

Ketterson / Nolan Research Group Collection

This document is part of a collection that serves two purposes. First it is a public archive for data and documents resulting from evolutionary, ecological, and behavioral research conducted by the Ketterson-Nolan research group. The focus of the research is an abundant North American songbird, the dark-eyed junco, *Junco hyemalis*, and the primary sources of support have been the National Science Foundation and Indiana University. The research was conducted in collaboration with numerous colleagues and students, and the objective of this site is to preserve not only the published products of the research, but also to document the organization and people that led to the published findings. Second it is a repository for the works of Val Nolan Jr., who studied songbirds in addition to the junco: in particular the prairie warbler, *Dendroica discolor*. This site was originally compiled and organized by Eric Snajdr, Nicole Gerlach, and Ellen Ketterson.

Context Statement

This document was generated as part of a long-term biological research project on a songbird, the dark-eyed junco, conducted by the Ketterson/Nolan research group at Indiana University. For more information, please see IUScholarWorks (<https://scholarworks.iu.edu/dspace/handle/2022/7911>).

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So you found a nest.....

(compiled from earlier versions by Nicole Gerlach, 2012)

Congratulations!

Before you leave the area:

Parents ID: If there are adults in the area, now is an excellent time to ID them! Get as much information on their color bands as possible; even a partial ID will be helpful in the future.

Nest Contents: Now and later you will need to record the state of the nest when you visit (stage of building, number of eggs, whether eggs look fresh or well incubated, how many nestlings, whether eggs gone and nest disturbed by predator, whether one or both parents were nearby and their behavior, etc.) and anything else of interest (a death, a visit from a neighboring male, a copulation, the fact that the nest was filmed or recorded, anything at all).

If you find a nest with 3 or fewer eggs, mark the eggs! (see instructions below)

If you find a nest with already-hatched nestlings, *and those nestlings are less than 7 days old* (not feathered, eyes closed or only slightly open), weigh the nestlings and measure their tarsi (and individually mark them, see below). The hatching date of the nest can be estimated by comparing these measurements to the “How Old Is Your Nestling” chart. *If the nestlings are 8 days or older* (eyes open, alert and looking at you), do not disturb the nest! Make your best visual estimate as to the number of nestlings and their age (based on feather development), but if you touch them, you risk inducing early fledging.

Flagging: Mark the nest with red/black flagging tape. Make a small (~1in) bow directly over the nest, on a small stick or available vegetation. For nests along the road/trail, tie a longer piece of flagging (~12 in) to the vegetation directly *across* the road from the nest. For nests that are in the woods off the road/trail, you will need to make a flagging trail from the closest point on the road/trail to the nest. Tie long pieces of flagging every ~50-100 feet, making sure that each piece of flagging is visible from the previous one. Near the nest, tie two hanging pieces of flagging (double flag) so that someone following the trail will know they’ve arrived. If the nest is on the ground/not immediately visible, tie your flags – and write good descriptions in the nestlog – so that someone approaching will not accidentally step on the nest.

There are of course exceptions to these flagging rules; nests that are on buildings, especially at the hotel, may not need to be flagged. A nest described as “above the light on the porch of condo 503” doesn’t need a flag; one that is “on a support beam under the gallery” probably does. The goal of flagging is to make things as easy as possible for co-workers who are trying to find the nest after you; use common sense.

When you return to the lab:

Please fill out a nest log sheet, including a good description of state of the nest when you found it, the identity of the adults, and *precisely* where it is. You need be very clear about the status of the nest when it was found and how to find it again. A nest does not get a number until at least one of the adults has been identified, but it does get a nest log sheet to help keep track of its progress.

You will also need to record some of this same information in other logs, e.g., nestling weight sheets, but the nestlogs are where we turn in order to piece together what really happened to the birds, so they need to be complete. **Also, please write so that others can read your writing!**

Nest ID: Once one of the adults associated with a nest has been positively IDed, that nest gets a nest ID. Nest IDs are sequentially numbered based on the location on the study site (HTL, JT, WVS, WP, DOL, STN, WVN; rarer designations include HP (horse pasture), BK (bald knob), GD (garbage dump road) IT (indian trail), GCR (golf course road), etc., although most of these can also be designated by the closest “main” area).

The first nest by a pair always just gets the number (i.e. STN 136). That number is the number for that pair for the season; subsequent nests by a pair get a suffix. If the first nest failed, the next nest gets an “R” (i.e. STN 136 R). If *that* nest fails, the next one gets an “R2”, etc. If the first nest fledged any offspring, the next nest gets a “II” designation.

Example:	Nest 1	STN 136	fledged
	Nest 2	STN 136 II	failed
	Nest 3	STN 136 II R	fledged
	Nest 4	STN 136 III	

Later in the season, make sure that you check to see if a pair has had a previous nest that year, so that the numbering scheme is correct. (This can be particularly tricky to spot for pairs that nest on the “border” between areas; i.e. a pair that is nesting by WVN tag 14 may have had their previous nest on the STN, etc.) If a pair divorces mid-season (or one partner dies), then new nests with a new partner are designated with a “B” in addition to the II / R. If both members of a pair re-nest with new partners, typically the male retains the Nest ID, and the female gets either a new number, or becomes the “B” nest of her new partner’s previous number. There’s been some variation on this in the past; the main concern is not to duplicate nest IDs!

Male & Female Information: If you only have a partial ID, please write it here, but in pencil! Once you have a positive ID for color bands, look up the band number (and blood number) in the appropriate sheets.

Nest Location: The “general” is typically the nearest tag # or building; the “specific” should be the detailed information about where the nest is (how many meters from the nearest tag, how far up a bank, whether it’s on the ground or elevated, nearby vegetation or landscape features, how the flagging trail goes, etc.) Draw a map if it’s needed!

Treatment Group: This is primarily used in years in which we did implants; should be filled in at the end of the season.

Date/Status When Found: The status is building/laying/incubating/nestling/fledgling family. Include the # of eggs/nestlings present when the nest was found, or details about the stage of building, etc.

Date 1st Egg:

eggs laid: The minimum number of junco eggs in the nest.

Date Incubation Onset / Expected Hatch Date: Incubation typically lasts 12 days, and the female typically begins to incubate the night before the day the last egg is laid (sometimes sooner as the season progresses). So, for example, if egg 4 is laid on May 1st, we expect hatching on May 12th. In a particularly cold spring incubation can run longer, but incubation is rarely (never?) less than 12 days. For most studies we conduct, it is important that we know hatching day. Estimates need to be as accurate as possible.

For nests found after incubation has begun, note the condition of the eggs when first found. Are they translucent as if they have just been laid, or are they opaque indicating that development is underway? Or are they just about to hatch, i.e., pipped? This will help to estimate when they might hatch.

Hatched, Date: The actual hatch date of the nest. If hatching is asynchronous, please record both how many hatched on the first day of hatching, *and* how many hatched in total.

Fledged/Fail, #, Date: This should be treated as the minimum number of fledglings. For fledgling families, date of fledging can be estimated based on tail length, and how good they are at flying (see below).

Typical nest timing (27 days):

Laying	1	2	3	4																																					
Incubation				1	2	3	4	5	6	7	8	9	10	11	12																										
Nestling																0	1	2	3	4	5	6	7	8	9	10	11	12													

How often to visit a nest:

- For nests found during building, return every other day to determine when laying begins and ends so we can have accurate estimates of clutch size and hatching day.
- For nests found during laying, return daily to mark eggs during laying. Use a sharpie (see separate instructions). Be very, very careful not to harm the eggs.
- After the clutch is complete, visit every 3rd day and note whether the female is on the nest. If she is off the nest, always note the number of eggs. For nests found with eggs, visit every other day.
- For nests found after laying complete, visit every other day to get the best possible estimate of hatching day.
- For nests found after the eggs hatch, what you do will depend on the age of the nestlings, see below.

Every time you visit a nest:

Please take great care when working with nests. It’s important not to impart your scent to the nests, eggs or young. To minimize nest predation, rub your hands in moist soil or ferns before touching eggs or young.

Also we need to keep disturbance to a minimum. We don't want females to start flushing too readily because our visits have been too frequent. If they do, they presumably become higher risk candidates for predation. So please don't flush a female unless it's necessary. Rather, if she's on, come back later and see what's up (females incubate roughly 70% of the time) or do what Eric Snajdr always called a soft flush. Briefly approach from the back and flush with some vegetation in a way that is 'natural' for the female.

Please use the sign-up sheets to be sure that nests are not visited by more than one person on the same day. If someone else signed up to check the status of a nest, then leave it alone. If you do accidentally disturb a nest that you didn't sign up for, you should still record it in the nestlog.

Every time you visit a nest, be sure to record the following in your notebook and the nestlog:

1. State of the nest: During building, does it have moss/grass/deer hair? What stage is it at? Does the lining look disturbed?
2. Nest contents: Number, and stage of incubation or age of nestlings. (NOT for QPCs, see below) Are all of the eggs junco eggs? Are the eggs warm and dry? Are the nestlings begging?
3. Adults: Are either the male or the female nearby? What are they doing? What are their colors? (Even if already known, it is worthwhile having multiple people confirm parent IDs.)
4. What you did: Note if you marked eggs, measured/bled nestlings, collected cowbird eggs, captured adults, performed behavioral trials, anything at all.
5. Anything else of interest: a death, a visit from a neighboring male, etc.

Marking eggs:

We mark eggs as they are laid in order to determine laying order, for egg measurements and/or collecting eggs. For many experiments, knowing each egg's order in the laying sequence is important. Eggs can be marked a sharpie (not the fine-tip ones). It is possible to mark the eggs without removing them from the nest (one spot for egg 1, 2 spots for egg 2, a circle for egg 3). In the past we marked like this: Carefully remove egg from nest, holding it gently in your left hand. Use sharpie to mark egg with a I, II, III, or X. Don't poke the shell! Never mark with a pencil.

What to do under other circumstances: If there are two eggs in the nest at the time you find it and they are fresh, mark them both with spots. If there are more than 2 eggs or incubation is underway (eggs warm) leave them unmarked.

Return the egg to the nest. If you marked egg by removing it, then nestle the egg in your fingers and let your fingers lead the way into the nest to avoid puncture of the egg by vegetation or nest material. Gently release egg into cup of nest. Again don't poke the egg! Record what you did in the nestlog.

Collecting eggs:

When the clutch is complete, collect the third egg. If the laying order is unknown, determine which egg is largest by eye and collect it. Clutches are typically 4 eggs in the early season, 3 eggs later on. On the day *after* the 4th egg is laid, *carefully* return the collected egg to the lab. We have egg carriers that have cotton on all sides to keep the egg from cracking in transit (e.g. film canisters or empty containers that once held microhematocrit tubes).

Measure the long axis and wide axis of the egg to the nearest 0.1 mm with calipers (hug the egg, but not too tightly). Take care not to break the eggs, and if you do have an accident, please keep complete notes. Weigh the egg to the nearest 0.01 g.

Wrap the egg tightly in Parafilm (which will catch any albumin that leaks when the egg cracks during freezing) and place it in a glass storage vial with a plastic top, labeled completely, freeze. Fill out the egg data log, noting the criterion used to select egg (3rd, 4th, or largest) and the egg's dimensions.

Do not collect more than one egg from a given female. Subsequent clutches should be left complete. The goal is to obtain eggs from as many females as possible but also to anticipate that many first clutches will fail, allowing observations of behavior to be made on replacement clutches that will be of full size.

Cowbird nest parasitism:

We want to know the rate of parasitism and how it varies annually and seasonally. We also don't want to have cowbird hatchlings in our nests. If a nest found during laying and has a cowbird egg in it, leave it until after the clutch is complete. If you were to remove it earlier, you might cause the female to desert. When the clutch is complete, collect the cowbird egg and return it to the lab, record its information, and freeze it. If a nest has more than one cowbird egg, only remove one per day. Make careful notes and be sure to record in the nestlogs what you did.

Determining nest fates:

If you find a nest (either with/without eggs) and you are not sure if the nest is active (i.e. if it is an abandoned build, if the eggs are cold, etc), the best tactic is to place a small bit of leaf in the nest and note whether it is there the next day. Females that visit their nests will remove small bits of leaf, even if she does not lay an egg.

If a nest has failed, particularly a nest with older nestlings, please note the circumstances. Was the nest lining torn out? Was the bottom smooth and unmussed? Were there any feces in the nest? Look around the nest very carefully. Do you see feathers from the female? Do you see bits of shell, partly eaten babies or color bands? Look carefully to see what you can see. Look beyond the nest itself, under it if it is elevated. If you find dead babies or mother, return them to lab and freeze for later extraction of DNA. Keep thorough notes and enter them in the nest logs!!

Weighing and measuring young:

Minimum data required for each nest is number hatched at day 0, number at day 6 + blood for DNA, and number fledged.

Hatching day (day 0):

On hatching day (day 0), we need to count the number of young, weigh them, and measure their tarsi. Weighings and measurements are done in the early afternoon. Each young should be marked so it can be identified later. Take scissors and clip feathers to distinguish young. Clip head, back, both, or neither. If there are 5 young, clip something unusual (only sections of the back fuzz, the wing fuzz, etc.), and/or use a sharpie.

Previously we did this with polish as red-R, red-L, red-both, and nothing. Nail polish works fine, but it can rub off, so it needs to be applied very carefully (a piece of grass dipped in the bottle works better than the brush to mark just the toes and not the foot - it takes a little while to dry, so think of this as an important task that needs to be done carefully).

Enter on nest log that young have been weighed, and fill out a nestling weight sheet. Nests should be aged according to first day of hatching and the majority of nestlings in the nest. If two hatch one day and two hatch the next, call day 0 (for the nest) the day that the first two hatched. If one hatches on Tuesday and three hatch on Wednesday, then Wednesday is day 0. Nests should be aged by their status in the PM.

Frequently, three eggs hatch one day, and one hatches on the next day. If any eggs remain unhatched on hatching day, then return the next day to process the young that have hatched in the meantime. (The first three nestlings should be clipped on back, head, and head+back, so that the fourth will be the “neither”.) If there is still an unhatched egg on the following day but you think there is a chance that an egg will hatch (no obvious flaws, obviously developed), then go back the next day (may be day 2 for the earliest hatched young) and either process the last hatched young or bring the egg(s) back to the lab for processing.

For eggs where it is obvious that they will never hatch, bring them back to the lab and fill out an “Unhatched Egg” sheet. Note the return of any eggs to the lab on the nest-logs.

Eggs that were punctured (by us or by the birds) and are light in weight and will never hatch. The same is true of eggs that are out of balance because they have dried out (eggs like this roll in circles on a flat surface). Eggs that remain translucent never developed and can be brought back. Eggs that fail during development are often dark looking. We are very interested in evidence of this kind of infertility, so as always keep complete notes and enter findings in nest logs. For eggs that develop but never hatch, we should also preserve embryos for DNA.

Day 3:

On day 3 count, weigh, and measure young. Collect ½ to 1 tube of blood. Note which young is which by the feather clips, and associate the measurements and the blood with the individual. To prepare for taking the blood samples, take the proper number of Eppendorf tubes with 500 ul of Longmire’s and needles. It is essential that the DNA be associated with the right individual! So be sure not to switch the DNA of the nestlings; keep track of whether the blood came from head clip, back clip, both clip, neither clip, etc.

Return to lab, process blood, fill out nestling weight sheets, blood sheet, and enter data in nest log.

Always be on the look out for definitive information about who the parents are. Bands seen? food in bill? close approach and chipping? Nests with unknown adults are not worth much and it’s easy to forget about this and lose opportunities.

Day 6 (traditional day of band and bleed):

If by day 6, you are not sure who the parents are, *watch the nest until both adults have fed the young and their bands have been identified*. We simply cannot have nests in which we have blood from young but are not sure who the parents are. If you find that one of the parents has still not been

bled, alert the crew and make this a TOP priority. If the nest fails there is no way ever to be sure who are parents are.

On day 6, band and bleed the young. Be sure to use unique band combinations from prepared lists. Collect two tubes of blood from each nestling (A & B sample). Measure mass and tarsus as usual. On the banding sheet, it is critical to note which young is which, i.e., which feather clipped young measured on day 6 becomes which color banded individual.

Return to lab to process blood samples. **AGAIN, BE SURE TO KEEP STRAIGHT WHO IS WHO!** If necessary, plan to bleed the nestlings again the next day or at fledging (recalling that the nest might fail).

Record data on nestling weight sheets and nest logs and DNA sheets.

Warning: do not attempt to handle the young after day 7 – they will fledge prematurely and probably not survive.

Day 9: conduct a quick passing check, QPC

On day 9, check the nest to determine whether it is still active (do parents chip? do they have food? do you see them feeding? going to and from nest? if you can see the nestlings without getting too close to the nest, estimate the number of nestlings present.). Remember, do not touch the young or go too close to the nest after day 7 or you may cause premature fledging and the young are likely to die! Simply check it out. Don't harass the adults.

Fledging day minus one (DAY 11):

On the afternoon of day 11 (early in the spring, later in summer, i.e., post 15 June go on day 10), go to the nest, make sure it is still active, and attempt to count the young. If you are in doubt about the age of the young, please be conservative and go on what you think is day 10. **Do not touch the nest/young.** If it is a nest where you simply cannot see (way back in the roots of a treefall, but sometimes a flashlight helps), then simply note whether the nest is active (e.g., parent with food chipping at you). Then state in the nest log that there was no way to be certain of the number of nestlings without risking early fledging.

Assess the situation for catching the young and the adults the next day. Set up a net (or nets) (it helps to have 2 people to do this) and do it in a way that will intercept the adults – or at least the female - as she goes to the nest to feed the young. Consider the light (i.e., visibility of the net to the parents) in the early morning and observe behavior because the adults typically use the same path to and from the nest. Then make a decision about the best time to return the next day to do the catching. E.g., given the lighting or the traffic on the road, is this one where you need to get there at dawn if you are to be successful?

Fledging day: (not before DAY 12, later in the year this will be moved up to day 11)

(1) When to go and what to take:

Go in the morning and take the following items: Bird bucket or bag; net, poles, stakes, and hammer (if net is not already set up); cooler with ice, bleeding kit, fledgling scream tape and tape recorder; and a potter trap; Balance, calipers, bands if necessary, and data sheets

(2) Begin by catching the adults

In almost all years, we attempt to catch the adults on fledging day to check their mass/condition, and to take a blood sample if needed. BE SURE to check before you go whether there is a DNA blood sample for each adult. If the adults can be caught passively – i.e. going to/from the nest – without causing the nestlings to fledge, this is the best way to do it. If you are unable to catch the adults in this way and they still need to be caught (e.g., to be weighed, to determine the band number, to get their blood for DNA), then you can sometimes catch them by using the young. To do this, grab the young (see below) and place them in a trap beneath the net. Then use a tape of fledgling screams placed next to the trap to get the adults in the net.

(3) Process the fledglings (ID, mass, tarsus, wing, tail).

THE COUNT OF THE YOUNG IS EXTREMELY IMPORTANT. You also need to weigh them and measure their tarsi.

Be extremely alert when you go for the young, or some will slip past you, and you will spend lots of time tracking them down. Have a container (bird bucket, paper bag) ready. Approach the nest slowly with your hands in front of you. Extend one or both slowly until you are in a position to strike like a snake at the nest. When ready, strike with your open hand(s) and cover the nest cavity so that no young can get out. Close your hands over the young and nest. In other words, grab the young and the nest as a unit. Have a back-up person if possible. Put all the young in the same bird bucket (with their parents if you have already caught the parents because parents remain calmer if they are with the young).

After the young are processed, ordinarily you put them back in or near the nest. They won't stay, but the parents will know where to look for them. Various people will have ideas about how best to do this (centrifugal force, baseball caps, striped maple leaves, etc.)

If you still need to catch the adults, and you don't have a trap, then put the bucket with young beneath the net. The young will call and the adults should fly into the net immediately. Treat adults as above. If you don't catch them right away, try using the tape. Keep track of time and conditions.

If the adults finally prove too wary to be caught, then process the young. If after having processed them you still need to capture the adults, then you can use the young to catch the adults (see above). Put them in one cell of a potter trap and place vegetation around the trap so the only way the parents can get to the young is by entering the cell adjacent to the young. Put the trap near the net. Hope; and don't let the young get hurt or too hungry.

Congratulations to you and to the juncos - they are launched!

Measuring survival to independence:

This is a tough assignment = very challenging but very satisfying too and juncos are one of the few passerines in which people have attempted this, and while this is very hard, it is not impossible.

1. Our procedure is to attempt to determine the number of young that survive to 1 day after fledging, 2 days after fledging, 3 days after fledging, 6 days after fledging, 9 days after fledging, and 14 days after fledging. The families move around a lot, and you may find them one day and not find them the next. Still we do a census on each of these days.

When looking for young use all your senses (including ESP). *The key to success:* is to go prepared. Have the information about band colors written down, so you know what leg and colors to look for. You will get only a few seconds, so preparation will make all the difference.

2. The day-14 measure is the most important, but you have to visit the territories all along in order to know the likely places to look on day 14. See data sheets for recording fledgling sightings. Be sure to record what you saw ASAP, so others will not duplicate your efforts.

3. If on day 14 you find all the fledglings that you think are alive, then you can quit looking. If you have not seen one or more of them, keep going to the territories even after day 14 (says days 15 and 16 and 17, until you are CONVINCED that certain young are not alive. On day 21 you may quit in any case.

4. Return to the lab and record your findings on the fledgling survival sheets. Note where you looked, how much time you spent, how sure you are of your observations, etc.

5. Be on the look out for signs of re-nesting. Is the female still around the fledglings? Did you see her feed young or just the male? I think that the earliest turn around is a case in which a female laid her first egg 8 days after the first brood fledged. But 12 to 15 days is more common.