

Freeze-Crush Apparatus

SK-Mill

(SK-100/SK-200)

Instruction Manual



Please familiarize with this Instruction Manual before using SK-Mill to operate it correctly. And please keep this manual near at hand to refer whenever necessary.

Funakoshi Co., Ltd.

Introduction

It is very grateful to your purchasing SK-Mill. This instruction manual explains how to operate our freeze-crush apparatus “SK-Mill (SK-100/SK-200) ”.

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Package contents

Please confirm the contents carefully before use.

Package contents	Quantity
Outer Case	1
Tube Holder	1
Crushers	1 0
Sample Tubes, 2 ml (Free samples)	3 0
Instruction manual (This manual)	1

1 Important Safety Information

Please review the following warnings and cautions precisely for safety.

WARNINGS

- Please operate liquid nitrogen with great care whenever handling, because liquid nitrogen is very dangerous. It is important to wear protective equipment, including safety goggles and protective gloves whenever handling liquid nitrogen. We recommend leather gloves which do not soak.
- It is dangerous to expose skin when you wear half sleeves and short trousers. Always wear working clothes not to expose skin.
- It is very dangerous to dip Sample Tubes or Tube Holders into liquid nitrogen directly when you wear gloves etc., because liquid nitrogen soaks into gloves. Be sure to use equipment, including wire gauzes and large spoons during operation.
- Do not pour liquid nitrogen into Sample Tube, because it is very dangerous. Such operation elevates internal pressure of Sample Tube and could burst.
- Please take care of the ventilation in your room whenever using liquid nitrogen. Poor ventilation is very dangerous, because it may cause low oxygen concentration and hypoxia.
- It is very dangerous to handle the cooled Sample Tube and/or the cooled Tube Holder with bare hands. In this case it is important to wear protective equipment, including safety goggles and protective gloves.
- Carefully avoid fragile matters and/or persons in your laboratory, because this apparatus may be broken because of fatigue and deterioration of material due to prolonged use of this apparatus. Vigorous shaking is dangerous due to coming off Outer Case cap and slipping from your hands. Please handle with adequate holding and shaking.
- Please operate this apparatus carefully, because you could injure your wrist and your arm etc., when you shake this apparatus too violently and operate it for many hours a day.
- Please confirm that Outer Case cap is closed surely. When the cap is open, the ultra low temperature Tube Holder could jump out and is very dangerous.

CAUTIONS

- Outer Case could be broken due to the influence of ultra low temperature. Stop immediately to operate Outer Case when it has grown old or cracked.
- Some kind of tubes does not fit SK-Mill. We supply special 2 ml Sample Tube. The other tubes may not fit and cause damage. Some tubes are weak at low temperature. Please consult FUNAKOSHI when you use tubes of the other makers.
- Do not strike Outer Case to bench-top etc. Outer Case made of plastic may be broken. Shaking this apparatus violently is enough to crush.
- Do not operate this apparatus except for freeze-crush.
- Be sure that the screw of each parts is tight before operation.

2 The name of each part (SK-100)

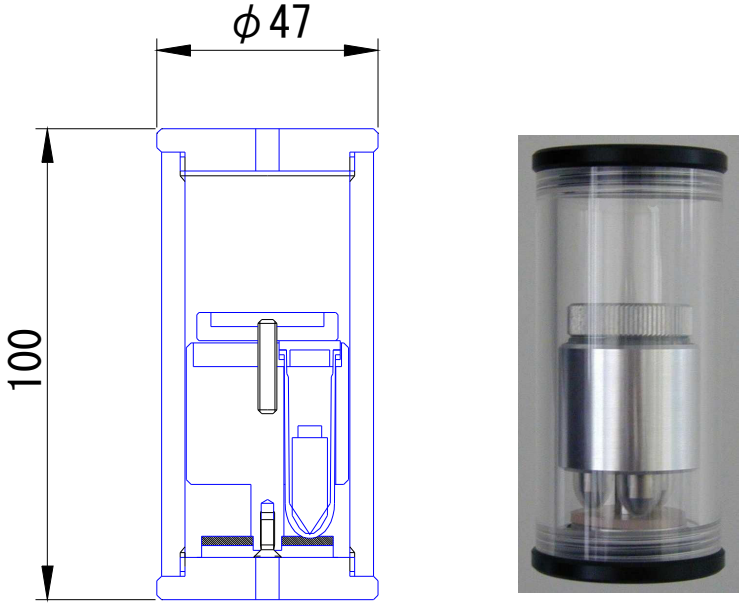


Fig.1 Structure and Overview



Fig.2 Outer Case

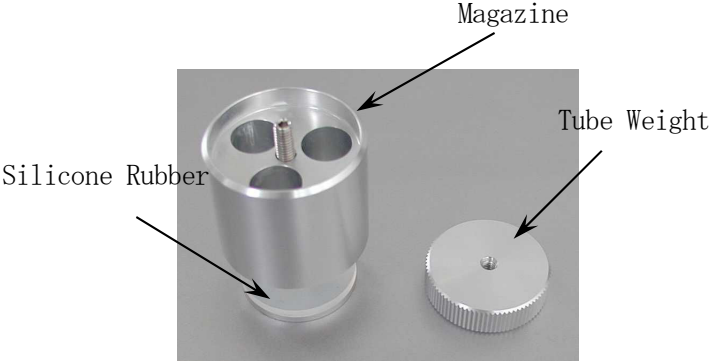


Fig.3 Tube Holder



Fig.4 Crusher



Fig.5 2 ml Sample Tube

3 Operation

3-1 Pretreatment

- (1) First of all, be ready to use your sample and SK-Mill.
- (2) Put the sample into Sample Tube. The standard sample size is like a grain of wheat.
- (3) Put Crusher into Sample Tube as shown in Fig. 1, and close the lid of Sample Tube.

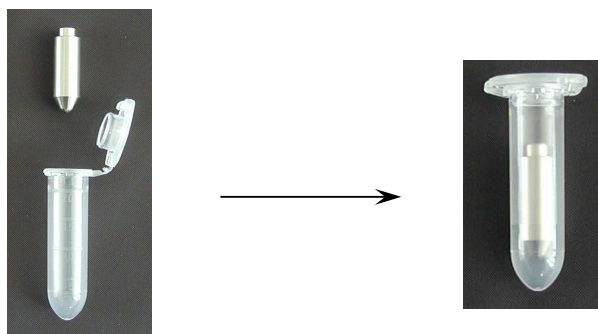


Fig.1 Setting Crusher

Note) Refer 4-1-2 when the sample is not solid.

- (4) Set the Sample Tubes to Magazine. Insert the Sample Tubes until the bottom of the Sample Tubes reaches the bottom Silicone Rubber of Tube Holder firmly, whenever setting.

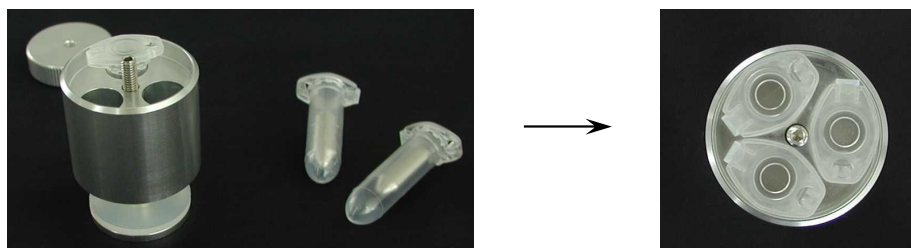


Fig.2 Setting Sample Tubes

- (5) Fix the Sample Tubes to Magazine firmly with Tube Weight. In this occasion, do not tighten Tube Weight too much to avoid damage of the Sample Tubes, because the Silicone rubber elasticity cannot be retained by too much pressure. As a standard, after the Tube Weight reached to the Sample Tube, please tighten it further half turn.



※. Above photo shows “Safe-Lock Tube”(Eppendorf Co., Ltd.) as the Sample Tubes.

Note) In continuous work, Tube Holder etc. may be at the Ultra low temperature after the first work. In this case use protective gloves etc. for safety.

- (6) Dip the set Tube Holder into liquid nitrogen for about one minute and freeze it.
A standard freezing period is that no bubbles generate as shown in Fig. 4.



Fig. 4 Dipping in liquid nitrogen

⚠ WARNING

Please operate adequate equipment, including wire gauzes and large spoons which can take out Tube Holder, because liquid nitrogen is very dangerous. Please use protective equipment, including safety goggles and protective gloves for safety.

- (7) Take out Tube Holder from liquid nitrogen when the freezing treatment is finished.
After liquid nitrogen adhered to Tube Holder has completely removed, insert Tube Holder into Outer Case and close Delrin Cap firmly.
Operate quickly not to thaw out the sample and crush it immediately.
If the freezing is sufficient, it takes about four minutes until the sample thaws out. Finish all operation within three minutes.



Fig. 5 Setting into Outer Case

Outer Case has neither up nor down direction.
Tube Holder can insert into Outer Case from every sides.

3-2 Crushing

(1) Crush the samples, holding Tube Weight upside, and shake up and down with your hand. Shaking speed is as you hammer a nail.

The standard crushing period is about one minute.

Note) Crushing condition is different with samples. Decide crushing time on experiment.

Refer 4-1-2 when the sample is not solid.

(2) Take Tube Holder out of Outer Case after the crushing is over.



Fig. 6 Crushing

Keep safety with enough protective equipments, for liquid nitrogen could remain in the inside of Outer Case.

(3) Take Sample Tubes out of Tube Holder.

(4) Take Crusher out of Sample Tube with tweezers etc.

Note) You can proceed to the next step such as melting the sample into solution without taking out. Doing so, you can use samples most efficiently.

3-3 Posttreatment

(1) Dispose Sample Tubes which has once used.

(2) As Crusher is made of stainless steel, you can use it repeatedly.

You should clean and store it avoiding any crack.

(3) Crusher tends to rotate on the bench-top and to fall down.

Check Crusher is free of crack.

Cracks could cause damage when you use with Sample Tube.

4 Troubleshooting

4-1 The sample can not be crushed well.

4-1-1 Is sample adhere to Sample Tube or to the sidewall of Crusher ?

- (1) Tap the arrow position in Fig.7 gently at the corner of desk etc. , and gather the sample to the bottom of Sample Tube. Then perform the freeze-crush again according to “3. Operation”.

Note) We advise you to crush the sample set on the bottom point of Crusher.



Fig. 7 Method to recover

4-1-2 Is the sample liquid such as yeast or Fungi ?

- (1) Insert the sample into Sample Tube and soak it into liquid nitrogen for the first time.
- (2) Taken it out of liquid nitrogen, insert Crusher into Sample Tube, set it into Tube Holder, and freeze again.
- (3) Insert the frozen Sample Tubes into Outer Case, shake the Outer Case up and down for two to three minutes as speedily as possible, and crush.

Note) If you continue crushing process more than three minutes, the samples may melt.

- (4) If the crushing is incomplete, take Sample Tube out of Tube Holder, tap the arrow position in Fig.7 at the corner of desk etc, and gather the sample to the bottom of Sample Tube.
- (5) Insert Sample Tube into Tube Holder, and repeat (2) and (3) again.

4-2 Low recovery

4-2-1 Does the crushed sample adhere to Crusher?

- (1) Tap the arrow position in Fig.7 gently at the corner of desk etc. and recover the adhered sample from Crusher.
- (2) There is another way. Remove the Crusher with tweezers : add solution and recover the sample adhered to Crusher.

5 Aftercare

Repairing is necessary due to damage and/or deterioration of Outer Case, Derlin Cap, and Silicone Rubber. Please inform to FUNAKOSHI.

FUNAKOSHI CO., LTD.

INTERNATIONAL BUSINESS DEPARTMENT

OCHANOMIZU MEIKEI BUILDING 2F, 2-3 KANDASURUGADAI, CHIYODAKU, TOKYO 101-0062, JAPAN

TEL:81-3-3292-6641 FAX:81-3-3292-6645

<http://www.funakoshi.co.jp/>

E-mail : trading@funakoshi.co.jp