

Extraction plan for APCL18_189-APCL18_282

2018-07-10

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)
```

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_189	APCL18_197	APCL18_204	APCL18_212	APCL18_220	APCL18_228	APCL18_236	APCL18_244	APCL18_251	APCL18_259	APCL18_267	APCL18_275
B	APCL18_190	APCL18_198	APCL18_205	APCL18_213	APCL18_221	APCL18_229	APCL18_237	APCL18_245	APCL18_252	APCL18_260	APCL18_268	APCL18_276
C	APCL18_191	APCL18_199	APCL18_206	APCL18_214	APCL18_222	APCL18_230	APCL18_238	APCL18_246	APCL18_253	APCL18_261	APCL18_269	APCL18_277
D	APCL18_192	XXXX	APCL18_207	APCL18_215	APCL18_223	APCL18_231	APCL18_239	APCL18_247	APCL18_254	APCL18_262	APCL18_270	APCL18_278
E	APCL18_193	APCL18_200	APCL18_208	APCL18_216	APCL18_224	APCL18_232	APCL18_240	XXXX	APCL18_255	APCL18_263	APCL18_271	APCL18_279
F	APCL18_194	APCL18_201	APCL18_209	APCL18_217	APCL18_225	APCL18_233	APCL18_241	APCL18_248	APCL18_256	APCL18_264	APCL18_272	APCL18_280
G	APCL18_195	APCL18_202	APCL18_210	APCL18_218	APCL18_226	APCL18_234	APCL18_242	APCL18_249	APCL18_257	APCL18_265	APCL18_273	APCL18_281
H	APCL18_196	APCL18_203	APCL18_211	APCL18_219	APCL18_227	APCL18_235	APCL18_243	APCL18_250	APCL18_258	APCL18_266	APCL18_274	APCL18_282

ONLY DO THIS ONCE

Generate extract numbers for database

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

Warning: Missing values are always removed in SQL.

Use `MAX(x, na.rm = TRUE)` to silence this warning

```
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))
```

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	E4649	E4657	E4664	E4672	E4680	E4688	E4696	E4704	E4711	E4719	E4727	E4735
B	E4650	E4658	E4665	E4673	E4681	E4689	E4697	E4705	E4712	E4720	E4728	E4736
C	E4651	E4659	E4666	E4674	E4682	E4690	E4698	E4706	E4713	E4721	E4729	E4737
D	E4652	E4647	E4667	E4675	E4683	E4691	E4699	E4707	E4714	E4722	E4730	E4738
E	E4653	E4660	E4668	E4676	E4684	E4692	E4700	E4648	E4715	E4723	E4731	E4739
F	E4654	E4661	E4669	E4677	E4685	E4693	E4701	E4708	E4716	E4724	E4732	E4740
G	E4655	E4662	E4670	E4678	E4686	E4694	E4702	E4709	E4717	E4725	E4733	E4741
H	E4656	E4663	E4671	E4679	E4687	E4695	E4703	E4710	E4718	E4726	E4734	E4742

```
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