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## 1. Description

This product is for research use only.

<b>Components</b>	7 vials, containing: 2.5 mL of Enzyme P 2× 50 mL of Buffer X 1.5 mL of Buffer Y 1 vial of Enzyme A (lyophilized powder) 1 mL of Buffer A 1 vial of Enzyme D (lyophilized powder)
<b>Size</b>	For up to 25 digestions. The specified number of digestions is valid when digesting up to 1 g tissue in 5 mL enzyme mix.
<b>Storage</b>	Upon arrival immediately store Enzyme P in aliquots at -20 °C. Store all other components at 2–8 °C upon arrival. Reconstitute Enzymes A and D before the date indicated on the vial label. For information about reconstitution and storage after reconstitution of the lyophilized components refer to chapter 2.

### 1.1 Principle of the Multi Tissue Dissociation Kit 2

Various tissues from different species, for example, mouse kidney or rat lung, can be dissociated into single-cell suspensions by combining mechanical dissociation with enzymatic degradation of the extracellular matrix, which maintains the structural integrity of tissues.

The tissue is enzymatically digested using the kit components, and the gentleMACS™ Dissociators are used for the mechanical dissociation steps. After dissociation, the sample is applied to a filter to remove any remaining larger particles from the single-cell suspension.

Cells should be processed immediately for downstream applications, such as cell separation, cell culture, cellular or molecular analyses.

### 1.2 Background information

The Multi Tissue Dissociation Kit 2 has been developed for the gentle, rapid, and effective generation of single-cell suspensions from various tissue. It is optimized for a high yield of viable cells. Dissociated cells can be subsequently cultured or isolated using MACS® Technology. Furthermore, the single-cell suspension can be analyzed *in vitro* for phenotype distributions, and other functional, genetic, or proteomic studies performed.

### 1.3 Applications

- Dissociation of various tissue into single-cell suspensions for subsequent cell separations using MACS Technology.
- Phenotyping or enumeration of cell populations by flow cytometry or fluorescence microscopy.

### 1.4 Reagent and instrument requirements

- Serum-free RPMI 1640 or DMEM
- gentleMACS Dissociator (# 130-093-235), gentleMACS Octo Dissociator (# 130-095-937), or gentleMACS Octo Dissociator with Heaters (# 130-096-427)
- gentleMACS C Tubes (# 130-093-237, # 130-096-334)

For additional requirements please refer to the dissociation protocol at [www.miltenyibiotec.com/130-110-203](http://www.miltenyibiotec.com/130-110-203).

## 2. Reagent preparation

▲ For cell culture experiments subsequent to tissue dissociation, all steps should be performed under sterile conditions.

1. Enzyme P is ready to use. Prepare aliquots of appropriate volume to avoid repeated freeze-thaw-cycles.  
Store aliquots at -20 °C. This solution is stable for 6 months.
2. Prepare Enzyme A by reconstitution of the lyophilized powder in the vial with 1 mL of Buffer A supplied with the kit. Do not vortex. Prepare aliquots of appropriate volume to avoid repeated freeze-thaw-cycles.  
Store aliquots at -20 °C. This solution is stable for 6 months.
3. Prepare Enzyme D by reconstitution of the lyophilized powder in the vial with 3 mL of serum-free RPMI 1640 or DMEM. Close the vial and wait for at least 5 minutes while inverting every minute. Prepare aliquots of appropriate volume to avoid repeated freeze-thaw-cycles. For cell culture experiments subsequent to tissue dissociation, Enzyme D should be sterile filtered prior to aliquoting.  
Store aliquots at -20 °C. This solution is stable for 6 months.

4. Proceed with dissociation protocol of choice.  
For tissue dissociation protocols, please refer to the product page at [www.miltenyibiotec.com/130-110-203](http://www.miltenyibiotec.com/130-110-203).

Refer to [www.miltenyibiotec.com](http://www.miltenyibiotec.com) for all data sheets and protocols. Miltenyi Biotec provides technical support worldwide. Visit [www.miltenyibiotec.com/local](http://www.miltenyibiotec.com/local) to find your nearest Miltenyi Biotec contact.

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