

# Extraction plan for APCL18\_283-APCL18\_376

2018-07-11

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_283	APCL18_291	APCL18_298	APCL18_306	APCL18_314	APCL18_322	APCL18_330	APCL18_338	APCL18_345	APCL18_353	APCL18_361	APCL18_369
B	APCL18_284	APCL18_292	APCL18_299	APCL18_307	APCL18_315	APCL18_323	APCL18_331	APCL18_339	APCL18_346	APCL18_354	APCL18_362	APCL18_370
C	APCL18_285	APCL18_293	APCL18_300	APCL18_308	APCL18_316	APCL18_324	APCL18_332	APCL18_340	APCL18_347	APCL18_355	APCL18_363	APCL18_371
D	APCL18_286	XXXX	APCL18_301	APCL18_309	APCL18_317	APCL18_325	APCL18_333	APCL18_341	APCL18_348	APCL18_356	APCL18_364	APCL18_372
E	APCL18_287	APCL18_294	APCL18_302	APCL18_310	APCL18_318	APCL18_326	APCL18_334	XXXX	APCL18_349	APCL18_357	APCL18_365	APCL18_373
F	APCL18_288	APCL18_295	APCL18_303	APCL18_311	APCL18_319	APCL18_327	APCL18_335	APCL18_342	APCL18_350	APCL18_358	APCL18_366	APCL18_374
G	APCL18_289	APCL18_296	APCL18_304	APCL18_312	APCL18_320	APCL18_328	APCL18_336	APCL18_343	APCL18_351	APCL18_359	APCL18_367	APCL18_375
H	APCL18_290	APCL18_297	APCL18_305	APCL18_313	APCL18_321	APCL18_329	APCL18_337	APCL18_344	APCL18_352	APCL18_360	APCL18_368	APCL18_376

## 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	E4839	E4847	E4855	E4863	E4871	E4879	E4887	E4895	E4903	E4911	E4919	E4927
B	E4840	E4848	E4856	E4864	E4872	E4880	E4888	E4896	E4904	E4912	E4920	E4928
C	E4841	E4849	E4857	E4865	E4873	E4881	E4889	E4897	E4905	E4913	E4921	E4929
D	E4842	E4850	E4858	E4866	E4874	E4882	E4890	E4898	E4906	E4914	E4922	E4930
E	E4843	E4851	E4859	E4867	E4875	E4883	E4891	E4899	E4907	E4915	E4923	E4931
F	E4844	E4852	E4860	E4868	E4876	E4884	E4892	E4900	E4908	E4916	E4924	E4932
G	E4845	E4853	E4861	E4869	E4877	E4885	E4893	E4901	E4909	E4917	E4925	E4933
H	E4846	E4854	E4862	E4870	E4878	E4886	E4894	E4902	E4910	E4918	E4926	E4934

```
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)

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