Dietary analysis (DNA)

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NGS

I will use NGS for the analysis of the dietary relationship between the functional groups

Procedure

- DNA-extraccion
  - Feacal extraction kit
    - PowerFecal DNA Isolation Kit code : MB-12830-50 Price : $439.00 ex. GST. kit of 50
- Concentration determination
  - Fluorecense at UTAS
- Amplicon PCR
  - Tailed primers + 10% untailed primers
    - Primers
      - Furseal
      - Lobster
      - Fish
  - Taq polymerase
  - ddH2O
- Clean Up
- Gel To check if PCR works

- Index PCR
  - Nextera index kit
  - Kapa mix
  - ddH2O
- Checking PCR
- Quantify amount of DNA
  - Quantiflour kit
- Normalize the samples
- Pool all the samples
- Sequence in illumina MiSeq
  - Sequencing reagents (1500 for ??)

Cost DNA procedure

-All the cost are in AUD

Step-materials	Total Amount	Price unit	Total Price (182)	Total (96)
Consumables	Lot	300	300	300
DNA-Extraccion	96	439(kit 50)	1756	878
Cons. Determination	96	2 per sample	364	192
Amplicon-PCR	96	40 (96-well plate)	80	40
Gel	96			
Cleanup	96	8 (10 samples)	160	80
Index-PCR	96	800 (96-well plate)	1600	800
Gel				
Clean-up	96	8 (10 samples)	160	80
Gel				
Sequence Ilumina	96	1500 (regents)	1500	1500
		Total	5620	3570

Functional groups

Functional Groups	Current Number	Needed	Total
Spiny$_{l}$*obster*		18	-161
Golden$_{C}$*rab*	1	17	
Breca	8	10	
Vidriola	2	16	
Anguila		18	
Otariid	2	16	
Octupus	3	15	
Large pelagic fish	2	16	
Small pelagic fish		18	
Small benthic fish	1	17	
Large benthic fish		18	
Per groups	18		

Clasification and samples

Functional Group	Species	N-Samples	adviced	Needed
Lobster	Lobster	0		
Golden Crab	Golden Crab	1		
Breca	Breca	8		
Octopus	Octopus	4		
Small Crustacean	Jaiva			
	King crab			
Anguila	Anguila			
Large Benthic Fish	Bacalao			
	Tollo			
	Lenguado			
	Congrio			
Small Benthic Fish	Colorado			
	Graniento			
	Cabrilla			
	Chancharro	1		
Large Pelagic Fish	Corvina	1		
	Jurel	1		
	Jurelillo	1		
Small pelagic Fish	Pampanito			
	Pez Mariposa	1		
	Jerguilla			
Pinnipedo	Furseal	2		
Vidriola	Yellowtail amberjack	8		
Total		28		