Mixing in (some) CKMR setups

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This is about basic properties of CKMR when your samples might not be "well-mixed". That could mean several things— this document just covers cases where:

1. there are several discrete spawning sites[1][2];
2. each adult tends to persistently breed at the same site;
    (a) NB it doesn't matter whether that preference is heritable or not;
3. you can't tell genetically which site an animal was born on, or breeds at[3];
4. some of your adult and/or juve samples are not "pure", ie they contain a mix of either juves that were born on different sites, or adults that breed at different sites.

The main intended audience is primarily me ;) or people like me who want to understand why/when/how things work or don't, at a semi-mathematical level: in particular, so you can confirm to yourself, or not, that the reasoning behind my general recommendations is OK (and please tell me if you don't think it is!). But it's not actually very mathsy, and the conclusions and general logic should be somewhat more generally accessible... I hope. I'd recommend reading this *fairly quickly* the first time, skipping all footnotes[4], and not getting stuck at the equations (though they are very simple— and some make important points). There's several places where I included long splurges of text, partly for my own benefit, that maybe should be somewhere else or nowhere; don't get stuck on those. This paragraph might be one of them.

The trigger for writing all this, is 2021 discussions about possible CKMR schemes for E-ABFT— although this document is *not* just aimed at E-ABFT. Actually, we did a big report on E-ABFT in 2017, which contained quite a bit of discussion about mixing; but because of the open-endedness of the sampling schemes that we considered back then— given the seeming impossibility of getting clear background information about anything— the conclusions were harder to interpret[5].

[1] Several *important* ones, that is. Unimportant ones are... unimportant.

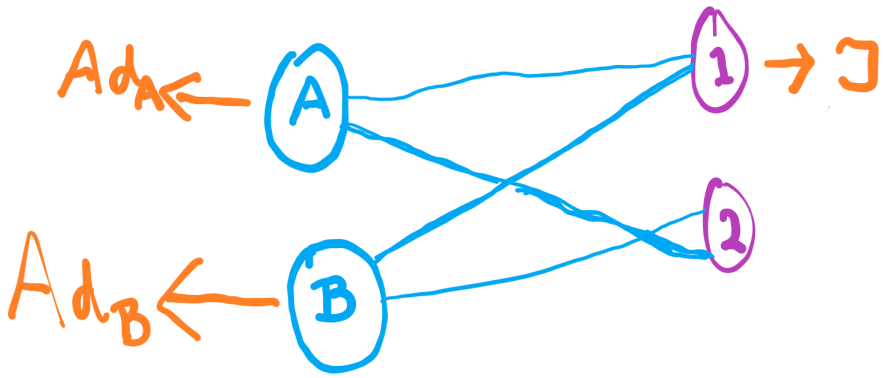
[2] If your situation is more about continuously-distributed species with potentially limited dispersal, but without discrete breeding sites, then see Conn et al. (2020).

[3] If you could, then you'd just do genetic assignment of individuals and run CKMR separately on each site.

[4] Did you? Ha!

[5] I think; I can't remember, and don't feel like re-reading. But for one thing, we were expecting mixed-site juveniles at that time. And then the fishery-in-question *closed down* around the time of the study! A few important things have changed since then, most notably the serious possibility to get large numbers of pure juveniles (larvae).

Figure 0.1: Basic setup. Juves are sampled only from spawning site 1; adults from sampling grounds A and B. Each sampling-ground-sample contains a mix of adults that spawn at site 1, and adults that spawn at site 2.



This time, to make things tractable (as well as more relevant to E-ABFT c. 2021), I'm going to specialize it a bit, in particular WRTO juvenile sampling. The spatial aspects here *are* relevant to E-ABFT; the demographics have been heavily simplified but most of the conclusions should still apply— there are some gotchas around that, as noted later (if I remembered to do so).

The spatial setup is as per Figure 0.1.

1. You get pure juvenile samples, but only from 1 breeding site.
2. You get mixed adult samples from one or more locations that are unrelated to the breeding sites;
3. Only considering females, with all female adults having equal fecundity (a "mammal")[6].
   (a) and female adults don't change their breeding-site preference over time[7].
4. Population in equilibrium— not considering time[8].
5. Focused on estimating *aggregate* abundance, i.e. the total N of female adults that breed at either site 1 or site 2;
6. Mostly concerned with POPs, though HSPs also mentioned in the last bit.

9

[6]Real CKMR has to consider male (adults) as well; they're not an optional extra! But they don't complicate the underlying logic here; you "just" have to do everything in a sex-disaggregated way.

[7]But it doesn't matter whether or not their offspring inherit Mum's preference for that site. IE there may be *absolutely no genetic signal at all even in principle* between juvenile sites. Of course, even if there was one because of heritability, there's a huge question about whether one could even detect it the presence of (realistic) low levels of changing preference within lifespan— i.e. the usual problem with pop gen "false negatives". Anyway, the point is that heritability is not the point— it's *within-adulthood-fidelity*.

[8]There's a caveat about this later on...

[9]Pure samples are not *necessarily* desirable for CKMR; IMO too many geneticists have an unhealthy obsession with "samples of pure origin"! If every sample was pure, then you'd need to do CKMR separately for every single breeding site, and that might be very onerous and not necessarily give the results that are useful for management—

Formal definitions

Personally I recommend you **skip over this box** initially, and just try to go with the flow. But if you are reading this for the 2nd+ time, or are someone who prefers to start with explicit albeit rather cumbersome and legalistic definitions (sometimes that is a Good Thing, sometimes it isn't), then here are some definitions related to "mixing" as the term is used in this doco. NB this is somewhat specific to the discrete-breeding-site setup outlined above; different biologies (e.g. clinal structure) might call for different definitions.
A sample from some location is:

Pure if all the adults in it breed at the same breeding-site, and all the juveniles in it were born at the same breeding site. It may contain zero adults or zero juveniles; this is a mathematical definition, remember!

Well-mixed if the adults and/or juveniles in the sample are in proportion to the adult and/or juvenile abundances in the population-at-large.

Not-well-mixed if the sample contains adults that use several different breeding sites, but not in proportion to their abundances in the entire population.

Mixed could in principle mean any of the above, legalistically speakingwise. In practice it probably means you're not sure in advance which one applies, except you probably don't think the sample is pure.

And while I'm being all legalistic, I suppose I should add that "well-mixed" and "pure" don't have to be 100% accurate. If the sample is 95% pure then it's pure enough to be called pure. The usual purpose of all this stuff is **to make sure "we're" not killing too many animals**; exactness is not necessary, and everything always gets estimated with uncertainty anyway because that's how the universe is, but we still have to deal with it, so *reasonable approximation* is at the core of everything. And sorry, but no I'm not going to define "reasonable"...

Table 1: This is a box actually...

1 Well-mixed

Let's suppose we only have adult samples from A not B, but we think the adults are likely well-mixed— that is, in proportion to the abundances of adults that breed at site 1 (site-1ers) and at site 2 (site-2ers). What's the prob of a POP[10] between adult Ann at A and juve Jill sampled at 1? It depends on whether Ann is really "from"[11] site 1 or site 2. We get:

$$\begin{aligned}
&\mathbb{P}\left[K_{\text{Ann,Jill}} = \text{POP} | \text{Ann at } A, \text{Jill from 1}\right] \\
&= \mathbb{P}\left[\text{Ann is from 1} | A\right] \times \mathbb{P}\left[K = \text{POP} | \text{both from 1}\right] + \\
&\mathbb{P}\left[\text{Ann is from 2} | A\right] \times \mathbb{P}\left[K = \text{POP} | \text{from different sites}\right] \\
&= \frac{N_1}{N_1 + N_2} \times \frac{1}{N_1} + \frac{N_2}{N_1 + N_2} \times 0 \\
&= \frac{1}{N_1 + N_2}
\end{aligned} \tag{1.1}$$

The point here is that Ann cannot be Jill's mum if Ann is actually a site-2er, which is where the "0" comes from. But if she is actually a site-1er, then she has an equal chance as any of the other site-1ers of being Jill's mum.

BTW I'm not going to explain the notation in general: but K is the nature of the kinship between two animals, which can be eg UP (Unrelated Pair), POP, HSP, etc.

In theoretical practice, we would do lots of comparisons of the Ann vs Jill kind, and then estimate that probability empirically— ie the A1 POP-rate. We could theoretically then use the reciprocal of the empirical POP rate to get an estimate of $N_1 + N_2$. [DON'T ESTIMATE ABUNDANCE THIS WAY FOR REAL[12]!]

Conclusion: if the adult sample is well-mixed, POPs will still estimate the total abundance of adults even when your juve samples are pure samples from just one site.

even enlightened management. That said, the formulae here are all assuming that the juvenile sample *is* pure— and this does seem to lead to cleaner conclusions than I remember seeing in 2017, when IIRC neither adult nor juvenile samples were expected to be pure nor to be well-mixed... So a bit of purity is probably good, but there's no need to go all OCD about it! The opposite end of purity is "well-mixed", and *that* is the ideal for CKMR. The murky area is in between purity and well-mixity...

[10] Actually a MOP, ie Mother-Offspring Pair.

[11] "From" meaning "breeds at". It doesn't matter where Ann herself was born, just what she does as an adult.

[12] The "invert the rate" notion corresponds ONLY to "the pink-and-blue cartoon fish", where there is just a single type of comparison between adults-all-of-which-are-identical and juveniles-ditto-blah-blah. You should NEVER try to construct an estimate that way on real fish data, because of size growth time everything— there are many different types of comparison each with its own probability formula, so build a proper CKMR model and let the computer do the job properly. However, the general notions about mixing are applicable in either case. I hope.

2 Not well-mixed

Now suppose that the site-1-adults and the site-2-adults differ in their per capita tendency to go to A (as opposed to B, C, or elsewhere); specifically, we now have

$$\mathbb{P}\left[\text{Ann is from 1}|\text{Ann at } A\right] = \frac{\alpha_1^* N_1}{\alpha_1^* N_1 + \alpha_2^* N_2} \tag{2.1}$$

So if $\alpha_1^* > \alpha_2^*$, it means that site-1ers are over-represented at A compared to site-2ers, and conversely.

It turns out to be annoying to work with eqn (2.1) in exactly that form, so I'll reparametrize it as

$$\mathbb{P}\left[\text{Ann is from 1}|\text{Ann at } A\right] = \frac{N_1}{N_1 + \alpha N_2} \tag{2.2}$$

where the "representation ratio" $\alpha \triangleq \alpha_2^*/\alpha_1^*$. So if $\alpha > 1$, then site-2ers are over-represented, or under if $\alpha < 1$.

The POP formula for Ann vs Jill is now

$$\begin{gathered}\mathbb{P}\left[K_{\text{Ann,Jill}} = \text{POP}|\text{Ann at } A, \text{Jill from 1}\right] = \\ \mathbb{P}\left[\text{Ann is from 1}|A\right] \times \mathbb{P}\left[K = \text{POP}|\text{both from 1}\right] + \\ \mathbb{P}\left[\text{Ann is from 2}|A\right] \times \mathbb{P}\left[K = \text{POP}|\text{from different sites}\right] = \\ \frac{N_1}{N_1 + \alpha N_2} \times \frac{1}{N_1} + \frac{\alpha N_2}{N_1 + \alpha N_2} \times 0 = \frac{1}{N_1 + \alpha N_2}\end{gathered} \tag{2.3}$$

which is clearly biased unless $\alpha \approx 1$. The other case where bias is low, is if $N_2 \ll N_1$; then bad-mixing doesn't really matter unless N_2 is massively over-represented at A. If $\alpha > 1$, i.e. if site-2ers are over-represented in your adult sample relative to site-1ers, then the bias will be positive, or negative if $\alpha < 1$.

Since there's only one piece of data on the table (the empirical A1 POP rate between site-A and site-1) you obviously can't estimate all 3 parameters in eqn (2.3), and unfortunately you can't estimate the combination that you really want either (i.e. $N_1 + N_2$).

Conclusion: if you only sample one adult ground (as well as your pure juveniles) and it's substantially not well-mixed, *and* if all you do is compare those adults to your juveniles, then you'll get bias that can neither be detected nor fixed. Direction of bias depends on which breeing site is over-represented (see above). But, maybe you can get more out of the data; see below.

2.1 Extreme cases

If $\alpha \gg 1$, i.e. if it's much less likely for a site-1 adult to go to A than it is for a site-2er, then the estimate will have a huge positive bias. Basically you are getting your juves from one "population" and your adults from a different one. So a naive application of oversimplified CKMR formulae would, of course, give a completely stupid answer (luckily with absolutely massive error bars, because you'll have very few kin. And the HSPs provide a sanity check— provided you've got a decent number of those. See later).

If $\alpha = 0$ then you have a pure sample of site-1 *adults* and you are just estimating their abundance, not the site-2ers.

If you are considering working with just one adult sampling ground, then you really need to be confident, in advance, that these extreme cases are very unlikely (or, if $\alpha \approx 0$ is plausible, that you don't mind the implication). Really, you should have a reasonable expectation that that single adult sampling ground should be not too far from well-mixed.

2.2 But that's not all we know!

If there's good age data, we can do something else: we can look at POPs just *within* the A-samples ("AA POPs"). What is the prob that Amelia is the mother of Alison? Thanks to the convenient assumptions (equilibrium, and mammaleseque dynamics, and good age) that I smuggled in earlier, we have

$$\begin{aligned}
&\mathbb{P}\left[K_{\text{Amelia,Alison}} = \text{POP} | \text{both at A, \& Amelia was alive and mature when Ali was born}\right] = \\
&\qquad \mathbb{P}\left[\text{Amelia is a site-1er and Alison was born at 1}\right] \times \frac{1}{N_1} + \\
&\qquad \mathbb{P}\left[\text{Amelia is a site-2er and Alison was born at 2}\right] \times \frac{1}{N_2} + \\
&\quad \mathbb{P}\left[\text{Amelia breeds at a different site to where Alison was born}\right] \times 0 = \\
&\qquad \left(\frac{N_1}{N_1 + \alpha N_2}\right)^2 \times \frac{1}{N_1} + \left(\frac{\alpha N_2}{N_1 + \alpha N_2}\right)^2 \times \frac{1}{N_2} + 0 = \\
&\qquad \frac{N_1 + \alpha^2 N_2}{\left(N_1 + \alpha N_2\right)^2}
\end{aligned} \tag{2.4}$$

So now we could conceptually equate this to the empirical AA POP rate (restricting comparisons to those that make sense— see the conditional in the first line of that equation) and now we have 2 equations. But there are still 3 unknowns (N_1, N_2, α) and only two observations, and you can't squeeze out an estimate of $N_1 + N_2$. On the plus side: if you had been thinking that the adult sample was well-mixed (i.e. that $\alpha = 1$), then these two numbers (A1 and AA POP rates) *may* give some power to detect any substantial violation of that assumption... let's look at that now.

2.3 Bounds

I will start this by saying that mostly *I do not advocate deliberately fitting a "wrong" (deliberately naive over-simplified) CKMR model and then trying to second-guess out how wrong it might be*. The *right* thing to do, is to organize your data collection so that you can build and fit a model that makes biological sense, as per the ERRO principles in Bravington, Skaug, and Anderson (2016); then you won't have to worry about bias. CKMR projects in fisheries are big tasks that involve a lot of blood, sweat, and tears[13] to get started (though they are *easy* to continue once everything is working)[14]. For once in fisheries, CKMR actually lets you do something *right*, without having to spend years arguing-the-toss afterwards about exactly how wrong it was; so why not do it properly?

OK, that said: sometimes it might be legit to start a project with simpler models in mind for the short term, *provided* you are confident that in the longer term the CKMR data (or some other piece of magic) *will* allow you to sort out what's really going on. (In contrast to, for example, CPUE data— where your structural uncertainties remain uncertain over time, and in practice usually get worse. Just sayin'.)

For the record, I don't think you should resort to single-imperfectly-mixed-adult-site CKMR in this setting unless you really cannot sample from any other adult ground (see below), and unless you are completely desperate. Anyway: what if we did have lots of AA and A1 POP data from 1 pure juvenile and just 1 mixed adult sampling ground, as just described? We know we can't estimate exactly what we want (3 parameters, 2 "data" i.e. empirical POP rates)— but if we did fit known-wrong model(s) in the short term, could we place any bounds or say much about bias from misapplying eqns (2.4) and (2.3)— see footnote[15]— as if they really were estimating total abundance?

Let's say we observe empirical POP rates c_J and c_a which corresponding to eqns (2.4) and (2.3), which should satisfy:

$$\begin{aligned} \mathbb{E}\left[C_J\right] &= \frac{1}{N_1 + \alpha N_2} \\ \mathbb{E}\left[C_A\right] &= \frac{N_1 + \alpha^2 N_2}{\left(N_1 + \alpha N_2\right)^2} \end{aligned} \tag{2.5}$$

Suppose we form two "naive estimates" that we know are biased for total abundance: $1/c_J$ and $1/c_A$. Can we place any bounds on real abundance?

To simplify the algebra, define $\rho \triangleq N_1/N_2$ and then "without loss of generality" set $N_1 = 1$, so

[13] The blood comes from the dead fish; you supply the sweat and, if you do it wrong, the tears as well.

[14] But usually *not* a lot of money, despite what people think— compared to fishery value.

[15] Of course, for real-world fish breeding several times, you are going to have to build a fairly proper CKMR model anyway, because of time-lags, size-specific fecundity, variability in growth, and uncertainty in age. You weren't thinking about just taking the reciprocal of some rate, were you? Of course not! Good. The "naive" and "misapply" stuff here only relates to the *spatial* (mixing) side of things— the implicit assumption here is that all the other bits of a CKMR model will be done right.

that the true total is $1+\rho$. Obviously, $\rho > 0$. Equating observed to expected (because we want to see what the bias would be), we have

$$\frac{1}{c_J} = 1 + \alpha\rho \tag{2.6}$$

$$\begin{aligned} \frac{1}{c_A} &= \frac{(1+\alpha\rho)^2}{1+\alpha^2\rho} = \\ &= (1+\alpha\rho)\,\frac{1+\alpha\rho}{1+\alpha^2\rho} \\ &= \frac{1}{c_J}\frac{1+\alpha\rho}{1+\alpha^2\rho} \end{aligned} \tag{2.7}$$

We can see the following:

1. If $\alpha = 1$ (i.e. no over/under-representation) then there's no difference in expectation (and no bias). So, reasoning the other way, if you have plenty of A1 *and* AA POPs and the two rates are also similar[16], then bias should not be a concern.

2. It's clear from eqn (2.7) that the AA-version will be bigger than A1 (on average) iff $\alpha < 1$. So we can check over/under-representation of site-2ers.

3. What about bias? It's also clear from eqn (2.6) that the A1 estimate is negatively/positively biased for total abundance according as $\alpha \lessgtr 1$.

   (a) It's true, but much less obvious, that the AA estimate is *always* negatively biased regardless of α (see section 6).

   (b) So when $\alpha > 1$, the two biases have opposite signs, and we kinda have bounds; they are *stochastic* bounds though, because the two estimates (which are independent) are both noisy.

All those "bounds" and "clearly different" and "similar", are stochastic— they depend on having found enough POPs of both types (A1 and AA) to make statistically defensible claims. Now, I am not going to encourage people to do this, because I don't recommend fitting deliberately biased models, but Table 2 summarizes bounds-and-bias for this situation.

Conclusion: If you only sample one adult ground (as well as your pure juveniles) and it's substantially not well-mixed, but you have enough samples and good age data to look for POPs just within the adults as well as across to the juves, then you can work out a fair bit about mixing and possible limits on abundance. Getting to that point might take some time, though.

[16] You need a reasonable *number* of POPs, in both categories, to keep down the statistical noise. But the quantities you actually compare are the *rates* of POP-per-comparison. Of course, you don't actually just compare two empirical rates because, as stated ad nauseam throughout this doco, you need to build a proper CKMR for teleosts, even if it's naive about mixing...

Table 2: Don't try this at home, folks: direction of bias in **naive estimates** from single-mixed-adult-sampling-ground POPs. Instead, go sample **more than one** adult ground! Signs are direction of bias; more symbols means worse (within that row). "AA" means "estimate based on AA POPs", etc.

	A1	AA	average	Least bad option / bounds
AA>A1	- - -	-	- -	Underestimate; use AA
AA<A1	+ +	-	+	Between[*] AA and (AA+A1)/2

[*] The straight average has a positive bias unless α is very large and ρ is very small— i.e. site-2 abundance is tiny, but they are massively over-represented in your adult samples.

2.4 Reality check: time and sample size

There's a shortcut in the notation here, because comparisons need to be "backdated" to the birth of the potential offspring. Hence some of the N's aren't actually the same as each other, because they pertain to different times. The general hope is that this can all be glued together in an age-structured pop dyn model when real data (definitely including HSPs... and perhaps catches) are used— as it does in "homogenized" (fully-mixed) CKMR settings. Of course I should use extra subscripts[17], but then you can't just blithely equate things and see whether "it should all work".

For a related reason, there may be a lot fewer *usable* AA POP comparisons than A1. You have to have one of your sampled adults being much older (at least the age-of-maturity) than the other. This is mainly why I have historically not paid much attention to the utility of AA POPs outside a few specific cases. In particular, with SBT there's been no point; with maturity at age 8, and adults not really getting into the swing of breeding effectively until say age 12 (so that in the first few years, we could only have expected any AA POPs between our very youngest and our oldest— 20yo+— adults), there just weren't many informative comparisons[18]. The *other* reason I have not thought about AA POPs in general, is that you need pretty good age information on the younger adult (the potential Offspring). For many species, I'd been expecting that adult age would have to be fudged from length alone (i.e. no otoliths nor epigenetic age).

So in case you are thinking "*aha! we can just sample one adult ground, fit two bad models, and get quickly get bound(s)*"— well, you do need to think about the time stuff and whether the sample sizes will actually allow that. If the project design is "economy bargain" and aimed only at *initially* getting enough A1 POPs and HSPs to fit a basic naive model (for cost reasons, which might be quite sensible), then don't expect to be able to magically get bounds and bias in the same timeframe from the same sample size.

[17] As a mathematician, here's what I've noticed about why people (including me) tend to get equations wrong sometimes:

1. Half the time it's because they *haven't put in all the subscripts they should have* (as per the warning just now);
2. Half the time it's because they put in *too many* subscripts, and got hopelessly confused;
3. And half the time it's because they just screwed up the algebra.

[18] We probably do have a few SBT AA POPs by now, and they might be mildly interesting to look at I guess, but it's not essential information.

I reckon the way to look at it is: over time, as your sample size accumulates (probably beyond what you originally planned), you will eventually get enough extra information to check your assumptions (e.g. that mixing is pretty good, with $\alpha \approx 1$). So if your mixing assumption was not too grossly wrong in the first place, then you probably won't make too bad a mess of management before you detect that. All depends on the numbers, innit.

3 A second adult sampling ground?

If we also sample not-necessarily-well-mixed adults at ground B, what can we then do? A lot more! Say the representation ratio at B is β rather than α. So now we can do comparisons between ground-B and site-1 as well:

$$\mathbb{P}\left[K_{\text{Betty,Jill}} = \text{POP} | \text{Betty at B, Jill from 1}\right] = \frac{1}{N_1 + \beta N_2}$$

and also within site B. And for a site-B site-A adult-adultcomparison,without over-explaining the notation which should be pretty obvious, the prob is

$$\begin{gathered}\mathbb{P}\left[\text{Belinda is Amy's mum}\right] = \\ \frac{N_1}{N_1 + \beta N_2} \times \frac{N_1}{N_1 + \alpha N_2} \times \frac{1}{N_1} + \frac{\beta N_2}{N_1 + \beta N_2} \times \frac{\alpha N_2}{N_1 + \alpha N_2} \times \frac{1}{N_2} + 0 = \\ \frac{N_1 + \alpha\beta N_2}{\left(N_1 + \alpha N_2\right)\left(N_1 + \beta N_2\right)}\end{gathered} \tag{3.1}$$

In all, we now have 5 pieces of POP data:

A1 POPs; B1 POPs; AA POPs; BB POPs; AB POPs

and 4 parameters $(N_1, N_2, \alpha, \beta)$. I think this is statistically identifiable[19] (see Appendix), and even has one spare degree-of-freedom for an internal cross-check; in particular, you can produce separate estimates of N_1 and N_2— which might of course be interesting in their own right— and you can add the two together.

So, a second adult sampling ground is a good thing. In fact, *technically speaking*, given huge enough sample sizes to yield enough kin-pairs to make all 5 empirical POP rates precise, you could in theory estimate the total abundance of mammal-like-fish-in-equilibrium using samples from one pure juvenile site and several not-well-mixed adult samples, without needing any well-mixed adult samples.[20]

[19] IE: given huge sample sizes, and if the underlying model is correct e.g. no other important breeding sites, then you'd be able to estimate all four parameters accurately. Well, the technical definition of "identifiability" is actually about determinants of a Hessian IIRC, but let's not go there just yet ;)

[20] That sentence is quite lengthy just to minimize the risk of it being quoted out of context a la "*you can just do whatever you want and it will all be fine la-la-la*". You can't. So don't.

But there are some caveats on that. Clearly, if $\alpha = \beta$, there's no extra information from having a second adult sampling ground; so you actually need quite *different* amounts of over/under-representation in the two sites. And if $\alpha \approx 0$ and $\beta \approx 0$, you haven't got much chance of estimating N_2 (because you sample almost no fish from site 2). And I strongly suspect is that *precision will be pretty low* at the level of individual site estimates (N_1 and N_2) for the kinds of sample sizes you can afford in the short term, unless you have somehow a good a priori idea of the value of α and/or β. The most likely example would be if it just makes sense that one of them is close-to-well-mixed on biological grounds. Sometimes tagging data might be very useful for that, but this is not a blanket mandate to go out and tag-tag-tag "because CKMR needs it". I try to design CKMR sampling so that "I" *don't* have to collect very expensive additional data.

Conclusion: with 2+ adult sampling grounds, you're in much better shape to validate a well-mixed assumption, and if it proves false then perhaps even to allow for it and get to an unbiased estimate in the longer term.

Remark 1. Someone should (I might eventually, but not soon) look into precision in a slightly more numerical/mathematical way— sticking to this simple 2+1-sampling-sites setup, but just considering different values of the $N_1 :: N_2$ ratio, α and β, and numbers of kin-pairs per category. I do suspect there are directions in N, α, β space that are quite poorly identified for *some* combos of ratio and rates, and others that are OK— which may or may not include "total N". That would sort-of extend the "bounds and bias" discussion of the one-adult-site case, in a more generic (but also more computation-dependent) way[21].

4 More options and more things

4.1 If there are more breeding sites...

What happens in the above if there are not 2 but actually 3 important breeding sites (ie N_3 which you suspect may be of similar size to N_1 and/or N_2, and suspect may be substantially mixed into

[21] I originally wrote this: "*There is some interesting maths around how to look at this in a general sense, and some more brute-force tools of varying brutishness. If you are that someone, please try not to "just simulate". Simulation is too often the crutch, and too often requires preposterously inordinate amounts of crutch-polishing.*" That is a bit of a hobby-horse of mine in fisheries... though it does not mean I am always opposed to simulation. *If* simulation helps you develop your intuition, and *if* you can avoiding spending years dealing with details that are unavoidable when writing code but that will be basically irrelevant to conclusions, then sure. I prefer algebra when I can do it, but there are limits to that.

On further thought about *this* case, I suspect algebra cannot take us much further, and something numerical might be unavoidable. But you don't need to simulate, at least not in the usual sense. In the case of CKMR (and a lot of MR, plus *a few* more classes of statistical design problems), there are some magic formulae from statistical theory that get you straight to the Hessian *without* any simulation. I reckon everyone should use them (I do), but since the CKMR ones aren't actually published yet, it is a bit much for me to complain when people don't! I think pretty much all the formulae are in a talk at ISEC 2014 or 2016 (Bravington/Skaug)... they are hinted at in BSA 2016, too, but there's a huge gulf between seeing those formulae and writing usable code. What I personally would do *here*, is to set that stuff up (it does need guesstimates of parameters and putatative sample sizes, and full CKMR model code, but it doesn't need data) and then look for eigenvectors in the Hessian that have large/small eigenvalues.

the adult samples from A, B, etc)?

First, we have to expand the definitions of mixing (and the number of parameters) so that

$$\mathbb{P}\left[\text{adult from A is site-2er}\right] = \frac{\alpha_2 N_2}{N_1 + \alpha_2 N_2 + \alpha_3 N_3}$$

etc. That is: we've added a subscript to α and β to distinguish site-2er properties from site-3er properties.

The POP formula become things like

$$\mathbb{P}\left[K_{\text{Amelia,Ann}} = \text{POP}\right] = \frac{N_1 + \alpha_2^2 N_2 + \alpha_3^2 N_3}{\left(N_1 + \alpha_2 N_2 + \alpha_3 N_3\right)^2} \tag{4.1}$$

and the sharp-eyed will notice that this is *not* quite the same as what you'd get by lumping N_2 with N_3 and pretending that a common α applied to that composite (look at the exponents). There is no single composite α and composite N_{2-3} that could *exactly* duplicate all the probabilities.

Clearly, you couldn't estimate $(N_1, N_2, N_3, \alpha_2, \alpha_3, \beta_2, \beta_3)$ (7 parameters) from just two adult sampling locations (5 empirical rates). With three adult sampling locations, then I think you have 9 empirical rates (A1 B1 C1 AA BB CC AB AC BC) so yeah *technically* you might be able to identify all the parameters given exceedingly large sample sizes— but I imagine the precision of individual parameter estimates would be dismal for many α, β values. Neverthless, the lumping approximation might be completely adequate for practical purposes as long as α_2 and α_3 are not expected to be that different, and ditto β; and also if N_3 is just small full-stop, or downright irrelevant for other reasons (e.g. not thought to be a big part of the catch, which is presumably the thing that is being managed). But all this might be going too far anyway; if even a 2-breeding site model couldn't be fitted reliably without some assumption of well-mixing, then worrying about how good an approximation you'd get to a 3-breeding-site reality is beside the point.

4.2 Fickleness

What if some (female or male) adults are fickle, and *do* change their breeding-site preference occasionally during adulthood? My hunch is this just makes everything better-mixed and reduces any bias problems; but it can stay as a hunch for now!

You can detect fickleness directly, and estimate its rate, if you sample enough juves from 2+ pure sites— by looking for HSPs across the sites, and how that proportion changes with increasing birth-gap. It's a very nice exercise; whether it's necessary is up to you.

4.3 What if we sampled more pure juvenile sites?

Well, yes, you probably could detect and/or eliminate bias in a few more scenarios. And you might be able to do better at estimating "site abundance" (at breeding sites, not on the mixed adult sampling grounds)— I haven't looked hard at that, but the discussions of identifiability above actually do involve site-specific estimates anyway. And you certainly can check for breeding-site-fidelity, as per previous point.

But (this is a bit E-ABFT-specific now) doing so would presumably require similar numbers of samples at each juve site as at the first one— i.e., a lot. And if that needs dedicated surveys, then you really have to question whether it's (a) worth it, and (b) has any chance of actually happening; if not, should it stand in the way of just going with one pure juvenile sampling site? I don't have the answers in your case, whoever you are, but those are some questions to consider :)

4.4 "CKMR at least gives you a lower bound on abundance..."

Well, sort-of... be careful here.

If you collect lots of samples and find hardly any kin-pairs (not enough to fit a model), then intuitively CKMR does still tells you *something* about abundance; if the abundance was tiny, then you would have seen plenty of kin-pairs, but you didn't, so the abundance isn't tiny. "In theory", even finding zero kin-pairs lets you construct lower confidence intervals on abundance! In particular, if you design your sample size etc so that it should give you enough kin-pairs for a nice precise result *based on an existing assessment*, and then you collect them, and then you find hardly any pairs: well, you did the best that can reasonably be expected and you can conclude that the assessment was far too pessimistic. You don't have a precise estimate like you hoped (at least, not until you collect further samples), but you definitely know that the true abundance is substantially higher than you thought! Happy times!

However, that lower-CI claim is only valid if you are confident about model structure[22], eg when you're sure mixing isn't a big issue (or e.g. that you've got well-mixing etc). In "nasty unknown mixing" cases when you are relying on the data to disentangle the true biology (and therefore the appropriate CKMR model equations), you mayn't be able to make the claim. For example, if the truth is that each of two adult sampling grounds is in fact pure WRTO a different breeding site (rather than being mixed or even well-mixed, as you had been assuming), then you'd easily detect that with a reasonable number of POPs— but if you find 0 POPs in each simply because your sample size was too small, then you learn 0 about it, and you don't realize that your basic assumptions were wrong.

[22] And to some extent about other CKMR-related parameters such as how fecundity changes with age/size, which you were expecting to have enough CKMR data to estimate. No kin-pairs means you have to plonk in guesses for that sort of thing, and you should put confidence intervals on your guesses, and... messy. Possibly still useful in desperation, but *your* problem not *mine*!

In general, I guess it's probably true that you need fewer kin-pairs to detect big qualitative differences, than you might need to get a decent "CV on N" (or whatever objective you then decide to retro-fit onto the project...). But the difference in requirements may not be as big as you'd hope. Everything is based on counts of kin-pairs, so variance-equals-mean applies, and you can consider the numbers in that light. Is 70 kin-pairs different from 100? Yes it is. Is 16 different from 23? Mmm, maybe, not sure. Is 6 different from 9? No idea.

Incidentally, if you have a clearly-too-small number of samples and no kin-pairs, and you are confident enough about the model to go ahead anyway, then your lower confidence interval may exist mathematically— but it will be useless. EG it would not be not worth spending $100K "because that's all we could raise" merely to prove that there are at least 1000 tuna left in the sea— if say 500,000 are being caught every year! All numbers made up of course.

4.5 How should the data really be used?

This is the obligatory health warning that I feel compelled to put into all such documents...

It is completely a bad idea to try to bodge up actual estimates directly from CK data using the kind of simple formulae given here. The right way to tackle "spatial close-kin" (*if* you have to tackle it— you might be able to dodge it if you can sample in such a way that you're comfortable with assuming adequately-well-mixed) is to write down explicit formulae for the kinship probs of each comparisons that allow for spatial location: given that Hilda was sampled Here, what's the prob she was over There when Thelma was born, etc? Such formulae would (in the case of a fish like E-ABFT) look very similar to the ones here— they are not complicated in themselves, but they do need to include all the subscripts that I've left out! The point is that there are lots of different formulae, depending on the covariates of the pair under comparison (dates, ages, places, sexes, sizes, ...; well, that's probably "all", actually, so no more than a 10– or 11–dimensional array ;). Then you plonk it all into the age-structure pop dyn model and let the computer do the estimation for you. BSA 2016.

If you put together the "proper" model, i.e. that implements a sensible-without-being-ridiculously-fussy qualitative version of the biology and sampling details, then you are **guaranteed** not to have bias[23]. However, you may find that the parameters are inestimable— ie have enormously large CVs. (Clearly, that's exactly what would happen in some of the above cases.) That does *not* give you an automatic excuse to merely fit a massively over-simplified model and trust it completely! Skill and sense are required...

[23] Asymptotically, that is; IE as your sample size "approaches infinity". Whereas if the model is wrong you will get bias regardless of sample size. However, that bias is IME usually much smaller than the CV, so it's not very important. Asymptotic lack of bias— the technical term is "consistency" in statistics— is as much as one can ever ask for; true small-sample unbiasedness is a furphy/mirage.

5 Recap / suggestion

All this is predicated on the idea that you will have pure juvenile samples from just one site.

Everything is then fine, even with just one adult sampling ground, if your adults are well-mixed. If you think it's pretty likely that they are not too-far-from-well-mixed, then it might be best to start just by *making* that assumption, preferably adding a second adult sampling ground as insurance; CKMR could well get you to a far better place than whatever deep cave of ignorance you are starting from. (If you have to add the caveat that there are some adults persistently using other breeding sites, which will just not be included in the abundance estimate, then it might be best to just see if you can live with that qualitative caveat; after all, you are doing all this work for a *reason* which is presumably more financially compelling than "because it's science"— such as, we need to know how many there are so we can avoid killing too many. So additional sites unrepresented in your adult sample(s) may or may not matter to you.)

Even with just one adult sampling ground, you should eventually be able to say *something* about mixing and to put at least some bound on total abundance[24]. But by adding a second adult sampling ground— assuming it's got different mixing characteristics to the first one— you gain an ability to quickly *cross-check* your adult sampling grounds to look at the validity of well-mixing[25]. If there is no discrepancy over time, then they are either both well-mixed, or *equally-badly-mixed*— which might be deemed an implausible coincidence. And over time you can build proper unbiased models based on two adult grounds (provided there are only two important breeding sites). I think it's OK if one ground is likely well-mixed but you don't expect the other one to be; for one thing, you are still going to get fec-at-age and that helps a lot[26]. If one adult ground is actually "pure" (i.e. taken from the same breeding site as your pure juveniles) that's OK too. All this gets into the realms where numerical design calculations are needed (which is not a 5-minute job— ie requires a contract!).

The reassurance then is that, with 2 adult sample grounds, *eventually* the data will reveal any substantial poor-mixing, and should allow some degree of correction (i.e. unbiased estimates). How long is "eventually"? Well, there's 3 things there.

1. "Eventuality" depends partly on how different the mixing actually is between those adult grounds (α vs β); see previous paragraph. Drastic effects that would make a big difference, will become apparent sooner than subtle effects that probably don't matter that much in the "grand" scheme of management.

2. The equations above completely bury the time/age element— "N is just N" but it should really be N_t, where t is the birth-date of some potential-offspring sample. In practice you

[24] Lower bound always (in theory); upper bound depending on the nature of under/over-representation

[25] This could be "quick" because the two adult grounds are being compared to the same young juveniles— whereas within-adult-ground comparisons tend to take longer, for reasons explained elsewhere.

[26] I probably should explain that more clearly, but it's really a point specific to one possible E-ABFT design which I'll pick up elsewhere

should *never*[27] fit an equilibrium CKMR model to anything, least of all fish! So when comparing two rates that ought to be "consistent", one problem is that N might have changed substantially between them, if they are being measured at different times. And if you are considering Adult-Adult POPs, the t of many of the "pseudojuvenile" candidate-offspring will, at the start of the study, be years earlier than the t for the true juveniles sampled from site 1 that are being used in Adult-TrueJuve POP comparisons. When it's all embedded in a proper pop dyn model, matters can be arranged so that the intrinsic-change-in-N *ought* to be accounted for, and appropriately-nuanced comparison statistics can be devised. But that doesn't change the fundamental issue that there might not be enough information to allow a precise direct comparison. Eventually this issue goes away, because the range of t's overlap between the various datasets. But not in the first couple of years.

3. "Eventuality" also depends on sample size. If you think you could get enough samples (assuming mixing really is good) to get a nice CV using just one adult ground, it's probably not going to work to merely split that total sample size across two adult grounds and still expect (a) the same nice CV *and* (b) enough kin-pairs to check for an appreciable difference; you'll get less kin from each site even if the same overall, and maybe not enough to check. In other words, the reassurance of checkability comes at some cost in sample size (though not as much as doubling it; it's not as bad as having two completely separate programs). But-but: you can always increase the sample size just by continuing another year or two with whatever sampling levels you've been taking up to that point; so you will have some options to later dig yourself out of an assumption hole, if you become worried about it later— *provided* that you remembered to collect the necessary samples in the past ;)

Although it might be *mathematically* possible to disentangle more complex "hidden" scenarios just by sampling (thoroughly) more badly-mixed adult grounds (i.e. giving up on the at-least-one-pretty-well-mixed idea), I personally would be very nervous about relying on that approach from the beginning unless I did have some a priori expectation of being adequately-close-to-well-mixed in at least one of the sites— because the identifiability stuff all feels rather sensitive to idealized model assumptions. At the minimum, some elaborate and defensive design work would be required to prevent subsequent disappointment from over-inflated expectations. If I really could not think of any adult sampling ground where the "probably-fairly-well-mixed" assumption might be tolerable in the medium term, then I might be pretty reluctant to start...

5.1 What's the problem with sampling lots of little places?

You don't get to "one well-mixed" by averaging together "many badly-mixed"; you just get to something that is almost certainly biased to an unknowable, and uncheckable, degree. Unless you have *a lot* of (adult) samples from one place, you just won't find enough kin-pairs there to check

[27] OK, almost never...

whether the empirical rate from that place matches that from other places, or if it does differ then by how much. Each time you sample a new place, you add new mixing parameters to estimate for that place. You don't have enough data to model them all properly, so you (or whichever modeller has been foolish enough to take on such a doomed task, which certainly would not be me) just end up having to lump-and-hope, and then the "weighting" of each imperfectly-mixed place towards the final ungodly estimate will reflect the arbitrary fact of its sample size, rather than anything biologically meaningful. This is *not* a situation where the Law of Large Numbers applies; the only Law that is gonna apply, is Murphy's.

I could go on to hammer this further into the ground, but hopefully the point is clear: *don't* launch into a CKMR study on something like E-ABFT[28] in the futile hope of stitching together a meaningful dataset from lots of little adult samplettes, even if you can also get one nice big pure juvenile sample.

5.2 What if the juvenile samples aren't pure?

Oh dear— the meter on my crystal ball has just expired!

Presumably, things get fairly complicated, and may or may not be estimable; see the 2017 report (which I have not re-read for several years, so I can't entirely remember) for more.

6 HSPs

Cross-cohort HSPs are IMO an essential part of putting together a reasonable CKMR model for most animals (certainly teleost fish). They let you: estimate Z (*independently of your N-estimate*) and often M; estimate average adult age (i.e. distinguish between lots-of-young and fewer-old adult populations of same TRO); bypass the need for other data/assumptions e.g. re selectivity. And they are FREE because you have already collected the juves for use in POPs!

However, for teleosts HSPs do not directly carry an absolute abundance signal, because of the quadratic effect of size-fecundity relationships (it's a long story...); they are an "essential piece of untangling equipment", but the "rope" itself is POPs. So at least for now I'm considering only what the POPs can tell us abundance— even though in practice the HSPs would be needed to extract that information, and might provide extra mixing-related info themselves.

But let's have some formulae anyway. Consider the main source: HSPs[29] among juves from site 1.

[28] I think there are species where it is perfectly sensible to just spread the sampling spatially in a fairly undesigned way; you can detect gross lack of mixing, and if you don't see it, you can proceed, and if you do, you have (a) learnt something pretty important and (b) may be able to build a proper spatial CKMR model a la BSA 2016 3.10. But those aren't situations where juveniles are coming from one pure site.

[29] Only Maternal HSPs for now, as distinguished by mtDNA. Similar stuff applies to PHSPs.

This formula is slightly expanded to illustrate general points about HSPs beyond just "mammals":

$$\begin{gathered}\mathbb{P}\left[K_{\text{Jill,Jane}} = \text{M(aternal)HSP}|\text{Jill born } T > 0 \text{ years before Jane, both in site 1}\right] = \\ \mathbb{P}\left[\text{Jill's mum (Xena) survived til Jane's birth}\right] \times \\ \{\text{adjustment for Xena's growth if she did live}\} \times \\ \mathbb{P}\left[\text{Jane's Mum was Xena}\right] = \\ e^{-zT} \times 1 \times \left(\frac{1}{N_1 @ \text{Jane's birth}}\right) \\ = \frac{e^{-zT}}{N_1} \end{gathered} \tag{6.1}$$

under the special assumptions here, e.g. no growth adjustment because these are "mammals"... see BSA 2016 3.9 for more general life histories (for which you have to sum over possible values for Xena's age), and for why $T = 0$ doesn't fit this pattern. It's important *not* to leap to the notion that you can use eqn (6.1) *on its own* and for *fish* to estimate abundance; there's an additional quadratic-ish factor to do with adult age-distribution and fecundity, which doesn't apply to "mammals"[30] and which affects the relationship between "abundance" and number of HSPs. And I mean bigly affects. For fish, you want both HSPs *and* POPs (did I already say that..? hopefully yes I did). Nevertheless, (6.1) is an emergency check against the possibility that all your adults are actually site-2ers!

If we have *good* ages from the adult A-samples, then we may also be able to do A1 HSPs and even AA HSPs. The key point is that the two animals have to have been born in the same site if they are to be HSPs, because the first one's Mum keeps breeding in that same site. NB that the ability to use adults in HSPs may be restricted because of (i) less accurate ageing (if T is uncertain then the information content is diminished) and (ii) the need to avoid too much risk of Grandma-Grandchild pairs: the *first-born* of the pair needs to be youngish when (lethally) sampled.

For A1 HSPs— assuming Ann is born before Jill— we have (left as an exercise):

$$\begin{gathered}\mathbb{P}\left[K_{\text{Ann,Jill}}|A, 1, T > 0\right] = \\ \left(\frac{N_1}{N_1 + \alpha N_2}\right)^2 \frac{e^{-z_1 T}}{N_1} \end{gathered} \tag{6.2}$$

where the adult mortality rate is now z_1 because it's only for site-1ers. Depending on the setting, it may or may not make sense to assume $z_1 = z$ overall. Equal m by breeding-site might make good sense, but it may get tricky thinking about how to apportion catches... assuming your catch data bear any resemblance to truth in the first place.

For AA HSPs, the formula is something like

[30] And turns out to be actually *helpful* for fish, *provided* you've also got POPs.

$$\begin{aligned}
&\mathbb{P}\left[K_{\text{Ann,Ali}}|A, A, T > 0\right] = \\
&\left(\frac{N_1}{N_1 + \alpha N_2}\right)^2 \frac{e^{-z_1 T}}{N_1} + \left(\frac{\alpha N_2}{N_1 + \alpha N_2}\right)^2 \frac{e^{-z_2 T}}{N_2} \\
&= \frac{N_1 e^{-z_1 T} + \alpha^2 N_2 e^{-z_2 T}}{\left(N_1 + \alpha N_2\right)^2}
\end{aligned} \tag{6.3}$$

If you feel able to make the approximation that $z_1 \approx z_2$, then this simplifies slightly to

$$\mathbb{P}\left[K_{\text{Ann,Ali}}|A, A, T, z_1 = z_2 = z\right] = \frac{e^{-zT}\left(N_1 + \alpha^2 N_2\right)}{\left(N_1 + \alpha N_2\right)^2} \tag{6.4}$$

Clearly, there's lots of stuff that might help with diagnosis here in the HSPs, and if you are ambitious then with estimating various mixy things. But... oh dear, *the meter has expired* on my crystal ball ;)

References

Bravington, Mark V., Hans J. Skaug, and Eric C. Anderson (May 2016). "Close-Kin Mark-Recapture". In: *Statistical Science* 31.2, pp. 259–274. DOI: 10.1214/16-STS552. URL: https://doi.org/10.1214/16-STS552.

Conn, Paul B. et al. (2020). "Robustness of close-kin mark–recapture estimators to dispersal limitation and spatially varying sampling probabilities". In: *Ecology and Evolution* 10.12, pp. 5558–5569. DOI: https://doi.org/10.1002/ece3.6296.

Mini-appendix 1: Direction of bias of naive single-adult-sampling-ground estimates

To check the bias direction of AA naive estimate compared to the true total abundance of $1 + \rho$, I write the two formula with an "either smaller or bigger" sign $\lessgtr$, and then step thru until it

hopefully becomes obvious which sign must apply. Like this:

$$
\begin{aligned}
1+\rho &\lessgtr \frac{(1+\alpha\rho)^2}{1+\alpha^2\rho} \\
\Leftrightarrow \left(1+\alpha^2\rho\right)(1+\rho) &\lessgtr (1+\alpha\rho)^2 \\
\Leftrightarrow 1+\alpha^2\rho^2+\left(1+\alpha^2\right)\rho &\lessgtr 1+2\alpha\rho+\alpha^2\rho^2 \\
\Leftrightarrow \left(1+\alpha^2\right)\rho &\lessgtr 2\alpha\rho \\
\Leftrightarrow 1+\alpha^2 &\lessgtr 2\alpha \\
\Leftrightarrow (1-\alpha)^2 &\lessgtr 0
\end{aligned} \tag{6.5}
$$

So it's always the lower option, regardless of whether $\alpha \lessgtr 1$, and by backtracking thru the equations we can see that $1+\rho > 1/c_A$ and thus that the AA-naive estimate is negatively biased.What about the arithmetic average of naive-AA and naive-A1? (A "Butterworthy" "solution" ;) Then we have

$$
\begin{aligned}
1+\alpha\rho+\frac{(1+\alpha\rho)^2}{1+\alpha^2\rho} &\lessgtr 2(1+\rho) \\
\Leftrightarrow (1+\alpha\rho)\left(1+\alpha^2\rho\right)+(1+\alpha\rho)^2 &\lessgtr 2(1+\rho)\left(1+\alpha^2\rho\right) \\
\Leftrightarrow 1+\alpha\rho+\alpha^2\rho+\alpha^3\rho^2+1+2\alpha\rho+\alpha^2\rho^2 &\lessgtr 2+2\rho+2\alpha^2\rho+2\alpha^2\rho^2 \\
\Leftrightarrow 3\alpha\rho+\alpha^3\rho^2 &\lessgtr 2\rho+\alpha^2\rho+\alpha^2\rho^2 \\
\Leftrightarrow \left(\alpha^3-\alpha^2\right)\rho &\lessgtr 2-3\alpha+\alpha^2 \\
\Leftrightarrow \alpha^2(\alpha-1)\rho &\lessgtr (\alpha-2)(\alpha-1)
\end{aligned} \tag{6.6}
$$

The average would only be worth considering if we were sure that AA<A1 so that $\alpha > 1$ (otherwise AA is less biased). If $\alpha < 1$, we can divide the last line by $\alpha - 1$ without changing the "sign", so that all-in-all the condition for the average to be negatively biased would be this:

$$
\alpha^2\rho < \alpha - 2 \tag{6.7}
$$

Now, that can only be true if ρ is very small (0.13 or less; see below) and even then only when α is very large (it is impossible for any ρ unless $a > 2$). So, outside of extreme situations (which you are supposed to know enough to avoid!), there will be some positive bias in the naive average when $\alpha > 1$, but clearly less than from using naive A1 alone. Hence, if AA>A1, then naive AA (which is negatively biased) and the naive average (AA+A1)/2 (which is positively biased) form "stochastic bounds" on total abundance. But, if you have any option, then you'd do better to sample additional adult grounds and just deal with things properly...The bounds on ρ required to

get negative bias as per eqn (6.7) are easily found with calculus:

$$\rho_{\max}(\alpha) \triangleq \frac{\alpha-2}{\alpha^2} = \alpha^{-1} - 2\alpha^{-2}$$
$$\implies \frac{d\rho_{\max}}{d\alpha} = -\alpha^{-2} + 4\alpha^{-3}$$
$$\frac{d\rho_{\max}}{d\alpha} = 0 \implies \alpha = 4$$
$$\rho_{\max}(4) = \frac{4-2}{16} = \frac{1}{8} = 0.125$$

All the algebra here is easy to check with a bit of R code.

Mini-appendix 2: Surplus algebra

This is a gruesome piece of algebra that no-one except me should probably ever read, to derive closed-form estimates for the 2-adult-ground 1-juve-site case: specifically, to estimate 4 parameters $(N_1, N_2, \alpha, \beta)$ based on 4 empirical rates (POPs in the categories A1, B1, AA, BB). I did this *before* realizing that there is actually a 5th rate (AB POPs) which really makes this irrelevant! Anyway, it looks to me as if "it works" but I haven't put in numbers to check. I'm a bit loath to discard perfectly good algebra even if it's irrelevant, and I don't know where else to keep it. You don't have to read it!

The scheme here is that the various c's are things that can be computed in succession just from the 4 observed POP rates (including the rates themselves). The symbol "$\triangleq$" means "is defined as". And when you see e.g. ... := c_6, it means you should now calculate c_6 using the other side of the formula. Actual parameter estimates (to be calculated from c's and/or other parameter estimate from previous steps) get a hat, eg $\hat{\alpha}$. For shorthand, I'll write $\mathbb{P}[\text{B1}]$ instead of the informal $\mathbb{P}[\text{Bertha from B is the mother of juvenile Jill from 1}]$. More formally:

$$\mathbb{P}[\text{B1}] \triangleq \mathbb{P}\left[K_{\text{Bertha,Jill}} = \text{POP} | \text{Bertha sampled at B, Jill at 1}\right]$$

and so on.

It starts by equating the 4 observed data to their expected values, and then moves towards solving for the 4 parameters. As noted somewhere, there's actually a 5th datum (AB POPs) so you would never ever do it like this— but the point of the exercise (apart from "fun") is to check if the parameters can be estimated even in principle.

$$c_1 \triangleq \mathbb{P}\left[\text{A1}\right] = \frac{1}{N_1 + \alpha N_2} \tag{6.8}$$

$$c_2 \triangleq \mathbb{P}\left[\text{B1}\right] = \frac{1}{N_1 + \beta N_2} \tag{6.9}$$

$$\implies (\alpha - \beta) N_2 = \frac{1}{c_1} - \frac{1}{c_2} := c_6 \tag{6.10}$$

$$c_3 \triangleq \mathbb{P}\left[\text{AA}\right] = \frac{N_1 + \alpha^2 N_2}{\left(N_1 + \alpha N_2\right)^2}$$

$$\implies c_3 = \left(N_1 + \alpha^2 N_2\right) c_1^2 \tag{6.11}$$

$$c_4 \triangleq \mathbb{P}\left[\text{BB}\right] = \frac{N_1 + \beta^2 N_2}{\left(N_1 + \beta N_2\right)^2}$$

$$\implies c_4 = \left(N_1 + \beta^2 N_2\right) c_2^2 \tag{6.12}$$

$$\& \, [6.11] \implies \left(\alpha^2 - \beta^2\right) N_2 = \frac{c_3}{c_1^2} - \frac{c_4}{c_2^2} := c_5$$

$$\implies (\alpha + \beta)(\alpha - \beta) N_2 = c_5 \tag{6.13}$$

$$\& \, [6.10] \implies \alpha + \beta = \frac{c_5}{c_6} := c_7 \tag{6.14}$$

$$[6.11] \, \& \, [6.8] \implies \left(\alpha - \alpha^2\right) N_2 = \frac{c_1 - c_3}{c_1^2}$$

$$[6.12] \, \& \, [6.9] \implies \left(\beta - \beta^2\right) N_2 = \frac{c_2 - c_4}{c_2^2}$$

$$\frac{\alpha - \alpha^2}{\beta - \beta^2} = \frac{c_2^2}{c_1^2} \frac{(c_1 - c_3)}{(c_2 - c_4)} := c_8 \tag{6.15}$$

$$\& \, [6.14] \implies \frac{\alpha - \alpha^2}{(c_7 - \alpha) - (c_7 - \alpha)^2} = c_8 \tag{6.16}$$

$$\implies \text{quadratic formula for } \hat{\alpha} \tag{6.17}$$

$$\& \, [6.14] \implies \hat{\beta} := c_7 - \hat{\alpha} \tag{6.18}$$

$$\& \, [6.8] \, \& \, [6.9] \implies \text{two linear simultaneous eqns for } \hat{N}_1 \text{and } \hat{N}_2 \tag{6.19}$$

Whew.