

Extraction plan for APCL18_377-APCL18_470

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This is a script for adding samples that are not in the laboratory database and have not been extracted. This is set up to handle one plate at a time.

Obtain a list of all clownfish sample ids from the Leyte database

```
# connect to leyte fieldwork db
leyte <- read_db("Leyte")
# import fish table
# select down to only sample_id numbers and remove any rows without a sample
fish <- leyte %>% tbl("clownfish") %>%
  # select only the column sample_id
  select(sample_id) %>%
  # remove any non-sample observations
  filter(!is.na(sample_id)) %>%
  distinct(sample_id) %>%
  # remove any repeat sample_ids (this should not be needed)
  collect()

# make sure each sample_id is only represented once
fish <- distinct(fish)
```

Select the range of samples in the plate

```
work <- fish %>%
  filter(sample_id >= params$first, sample_id <= params$last)

# define wells
plate <- data.frame(row = rep(LETTERS[1:8], 12), col = unlist(lapply(1:12, rep, 8))) %>%
  mutate(sample_id = ifelse(row == "D" & col == 2, "XXXX", NA),
         sample_id = ifelse(row == "E" & col == 8, "XXXX", sample_id))

samples <- plate %>%
  filter(is.na(sample_id)) %>%
  select(-sample_id)

plate <- anti_join(plate, samples, by = c("row", "col"))

samples <- cbind(samples, work)

plate <- rbind(plate, samples) %>%
  arrange(col, row)
```

Make a plate map of sample IDs (for knowing where to place fin clips)

```
platemap <- as.matrix(reshape2::acast(plate, plate[,1] ~ plate[,2]), value.var = plate[,3])
knitr::kable(platemap, booktabs = T) %>%
  # use scale_down to get map to fit within the bounds of the pdf
  kable_styling(latex_options = "scale_down")
```

	1	2	3	4	5	6	7	8	9	10	11	12
A	APCL18_377	APCL18_385	APCL18_392	APCL18_400	APCL18_408	APCL18_416	APCL18_424	APCL18_432	APCL18_439	APCL18_447	APCL18_455	APCL18_463
B	APCL18_378	APCL18_386	APCL18_393	APCL18_401	APCL18_409	APCL18_417	APCL18_425	APCL18_433	APCL18_440	APCL18_448	APCL18_456	APCL18_464
C	APCL18_379	APCL18_387	APCL18_394	APCL18_402	APCL18_410	APCL18_418	APCL18_426	APCL18_434	APCL18_441	APCL18_449	APCL18_457	APCL18_465
D	APCL18_380	XXXX	APCL18_395	APCL18_403	APCL18_411	APCL18_419	APCL18_427	APCL18_435	APCL18_442	APCL18_450	APCL18_458	APCL18_466
E	APCL18_381	APCL18_388	APCL18_396	APCL18_404	APCL18_412	APCL18_420	APCL18_428	XXXX	APCL18_443	APCL18_451	APCL18_459	APCL18_467
F	APCL18_382	APCL18_389	APCL18_397	APCL18_405	APCL18_413	APCL18_421	APCL18_429	APCL18_436	APCL18_444	APCL18_452	APCL18_460	APCL18_468
G	APCL18_383	APCL18_390	APCL18_398	APCL18_406	APCL18_414	APCL18_422	APCL18_430	APCL18_437	APCL18_445	APCL18_453	APCL18_461	APCL18_469
H	APCL18_384	APCL18_391	APCL18_399	APCL18_407	APCL18_415	APCL18_423	APCL18_431	APCL18_438	APCL18_446	APCL18_454	APCL18_462	APCL18_470

ONLY DO THIS ONCE

Generate extract numbers for database

```
lab <- read_db("Laboratory")
extracted <- lab %>% tbl("extraction") %>%
  summarise(last = max(extraction_id, na.rm = T)) %>%
  collect() %>%
  mutate(last = substr(last, 2,5))

plate <- plate %>%
  mutate(well = 1:nrow(plate)) %>%
  mutate(extraction_id = paste("E", well + as.numeric(extracted$last), sep = "")) %>%
  mutate(well = paste(row, col, sep = "")) %>%
  mutate(method = "DNeasy96",
         final_vol = "200")

plate_name <- plate %>%
  summarise(first = min(extraction_id),
           last = max(extraction_id, na.rm = T))
```

Make a platemap with extraction ids

```
map <- plate %>%
  select(row, col, extraction_id)
platemap <- as.matrix(reshape2::acast(map, map[,1] ~ map[,2]), value.var = map[,3])

## Using extraction_id as value column: use value.var to override.

knitr::kable(platemap, booktabs = T) %>%
  kable_styling()
```

	1	2	3	4	5	6	7	8	9	10	11	12
A	E4935	E4943	E4951	E4959	E4967	E4975	E4983	E4991	E4999	E5007	E5015	E5023
B	E4936	E4944	E4952	E4960	E4968	E4976	E4984	E4992	E5000	E5008	E5016	E5024
C	E4937	E4945	E4953	E4961	E4969	E4977	E4985	E4993	E5001	E5009	E5017	E5025
D	E4938	E4946	E4954	E4962	E4970	E4978	E4986	E4994	E5002	E5010	E5018	E5026
E	E4939	E4947	E4955	E4963	E4971	E4979	E4987	E4995	E5003	E5011	E5019	E5027
F	E4940	E4948	E4956	E4964	E4972	E4980	E4988	E4996	E5004	E5012	E5020	E5028
G	E4941	E4949	E4957	E4965	E4973	E4981	E4989	E4997	E5005	E5013	E5021	E5029
H	E4942	E4950	E4958	E4966	E4974	E4982	E4990	E4998	E5006	E5014	E5022	E5030

```
plate <- plate %>%
  mutate(plate = paste(plate_name$first, plate_name$last, sep = "-")) %>%
  select(-row, -col)
```

Import the extract list into the database

Make sure you have created your output PDF for this labwork before sending to the database

```
rm(lab)
lab <- write_db("Laboratory")

## Loading required package: DBI
dbWriteTable(lab, "extraction", plate, row.names = F, overwrite = F, append = T)

## [1] TRUE
dbDisconnect(lab)

## [1] TRUE
rm(lab)
```

Load fin clips

Calculate the amount of lysis buffer to make

```
num_samples <- params$num_samples
w_error <- num_samples * 1.1

mix <- readr::read_csv("num_samples_w_error, ul_prot_k, ml_ATL_buff
0,0,0") %>%
mutate(num_samples_w_error = w_error,
ul_prot_k = w_error * 20 * 0.001,
ml_ATL_buff = w_error * 180 * 0.001)

kable(mix)
```

num_samples_w_error	ul_prot_k	ml_ATL_buff
105.6	2.112	19.008

1. Set up tubes of fin clips into the plate formation on tube holder block according to plate map.
2. Create the lysis mix in a 50mL falcon tube, **do not vortex (foamy)**, and pour into 50mL reservoir.
3. Place an 8 well strip of collection tubes (H position labeled with the column number) onto the loading plate and fill with 200uL of lysis mix.
4. Double check that the next column of tubes matches the plate map
5. Place the fins in the collection tubes
6. Cap the tubes
7. When the entire plate is done, place in the incubator overnight.
8. Allow the plate to cool and check the caps to make sure they are on securely.
9. Follow the Qiagen protocol for plate extraction.

Next follow the protocol for gel and pico_plate to check quality and quantity of extracts