintroduction is not practical, nor is treating individual animals and releasing them.)

Kendrick: What about cycles over time? Animals come back; maybe there is no reason to do anything.

Mike: Maybe there are things we can learn...what species are susceptible? What populations have been depleted? Can we document any anthropogenic causes?

Ann: There is a concern about frogs in Asian markets and other places. Are varieties being shipped here from Asia? Is that strain of fungus the same as we have, or is it different?

Sathya: Those could be different strains of the same species of fungus, but they are highly related and present the same pathogenic risk.

Gabrielle: Is the chytrid we have our “native” chytrid?

Sathya: Are other strains coming into play that our natives have not evolved to adapt? There are other pathogens out there that cause disease. This is not the first. Some suggest the strains we have today go back 50 years to different parts of the country, from museum specimens. We don't have many good biological samples to test further back than that.

Mike: Has chytrid ever been documented on reptiles?

Sathya: Snake sheds is a good way to grow chytrid.

Ann: Eels?

Mike: Fish?

Sathya: Yes, the keratin on scales could carry it.

Mike: Most ambystomatids have little keratin in their skin—has chytrid been documented in them?

Sathya: Only in their mouth parts.

Jeff Humphries: Would it cause hellbender legs to fall off? Tissue sloughing?

Sathya: Yes, sloughing, bloody blisters, etc. Other pathogens can cause limb deformation.

Mike: So this is something we should be aware of and talk about. I don't see that any one entity (for example, the WRC or *Wildlife Action Plan*) should be the lead on this or take responsibility for this.

Jeff Hall: It seems like if we are to focus on something, we should focus on something that is isolated, where whole populations could easily be wiped out (for example, green salamanders, pine barrens treefrogs, etc.)

John: There has been discussion among zoos about assurance populations, with a focus on southeastern amphibians, NC endemics, etc. But, again, is it really going to help?

Mike: With box turtles, I can see some utility with that. With many species, reintroductions are usually a bad idea in many ways. It depends on what the central threat is that put that species in jeopardy anyway, but chytrid is a fundamentally different thing. It is not because a species is being eaten to extinction like with turtles.

John: The idea is that if we can figure this out, say, with 500 animals at a minimum for assurance populations, how do we do it?

Mike: Joe Mendelson has some breeding animals for assurance colonies that are now extinct in the wild.

Ann: There should be continual testing of captive colonies, to find disease-resistant individuals.

John: We're going ahead with it, projected for a minimum of 50 years—a huge investment for a project like this.

Mike: A third of the world's amphibians are predicted to be extinct in the next several decades. There has not been a mass extinction event like that since the dinosaurs.

Jeff Hall: One more thing...is there a test for water?

Sathya: We are supposed to be able to test zoospores in the water. Techniques have not been validated yet, but it is close to being possible.

John: Will you also test the soil?

Sathya: Yes.

Jeff Hall: Playing the devil's advocate...we need to be very careful what we assume when we find a chytrid-positive animal. We assume we know chytrid is responsible for something when we don't know. There may be other factors at work besides chytrid.

Sathya: With concurrent diseases, no one has co-infected animals yet, for example, with chytrid and ranovirus. There is still so much we don't know. If we have a swab bank, we'll be able to test for many years in the future, particularly as future tests are developed.

Mike: With all the WRC surveys being done, why not go ahead and swab some animals? I propose to those of you doing surveys, go ahead and start keeping a bank of swabs, for use now or in the future.

Jeff Hall: We can keep the swab in the freezer indefinitely?

John: We're going to start a survey at our Zoo sites, so can we send swabs to you (to Mike)?

Mike: We are hoping to do some at Davidson. We can't devote the entire lab to this, but if it fits in with what we are already doing, we should be able to run some tests. Or, we could incorporate testing into existing courses with lab components.

Jeff Hall: There is a researcher at Wake Forest University, who is permitted to do some testing on a couple of species. It might be good to talk to him and see if we can collaborate or possibly use his lab.

John: The AZA has designated 2008 as the "Year of the Frog" and has planned all kinds of programs, particularly on February 29th, since it is a leap year.

Mike: Wrapping up today's meeting, (to Jeff Hall) what is the status of the NCPARC meeting destination and date? It is at the Trinity Center?

Jeff Hall: That is what I have so far—the Trinity Center at Emerald Isle, March 6-7.

Lori: What is the theme this year?

Jeff Hall: We are still working on that, but it will likely have to do with coastal herps, with an opportunity for field work. We are still working on the agenda.

Gabrielle: The SEPARC meeting will be February 21-24th in Athens, GA. There is some information on the website, but more will be coming. There will be symposia the first day or two, then working groups, and field trips on the last day. The theme is "Tools for Conservation".

Mike: So everyone knows, Gabrielle is the new co-chair for SEPARC along with Betty Crump.

Mike: Thanks to everyone for coming today.

The meeting adjourned at 3:23 pm.

Respectfully Submitted By,
Lori Williams