

Before the day,

- 1.) Make Guba solution of .2L
- 2.) Make 4% NaHCO₃ of 1L (≈ 50mM).
- 3.) Cut the gauze of 100 x 150 cm and autoclave.
- 4.) Prepare cold dH₂O of 10 L (4 degree).
- 5.) Wash the grinder and put it in the cold room.
- 6.) Wash the 3-5 L beaker or a can and put it in the cold room.
- 7.) 1L cylinder.
- 8.) Plastic spoons or similar stuff.
- 9.) 2-3 kitchen knives.
- 10.) Check the amount of ice.
- 11.) A plastic box or similar for burying a rabbit
- 12.) Put 1 bottle Acetone in the cold room.

The day,

Chicken breast (Sasami)

Put (bury) the body in the ice for 20 min.

-Remove muscle from the body

-Measure weight (g).

≈ 1vol (ml) 461 g @2017/08/01, 7:45am

-Add 3 vol. Guba solution.

1383ml

-Gently stir at 0 degree for 3-5 minutes.

(5 min)



Centrifuge at 5K, 10min, at 0 degree.

Get the pellet. (Supernatant includes Myosin.)

-Scoop pellet and put it into a 5L container at Room Temperature (RT).

-Add 4 vol. Cold dH₂O and stir FAST and SMOOTHLY for 2 min.

F Filter with gauze.

-Place pellet into 5L container carefully.

(Pick pellet from gauze using spoons.)

-Add 4% NaHCO₃ (≈ 50mM) of 500mL + dH₂O 4.5 L (Room temperature) **Make 0.4%** into 5L container at RT.

-Stir gently and leave it for 10 min.

(Scoop fat and metal involved in pellet with a beaker.)

F Remove water well.

(Repeat)

@8:40am

-Place pellet into 5L container carefully.

-Add 4% NaHCO₃ (≈ 50mM) of 500mL + dH₂O 4.5 L (Room temperature) **Make 0.4%** into a can at RT.

-Stir gently and leave it for 10 min.

F. Filter with gauze.

Place pellet into 5L container carefully.

-Add cold dH₂O of 5 L into the container.

-Stir the pellet quickly and well.

F. Filter with gauze.

(Repeat)

Place the pellet into 5L container carefully.

-Add cold dH₂O of 5 L into the container.

-Stir pellet quickly and well.

F. Filter with gauze.

Place pellet into 5L container carefully.

Add Cold dH₂O of 1-2 L.

Make the viscous liquid. i.e.) Loose (half melted) Milk shake.

Incubate the viscous liquid at 0 degree (in the ice bath) for 6 hrs. @9:00am



Centrifuge at 6 K, 20min, at 0 degree.

@14:35

Get the pellet. (Supernatant includes Myosin.)

Place pellet into 5L container carefully.

@15:00

-Add cold Acetone of 1.5L (about a half bottle).

-Stir pellet **quickly** and well.

F. Filter with gauze.

(Repeat)

Place pellet into 5L container carefully.

-Add cold Acetone of 1.5L (about a half bottle).

-Stir pellet **quickly** and well.

F. Filter with gauze.

(Repeat x 3 times, 5 min for 1st, 10 min for 2nd, 20 min for 3rd wash)

Place pellet into 5L container carefully.

-Add Acetone of 1.5L (about a half bottle) **ROOM TEMPERATURE.**

-Stir pellet quickly and well.

F. Filter with gauze.

@15:55

Wrap acetone powder with gauze and put it under a chemical hood for over night at RT.

Aliquot acetone powder and freeze at -20 degree. **Total yield 34 g**

Fin.