Freeze Crusher µT-48

Powerful crushing of frozen samples with liquid nitrogen. 48 samples can be treated simultaneously. Optimum for extracting hard samples, proteins susceptible to heat denaturation, RNA, etc.

•Example of various frozen crushed samples including inanimate objects. --> P.106-P.107

Features

- Crushing of frozen sample in vessels with liquid nitrogen
- •2 mL Microtube or Dedicated metal container are used
- The throughput is 0.2 g to 2 g (Depends on the vessels)

Applications

- Crushing of Yeast, Mold, Tissue piece of animals and plants
- Crushing of bones, teeth and limbs of small animals
- Crushing of Wire covering and Plastics, Asbestos samples, etc.





The procedure for freeze crushing Work gloves are worn when freezing the sample with liquid nitrogen in the photo. Use gloves that are suitable for handling liquid nitrogen in actual use



Put the specified amount crusher into the vessel



When a vessel requires a holder, attach it in the holder.



Soak and freeze in liquid nitrogen until bubbling stops.



Quickly attach to the $\mu\text{T-48}$

Power supply



Close the hood and check the time and shaking speed, and then start crushing.



After crushing, take out the next process

Adapted to various samples with 3 types of vessel holders

Powerful crushing of frozen samples with liquid nitrogen. Living tissues and organs, Hard tissues such as bones, etc. and also some inanimate samples such as rubber and plastic can be crushed. Adapted to Marketed 2 mL tubes and Dedicated stainless steel crushing vessels.

Optional accessories: Vessel holders



Product Name / Mo	odel / Remarks
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O 48pcs-Holder for µT-48 TH-0248T

1 pc of Holder (Capacity: 48 pcs of 2.0 mL round bottom Microtubes) and 100 pcs of Metal crushers come as a set. Sample throughput is 0.1 to 0.2 g/1 pc.

2 3pcs-Holder for µT-48 TH-0203T

4 pcs of Holders (Capacity: 3 pcs of 2.0 mL round bottom Microtubes), 24 pcs of Metal crushers and Rack come as a set. 4 holders can be attached to μT -48 (max. 12 tubes of 2.0 mL), which is superior to TH-0248T in heat retention.

⊙ Stainless steel-made strong crushing vessel TH-SPT

Crushing vessel 4 pcs. Dedicated crusher and Rack come as a set. Suitable for samples that cannot be crushed by Microtube with Metal crusher. Larger amount of samples (1.0 to 2.0 g/1 pc) can be crushed than that of Microtubes.

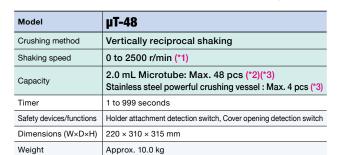
Recommended shaking speed in each crushing sample

- •Stainless steel strong crushing vessel: Up to 1000 r/min
- •Metal crusher: Up to 1200 r/min
- •Crushing beads: Up to 1600 r/min

A shaking speed that is higher than the above speeds may cause breakage of tubes and vessels. Therefore, make sure to observe the shaking speeds above.

USER'S VOICE

Very useful when extracting substance susceptible to denaturation and degradation of RNA and proteins



(*1)Around 1200 r/min should be necessary and sufficient condition to crush the sample in actual use. (*2)Eppendorf "Safe-Lock Tube 2 mL" is recommended. (*3)Microtube and Stainless powerful crushing ssel are available as an option.

AC100V/1A (Need a step-down transformer)



Example 1 Freeze crushing of various samples including inanimate samples

Embrittlement by freezing enables strong crushing. The freeze crushing with µT-48 is also suitable for Obligatory anaerobe samples.

•Freeze Crusher µT-48 --> P.105

Test results

•Freezing method Immerse the vessels with the sample and crusher into liquid nitrogen (2.0 mL: Vessel holder) and then freeze them.

•Rushing time 30 sec (Additional 30 sec if not completely crushed)

Whether powder forms or nearly forms (Cut samples into any size that can be put in vessels). •Judgment whether sample is crushed

 Vessels Safe-Lock tube 2.0 mL ...Marketed product (Made by Eppendorf)

> Metal crusher......Included in Optional 48 pcs-holder for µT-48 (used in this experiment) Stainless steel-made strong crush vesselsOptional parts (Dedicated crusher is included.)

Chicken thigh





Vessels: Safe-Lock tube 2.0 mL Sample volume: 0.1 g Shaking speed: 1200 r/min Crushed with: Metal crusher

Human hair





Vessels: Safe-Lock tube 2.0 mL Sample volume: 0.1 g Shaking speed: 1200 r/min Crushed with: Metal crusher

Human nails





Vessels: Safe-Lock tube 2.0 mL Sample volume: 0.2 g Shaking speed: 1200 r/min Crushed with: Metal crusher

Mouse skin (with body hair)





Vessels: Safe-Lock tube 2.0 mL Sample volume: 0.2 g Shaking speed: 1200 r/min Crushed with: Metal crusher

Mouse heart





Vessels: Safe-Lock tube 2.0 mL Sample volume: 0.2 g Shaking speed: 1200 r/min Crushed with: Metal crusher

Mouse tail





Vessels: Stainless steel-made strong crush vessel Sample volume: 1 g Shaking speed: 1000 r/min Crushed with: Dedicated crusher

Hypocotyl of Radish





Vessels: Safe-Lock tube 2.0 mL Sample volume: 0.2 a Shaking speed: 1200 r/min Crushed with: Metal crusher

Okra seeds





Vessels: Safe-Lock tube 2.0 mL Sample volume: 2 pcs Shaking speed: 1200 r/min Crushed with: Metal crusher

Hard rubber (Polychloroprene)



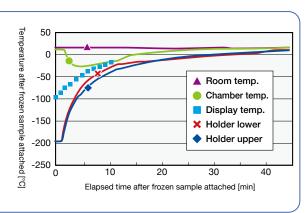


Vessels: Stainless steel-made strong crush vessel Sample volume: 2 q Shaking speed: 1000 r/min Crushed with: Dedicated crusher

Frozen sample/Holder temperature (Reference)



The 48 pcs-holder for 2 mL tube TH-0248T with 48 tubes and Metal crusher that was frozen with liquid nitrogen was attached to the unit. Then, the temperature change of each part while shaking at 1200 r/min was measured. The cryogenic temp. was completely kept for 30 to 60 sec which was required for crushing. The display temp. indicates the temp. stage of the top surface on which the holder is placed.



Freeze crushing of Plastic samples using Freeze crusher µT-48 with Stainless steel-made strong crush vessel.

•Freeze Crusher µT-48 --> P.105

Results and Examination

We tried some crushing of samples such as polystyrene, polypropylene, and polycarbonate that are well known. Each result is as follows.



Polystyrene(1): Fair





Sample shape and volume: φ6 mm ball shaped, 1 g Shaking speed: 1000 r/min Crushing time: 300 sec

Polystyrene 2: Fair



Sample shape and volume: φ6 mm ball shaped, 0.4 g Shaking speed: 1000 r/min Crushing time: 300 sec

Polystyrene3: Excellent





Sample shape and volume: 10 mm square chip shaped, 0.5 g Shaking speed: 1000 r/min Crushing time: 180 sec

Polypropylene 1: Good

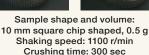




Sample shape and volume: 10 mm square chip shaped, 0.5 g Shaking speed: 1000 r/min Crushing time: 150 sec

Polypropylene2: Good





Polycarbonate: Poor





Sample shape and volume: 10 mm square chip shaped, 0.5 gShaking speed: 1000 r/min Crushing time: 300 sec

Polystyrene was able to be powdered completely (3). However, in case of the \$\phi6\$ mm ball-shaped sample, there were large fragments that remained at a certain rate even after trying with different amounts and crushing times (①②). It seems that the ball-shaped sample remained uncrushed if it is stuck in upward of the crusher.

The result suggests that the shape of the sample is better to be like a chip shape (or a tablet shape) Polypropylene in order to be crushed into fine fragments. However, it was not crushed into powder (1). In order to improve (although the shaking speed limit is exceeded when using a strong crushing container), the shaking speed was performed at 1100 r/min for 5 minutes. It becomes fine, but like a braided piece of cotton (2). At this stage, it becomes difficult to collect unless suspended in a solvent.

Polycarbonate proved to be difficult to crush. Even if the shaking speed was reduced to 1100 r/min or by reducing sample amount, the result of this experiment was that only a small amount of powder was produced and the chip shape remained almost unchanged.

Freeze crushing procedure when using stainless steel-made strong crushing vessel

An adjustable plier is useful for taking the Stainless steel-made strong crushing vessel (referred to as crushing vessel) in and out of the liquid nitrogen. Make sure to wear leather gloves when touching the frozen crushing vessel or the vessel holder that has become cold by contacting the frozen vessel. Make sure to ventilate the room well when using liquid nitrogen as there is risk to get Anoxia unknowingly because vaporized liquid nitrogen can become a huge volume of nitrogen gas.



Pour liquid nitrogen into a



Put the weighed sample *2 into the crushing vessel.



Put the dedicated crusher into the crushing vessel and



Submerse the crushing vessel in liquid nitrogen completely with the adjustable plier, etc.



Close the Styrofoam container with the lid and wait until the liquid nitrogen is boiling.



Take out the crushing vessel with *3 adjustable plier, etc. when the boiling settles down



With your glove, take the crushing vessel that you have



Place the crushing vessel on the unit and put on the lid of the mounting rack. *4



Secure the lid of the rack with the black-colored knob. *5



Shake at the prescribed speed and time.



After the shaking is complete, open the lid and



- Desirable to use the minimum-sized polystyrene foam container that the required number crushing vessels can be immersed to minimize the amount of liquid nitrogen used.
 The processing capacity of the crushing vessel is 1.2 g per 1 pc while it is better to make it to 0.5 g per 1 pc for plastic samples (Up to 1 g polystyrene can easily be crushed by freezing).
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 *3.Wait for at least 2 minutes after the boiling is settled out to freeze the sample in the crushing vessel sufficiently.

 *4. Place at least two "frozen" Stainless steel-made strong crushing vessels for balance and secure fixation. Because metal shrinks when frozen, both vessels must be frozen to ensure a firm fixation.

 *5.In December 2017, the rack was changed to a new type that does not require thumbscrews to secure vessels.

 *6.The crushed sample may stick to the crusher so tap it with the inner wall of the vessel to drop it.

 *7.If the crushing is insufficient, return the crusher to the unit to freeze it and shake again.



the sample is crushed