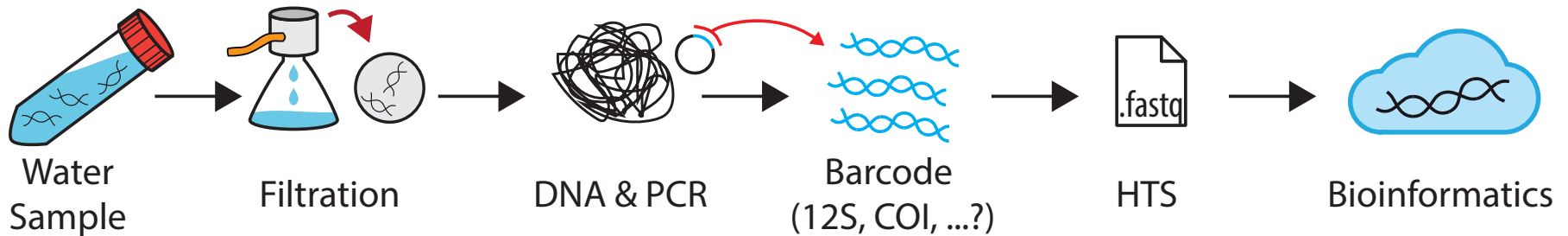


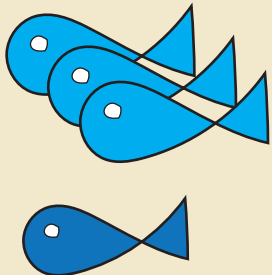
# eDNA metabarcoding: Sounds too good to be true?

“just” collect a water sample - Capture **all** the biodiversity!?



## Traditional surveys

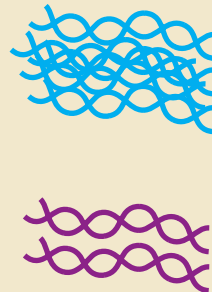
- Lot's of work
- Count data (Abundance)



3 x Fish **A**  
1 x Fish **B**

## eDNA surveys

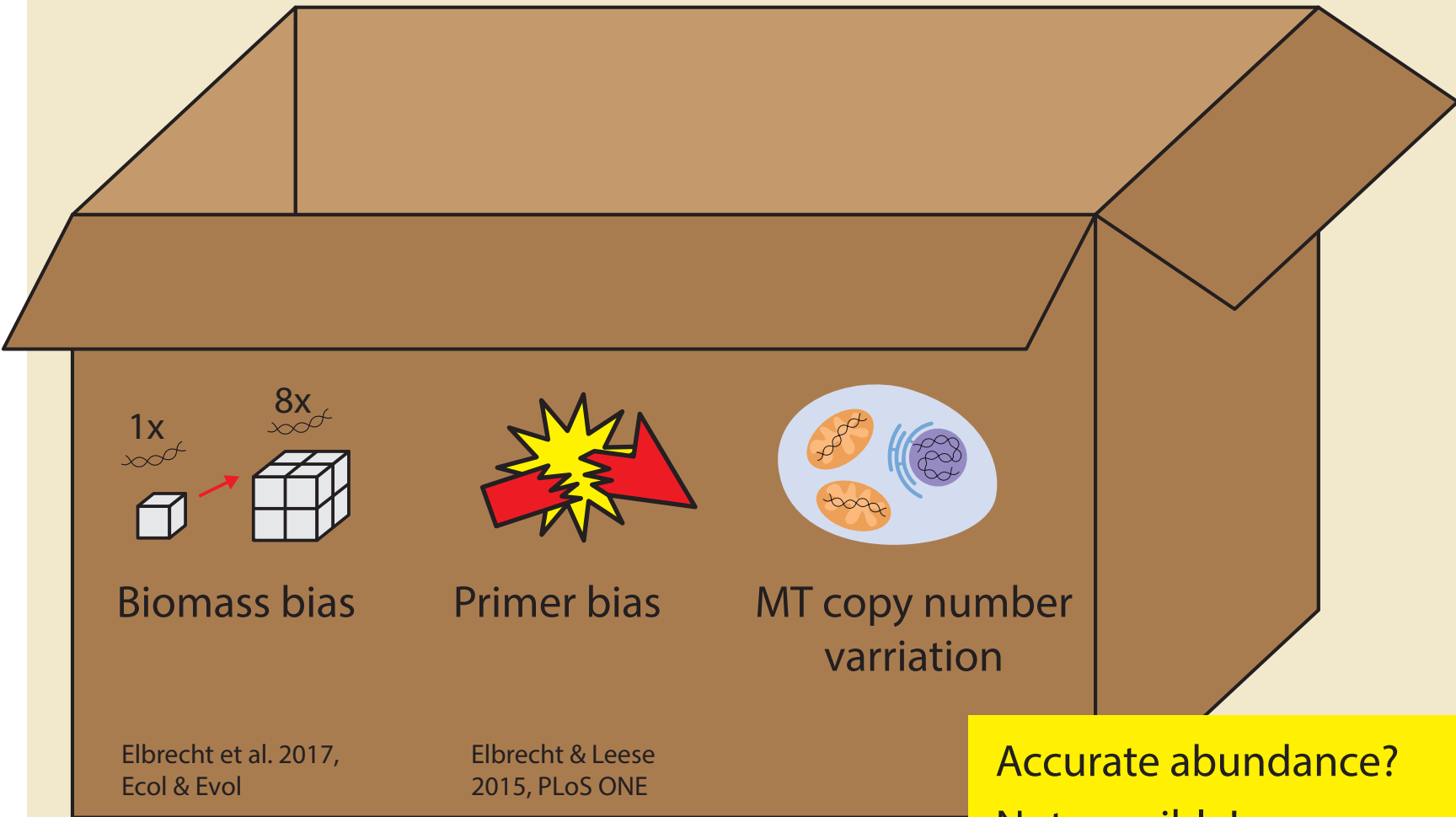
- What count data do we get?
- Biomass, relative abundance, p/a!?



1000 reads Fish **A**  
0 reads Fish **B**  
300 reads Fish **C**



# Tissue based metabarcoding: Well understood biases



Accurate abundance?  
Not possible!

# eDNA metabarcoding

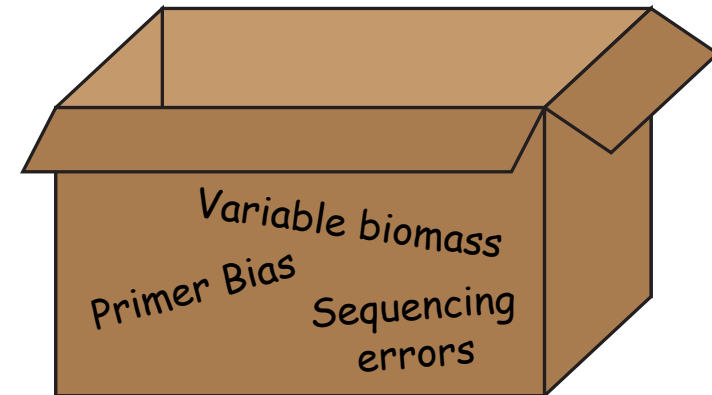
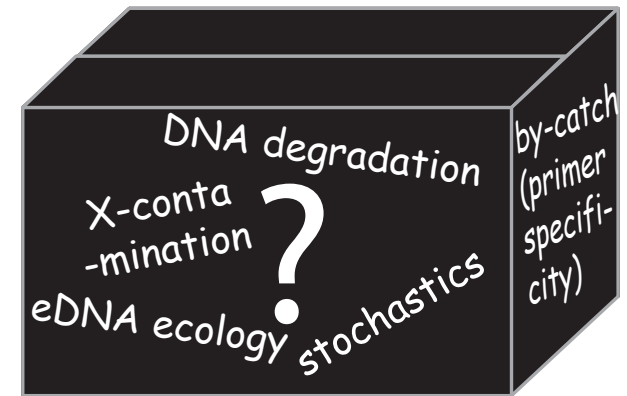
## Adding (unknown) biases on top!

- eDNA metabarcoding:
  - Metabarcoding biases (previous slides)
  - Many eDNA biases and unknowns!
- Reviews:
  - **Ecology of eDNA:** Barnes & Turner 2015, Conserv Genet
  - **eDNA abundance / good introduction:** Deiner et al. 2017, Mol Ecol
  - **eDNA uncertainty:** Cristescu & Hebert, 2018 Annual Reviews
- Less a problem for targeted qPCR / ddPCR

**eDNA Metabarcoding = amazing tool!**

- But let's use it cautiously
- Maybe a bit over-hyped ;)

### eDNA biases (black box)



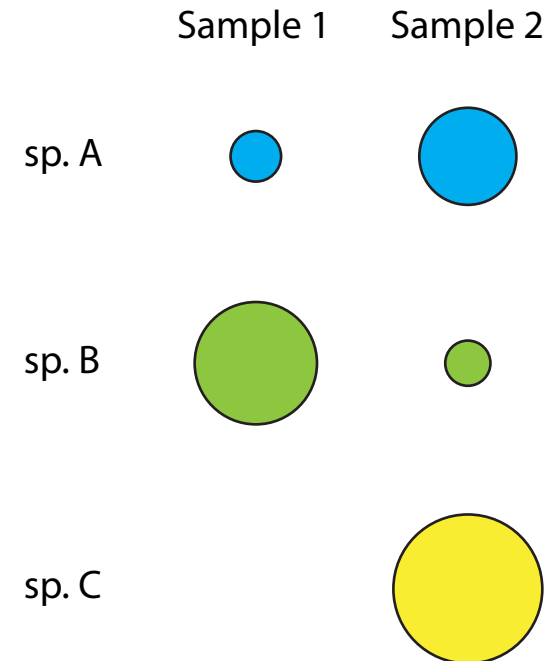
### Metabarcoding biases

*IMO: What can we (reliably) say with eDNA metabarcoding?*

- Exact number of fish? **NO**
  - Biomass of each fish? **NO**
    - With species specific markers: **Maybe**
- Levi et al. 2018, bioRxiv
- Presence / absence of fish species? **YES!**
  - Relative abundance? **YES!**

eDNA has it's biases!  
Will **not** capture everything

How to use relative abundance?



- 1.) Sample 1 contains more sp. B!
- 2.) But due to primer bias, species abundance is unknown!

# Example: Primer bias

- “Solving” primer bias;

- We need multiple markers!

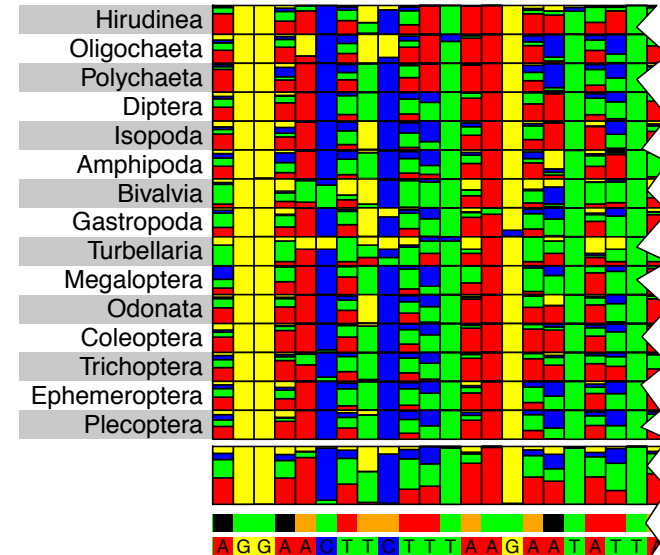
Alberdi et al. 2017, MEE, Zhang et al. 2018, Ecol Appl

- Ribosomal markers are great!

Deagle et al. 2014, Biol Letters

- My COI primer (without degeneracy) is great

Jusino et al. 2018, Mol Ecol Resour



Elbrecht & Leese 2016, MEE

Maybe true in *some* cases!

But how about extensive primer validation!?

Braukmann et al. 2018, bioRxiv

- Tissue bulk sample containing 374 taxa (BINs)
- Metabarcoding with 21 different primer sets!

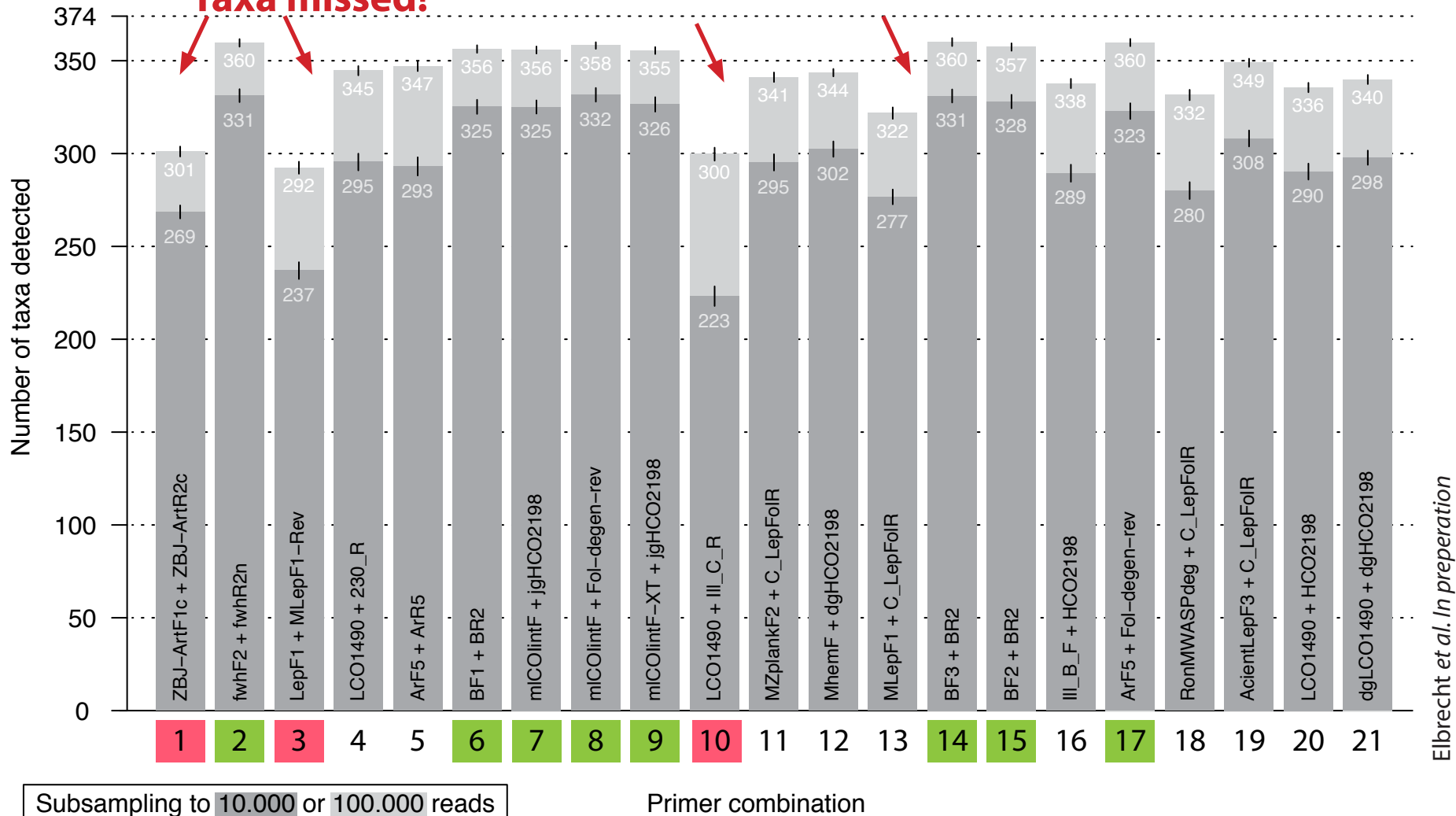


# Primer bias!

## 21 Primer sets evaluated on 374 bulk insect taxa



**Taxa missed!**



Elbrecht et al. In preperation

- Metabarcoding or single species detection?
- Primer validations are key!
- Need to understand biases in:
  - eDNA "ecology"
  - Lab-methods & bioinformatics
- eDNA based methods already useful, embrace them!

"As this study clearly shows, there are excellent [#metabarcoding](#) primers out there, we just have to use them."  
[@VascoElbrecht](#) via Twitter

