

User guide

# **10x Genomics Sample Requirements**

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# 10X genomics sample requirements

For our 3' gene expression and 5' V(D)J immune profiling services

## Sample types

We accept:

1. **Cryopreserved samples**
2. **Methanol fixed samples**  
(not compatible with immune profiling V(D)J 5')
3. **Fresh samples**  
(only when agreed upon prior to experiment)

Please make sure to report any Biological hazards.

## 1. Cryopreserved sample requirements

### 1.1 Cryopreservation

Cryopreservation protocol	The protocol depends on your cell type, and as such, the protocol varies. If you have a protocol available and tested for your cell type, we strongly advise using this. 10X genomics has a few demonstrated protocols for cryopreservation, but note that these may not be suitable for all cell types. You can find those protocols <a href="#">here</a> (human/mouse cell lines) and <a href="#">here</a> (PBMCs). For the cryopreservation of nuclei, 10x genomics recommends using <a href="#">this</a> protocol (note: this is not the recommended protocol for isolating the nuclei).
Thawing protocol	When available, please send your validated thawing protocol to <a href="mailto:lab@scdiscoveries.com">lab@scdiscoveries.com</a>

## 1.2 Cell parameters

Always check cell number and viability of all samples before cryopreservation to ensure the best processing quality. When it is the first time using a cryopreservation method, please thaw a test sample and recheck cell number and viability before submitting your samples.

<b>Cell numbers*</b>	Recommended	>10 <sup>6</sup> cells/sample, in duplicate
	Minimal	500,000 cells/sample, in duplicate
<b>Viability*</b>	Recommended	>90%
	Acceptable	70%-90%

*\* These are optimal numbers for the processing of samples. However, when it is not possible to obtain enough cells, duplicate samples, or a high enough viability: please do not hesitate to contact us so that we can think along with you. We can discuss the options together and find the best solution to run the experiment.*

## 1.3 Storage

<b>Container</b>	Cryovial
<b>Medium</b>	Cryopreservation media, composition depending on protocol
<b>Labeling</b>	Label tubes clearly and use compatible markers and/or labels
	Use short unambiguous names (e.g., CTR1, EXP1a, GR2-M3)
<b>Temperature</b>	Store according to protocol (-80°C/Liquid nitrogen), ship on dry ice

## 1.4 Shipping

All shipments can be sent to:  
Single Cell Discoveries  
Attn. Lotte Koopman  
Uppsalalaan 8  
3584CT Utrecht  
The Netherlands

It is also possible to personally hand over your samples at this same address. If you prefer this, please let us know via email ([lab@scdiscoveries.com](mailto:lab@scdiscoveries.com)). At the reception, you can ask for Single Cell Discoveries.

Sample submission	Please complete our online sample submission form before shipment: <a href="https://www.scdiscoveries.com/customers/submit-samples/">https://www.scdiscoveries.com/customers/submit-samples/</a>
	Add the excel form of the sample submission form to the package. This way, our lab team can quickly identify the samples.
Shipping method	Ship with dry ice.
	A <b>minimum of 5 kg dry ice</b> for all EU/USA shipments is required
	Please ship the samples on a <b>Monday or Tuesday</b> , to prevent the package from getting stuck over the weekend.
	After shipment, please send the tracking code to <a href="mailto:lab@scdiscoveries.com">lab@scdiscoveries.com</a>
Packaging	Use a sturdy dry ice box and place it inside a cardboard box.
	Include a "this side up" marking on the box.
Sample packing	Close the cryovials well
	Wrap the cryovials tightly in parafilm
	Secure the cryovials in an Eppendorf box with an insert
	Make sure the samples are upright within the package.

## 2. Methanol fixed samples

Please note that immune profiling 5' V(D)J is not supported on methanol fixed cells. Also, RNA complexity can decrease with methanol fixation. We do have good experiences with methanol fixation, and it can be a good option for example when samples need to be collected at strict time points, when cryopreservation is not feasible or when pathogens are present within the sample.

### 2.1 Methanol fixation

MeOH fixation protocol	If you have a protocol available and tested for your cell type, it is possible to use this protocol.
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	In general, we recommend using the 10x genomics demonstrated protocol for MeOH fixation. You can find that protocol <a href="#">here</a> .
Rehydration protocol	Our lab team uses the 10x genomics demonstrated protocol to rehydrate the cells.

## 2.2 Cell parameters

Always check cell number and viability of all samples before MeOH fixation to ensure the best processing quality. Due to the fixation, it is not possible to check viability after rehydration.

<b>Cell numbers*</b>	Recommended	>10 <sup>6</sup> cells/sample, in duplicate
	Minimal	200,000 cells/sample, in duplicate
<b>Viability*</b>	Recommended	>90%
	Acceptable	70%-90%

*\* These are optimal numbers for the processing of samples. However, when it is not possible to obtain enough cells, duplicate samples, or a high enough viability: please do not hesitate to contact us so that we can think along with you. We can discuss the options together and find the best solution to run the experiment.*

## 2.3 Storage

<b>Container</b>	(Screwcap) Eppendorf tubes
<b>Medium</b>	(80%) MeOH
<b>Labeling</b>	Label tubes clearly and use compatible markers and/or labels
	Use short unambiguous names (e.g. CTR1, EXP1a, GR2-M3)
	Always label your tubes on the lids and the side
<b>Temperature</b>	Store according to protocol (-20°C or -80°C ≤ 6 weeks), ship on dry ice

## 2.4 Shipping

All shipments can be sent to:  
 Single Cell Discoveries  
 Attn. Lotte Koopman  
 Uppsalalaan 8

3584CT Utrecht  
The Netherlands

*It is also possible to personally hand over your samples at this same address. If you prefer this, please let us know via email ([lab@scdiscoveries.com](mailto:lab@scdiscoveries.com)). At the reception, you can ask for Single Cell Discoveries.*

<b>Sample submission</b>	Please complete our online sample submission form before shipment: <a href="https://www.scdiscoveries.com/customers/submit-samples/">https://www.scdiscoveries.com/customers/submit-samples/</a>
	Add the excel form of the sample submission form to the package. This way, our lab team can quickly identify the samples.
<b>Shipping method</b>	Ship with dry ice.
	A <b>minimum of 5 kg dry ice</b> for all EU/USA shipments is required
	Please ship the samples on a <b>Monday or Tuesday</b> , to prevent the package from getting stuck over the weekend.
	After shipment, please send the tracking code to <a href="mailto:lab@scdiscoveries.com">lab@scdiscoveries.com</a>
<b>Packaging</b>	Use a sturdy dry ice box and place it inside a cardboard box.
	Include a "this side up" marking on the box.
<b>Sample packing</b>	Close the tubes well (especially MeOH tends to leak quickly)
	Wrap the tubes tightly in parafilm to prevent leakage
	Secure the tubes in an Eppendorf box with an insert
	Make sure the samples are upright within the package, not sideways or upside down.

### 3. Fresh samples

Fresh samples need to be loaded onto the 10X chip as soon as possible after dissociation. Fresh samples, therefore, need to be at Single Cell Discoveries within 1-1.5 hours after dissociation. Please discuss this option beforehand with us, as it requires a more tailor-made planning.

#### 3.1 Cell parameters

Always check cell number and viability of all samples before bringing the samples, to ensure the best processing quality.

<b>Cell numbers*</b>	Recommended	>10 <sup>6</sup> cells/sample, in duplicate
	Minimal	200,000 cells/sample, in duplicate
<b>Viability*</b>	Recommended	>90%
	Acceptable	70%-90%

*\* These are optimal numbers for the processing of samples. However, when it is not possible to obtain enough cells, duplicate samples, or a high enough viability: please do not hesitate to contact us so that we can think along with you. We can discuss the options together and find the best solution to run the experiment.*

### 3.2 Storage

<b>Container</b>	Closed FACS tubes or Eppendorf tubes
<b>Medium</b>	You can hand your dissociated cells over in a cell culture medium of choice (max. 10% FCS), unless the number of cells is very low (this will limit washing steps). In that case we can discuss the appropriate medium.
<b>Labeling</b>	Label tubes clearly and use compatible markers and/or labels
	Use short unambiguous names (e.g. CTR1, EXP1a, GR2-M3)
	Always label your tubes on the lids and the side
<b>Temperature</b>	Delivery of cells on ice

### 3.3 Shipping

It is possible to hand over your samples at the following address:

Single Cell Discoveries

Uppsalalaan 8

3584CT Utrecht

Phone number: +31 30 212 1905

At the reception, you can ask for Single Cell Discoveries.

<b>Sample submission</b>	Please complete our online sample submission form before delivery: <a href="https://www.scdiscoveries.com/customers/submit-samples/">https://www.scdiscoveries.com/customers/submit-samples/</a>
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	Add the excel form of the sample submission form to the package. This way, our lab team can quickly identify the samples.
<b>Shipping method</b>	Personal delivery of the samples on ice
	Please let us know at what time you will approximately arrive
	Samples need to be at Single Cell Discoveries before 16:00 CEST, unless agreed upon otherwise.
<b>Packaging</b>	Use a sturdy dry ice box with lid
<b>Sample packing</b>	Close the (FACS) tubes well
	Make sure the samples are upright and stable within the package