**Working title**

Can we use metabarcoding data for food web studies of invertebrate communities or

What is learned from these types of data that complements/revises/boosts other methods?

**Introduction**

Challenge: Invertebrate communities have many cryptic interactions, making it challenging to build interaction data based on real observations. Metabarcoding diet could be a solution in these systems to otherwise inferred or unknown data.

There is a dearth of terrestrial food webs in the literature, due to many historic limitations, one including lack of an ability to capture interactions for many consumers who are invertebrates. This also has the potential to re-evaluate and disaggregate data for aquatic food webs with invertebrate consumers.

Werewolf: If we use metabarcoding data to build food webs and interaction networks, we want to know that it is giving us information comparable to or better than inferred data.

Silver Bullet: We look at metabarcoding data of generalist predator species at the individual level and show some examples of comparable or better than inferred data.

**Takeaways**

**Molecular diet (hopefully) performs as well** as others in building food webs? What does it add, how does it compare, and why?

1. Analyses: Link comparison (might also consider taking species-level data here for the ASVs that were assigned to species…), trophic position of links (e.g. degree of omnivory?)
   1. We captured more links as a proportion of the total possible number of families where links were possible than a web with comparable family richness, though fewer of the links for a web with fewer families. When compared to just the total number of links per predator species, on Palmyra we observe a similar number of links per species for a web with double the number of species and an similar number of families, and more links per species for a web with the same number of nodes but fewer families.

**Molecular diet adds the ability to examine individuals**, which allows you to explore ideas related to interaction strength (e.g. frequency), individual body size ratios, and ontogeny (look at Ings individual-level conversation here).

1. Analyses: interaction strength (frequency), individual body size ratios compared to published species-level (maybe here go back to original data and subset data that came back at species level to explore this more), ontogeny or lack thereof, body size ratio and interaction frequency (Emmerson and Raffaelli 2004)

**Molecular diet allows for statistical validation of sampling and methods planning**. Ability to quantify data resolution and build methodologies off of it (have we captured full diet ala Polis and Ings conversations? Are we missing important ontogenetic stages, and how much do we need to collect to get at this?)

1. Analysis: link accumulation and diet richness estimates in addition to how much do you need to sample to capture ontogenetic shifts?

**Analyses:**

1. Does molecular diet give you more diet resolution (links) than other approaches?
   1. Per species links comparison between published webs and molecular
      1. Compare to Hines et al. 2019 (rmangal), Laigle et al. 2017 (Dryad)
   2. The ability not only to capture links but build a confidence interval of capturing full links based on individual accumulations of new links (accumulation curves).
      1. Richness estimates for our data
   3. No published terrestrial food webs with interaction strengths – added value of being able to add this layer of data to these food webs (maybe think about a modelling approach here to highlight importance, or just report interaction frequency distributions?
      1. Our predators with and without interaction strengths, is there an easy way to model this?
      2. Laigle et al. has a bunch of different food webs, could consider constructing a frequency-based model here, but highlighting that this is across-communities vs. within-communities, so consider difference of scale here…
2. How do individual-level predator-prey body size ratios compare to species-level, and are there differences between individual-level and species-level body size ratios for published versus molecular?
   1. Species-level predator-prey body size ratios comparison between published and molecular
      1. Laigle et al. 2017
   2. Relationship between predator-prey body size ratios and interaction strength – is this something to tease out here? Ala Emmerson and Raffaeli 2004.
3. Do molecular diet allow you to determine ontogenetic diet shifts within species?
   1. No ontogeny in published food webs for continuous-growth predators (but check aquatic)
   2. Lack of evidence for ontogeny in our dataset, but is this a limitation of data amount? And how would we give a prescriptive of how much we need to add?
      1. (maybe iNEXT accumulations based on arbitrary size classes of predators and/or core prey within the whole species comparisions, rather than all prey comparisons). Give a prescriptive of how much you would have to collect to get at ontogeny? Or?

**Methods**

Sample collection methods:

How many predators of total predators in Palmyra food web?

Sample extraction, PCR, and sequencing methods:

Sample cleaning, denoising, taxonomic assignment, and rarefying methods:

Literature search:

Looking for datasets of terrestrial invertebrate interaction webs with comparable numbers of species to those present in the Palmrya food web (~400). We used various sources that curate online food webs, including Mangal (through rmangal in R version), Dryad data, NCEAS InteractionWeb Database, and the Global Web Database. For rmangal, only one dataset of 172 (Hines et al. 2019, 714 nodes, 51,496 interactions, accessed June 24, 2020) fit this specification. For Dryad datasets, we searched using multiple search terms (all accessed between June 24-July 1, 2020: “food web” (463 papers), “interaction network” (553 papers), “predator prey interactions” (520 papers), “diet analysis insect” (42 datasets), “diet analysis spider” (16 datasets), “gut content analysis” (41 datasets), “diet analysis invertebrate” (24 datasets)). Of these datasets, 13 reported invertebrate predation interactions, 5 represented full interaction networks, and 1 of these represented an interaction networks with greater or comparable species diversity to the Palmyra community (Laigle et al. 2017: 878 nodes, 65,536 documented interactions across multiple food webs). There were no terrestrial food webs in the NCEAS InteractionWeb Database on our access date (June 2020), and no terrestrial food webs with comparable species counts (none with more than 100 taxa) in the Global Web Database.

Outputs:

Number of predators in each food web? Proportion of consumers that are predators?

Proportion for Palmyra food web as well as the proportion we have represented in our datasets here.