

9/10 Phil #312

40 μ l DNA
6 μ l H_2O

coveris spin 11,000 rpm 1 min x 2

① DNA Repair

45 μ l DNA
8.5 μ l H_2O
6.5 μ l FFE buffer
2 μ l enzyme
102 μ l

into PCR tube \rightarrow inc. 20°C x 15 min

transfer to 1.5 ml tube

add 62 μ l AMPure beads

inc. 10 min RT & flicking

spindown & pellet on magnet

wash w/ 70% EtOH 200 μ l x 2

spin & remove last of EtOH & dry

resuspend w/ 48 μ l H_2O incubate 5 min.

pellet & remove 46 μ l H_2O

Qubit 11.4 ng/ μ l (508 ng)

② End Prep

45 μ l DNA
7 μ l Ultra II end prep buff.
3 μ l " mix
5 μ l DCS
60 μ l

inc 20°C 5 min, 65°C 5 min

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trans. to 1.5ml tube
 add 60 μ l Ampure beads
 inc. 10min
 spin down & magnet
 wash w/ 70% EtOH 200 μ l x 2
 spin down & dry
 resuspend w/ 3 μ l H₂O & inc. 5min
 Qubit 17.6 ng/ μ l (526 ng)

③ Adapter Ligation

30 μ l DAA
 20 μ l Adapter mix (AMX10)
 5 μ l Blunt/TA Master mix
 10 μ l

incubate RT 10min

④ XP Bead Binding

add 40 μ l Ampure Beads
 mix & incubate flicking RT 10min
 spin & pellet

add 140 μ l ABB resuspended pellet x 2

remove from magnet add 15 μ l ELB 10min
 pellet & remove

⑤ Run

35 μ l RBF
 4.5 μ l H₂O
 25.5 μ l UB
 10 μ l Library
 75 μ l

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