

# thermoMACS™ Separation Unit

User manual

Order no. 130-091-136



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## Warnings and precautions

- ▲ The thermoMACS™ Separator is for research applications only. Read and follow all operating instructions carefully before proceeding with installation and operation of the system.
- ▲ The thermoMACS Separator is equipped with an extremely powerful magnet. Its magnetic field can damage computer screens, watches, floppy discs, and other objects that react to magnetic fields. Never allow the thermoMACS Separator to be closer than 20 cm to any magnetically sensitive object.
- ▲ Electric devices pose the risk of an electric shock. To reduce the risk of an electric shock, take special care while handling fluids. Caution must be taken that fluids do not enter the interior of the device. Clean up spills immediately. Unplug the power cord before cleaning the thermoMACS Separator.
- ▲ Use the thermoMACS Separator only with the enclosed power supply. Use of a non-specified power supply may damage the device.

### Limited warranty

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- If damage is due to the misuse of the thermoMACS Separator, all warranties shall be void. This includes damage to the device itself as well as any consequential damages.
- The appropriate use of the device is specified in the thermoMACS user manual.
- Warranties are void if the damage is due to the use of separation reagents which are not produced and distributed by Miltenyi Biotec. This includes damage to the device itself as well as any consequential damages.
- Under all circumstances, liability is limited to the value of the thermoMACS Separator. Any liability beyond the value is excluded.
- The thermoMACS Separation Unit is warranted for 12 months from the date of delivery.

## Warnhinweise und Vorsichtsmaßnahmen

- ▲ Der thermoMACS™ Separator darf nur zu Forschungszwecken eingesetzt werden. Vor Aufstellung und Inbetriebnahme bitte sorgfältig die Bedienungsanleitung lesen und befolgen.
- ▲ Der thermoMACS Separator ist mit einem sehr leistungsstarken Magnetfeld ausgestattet, so dass magnetisch empfindliche Objekte wie Armbanduhren, Disketten u.a. dauerhaft beschädigt werden können. Achten Sie darauf, dass der thermoMACS Separator stets mindestens 20 cm von magnetisch empfindlichen Objekten entfernt ist.
- ▲ Bei elektrischen Geräten besteht die Gefahr eines Stromschlags. Deshalb sollte mit Flüssigkeiten im Bereich des thermoMACS Separators sorgfältig umgegangen werden. Vermeiden Sie es, Flüssigkeiten in den Geräteinnenraum eindringen zu lassen. Ziehen Sie vor der Reinigung des thermoMACS Separators den Netzstecker.
- ▲ Der thermoMACS Separator darf nur mit dem mitgelieferten Netzteil elektrisch betrieben werden. Die Benutzung anderer Spannungsversorgungen kann zu irreparablen Schäden am Gerät führen.

## Gewährleistungs- und Garantieansprüche

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- Führt die unsachgemäße Handhabung des thermoMACS Separators zu einem Schaden, verliert der Käufer sämtliche Gewährleistungsrechte. Dieses umfaßt sowohl Schäden am Gerät selbst als auch Mangelfolgeschäden.
- Die sachgemäße Handhabung des Gerätes ist dem thermoMACS Handbuch zu entnehmen.
- Die Haftung ist ebenfalls ausgeschlossen, wenn durch die Nutzung von Fremdreagenzien ein Schaden am Gerät oder ein Mangelfolgeschaden entsteht.
- Die Haftung beschränkt sich in jedem Fall auf den Wert des thermoMACS Separators. Ein darüber hinausgehender Schaden wird in jedem Fall ausgeschlossen.
- Die Gewährleistungsfrist der thermoMACS Separation Unit beträgt 12 Monate ab Auslieferung des Gerätes.

# Précautions d'emploi

- ▲ Le séparateur thermoMACS™ est conçu pour des activités de recherche uniquement. Merci de bien vouloir porter une attention particulière aux conseils d'utilisation avant de mettre l'appareil en fonctionnement.
- ▲ Le séparateur thermoMACS est doté d'un aimant de forte puissance. Veiller à ne pas disposer le séparateur thermoMACS à moins de 20 cm d'un objet sujet à des dérèglement en présence d'un champ magnétique: montre, écran d'ordinateur, disque diquette, etc.
- ▲ Comme tout appareil électrique, il est indispensable de prendre les précautions nécessaires lors de l'utilisation de liquide à proximité du séparateur thermoMACS, afin de prévenir tout risque de choc électrique. S'assurer qu'aucun liquide ne pénètre à l'intérieur de l'appareil et que l'électricité est coupée avant tout nettoyage du séparateur thermoMACS.
- ▲ Veiller à respecter le voltage maximal indiqué sur l'appareil. Un survoltage pourrait endommager le séparateur thermoMACS de façon irréversible.

## Garantie limitée

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- Tout dommage subit par le séparateur thermoMACS suite à une mauvaise utilisation de l'appareil est exclu de la garantie. Ceci inclut aussi bien les réparations de l'appareil que le versement éventuel de dommages et intérêts.
- Pour une bonne utilisation du séparateur thermoMACS, merci de consulter avec attention le manuel d'utilisation.
- La garantie de l'appareil ne comprend pas les pannes rencontrées suite à l'utilisation de réactifs de séparation magnétique, autre que ceux commercialisés et/ou distribués par Miltenyi, en combinaison avec le séparateur thermoMACS. Ceci inclut aussi bien les réparations de l'appareil que les éventuels versement de dommages et intérêts.
- Dans tous les cas, la responsabilité du fournisseur est limitée au prix public du séparateur thermoMACS. Toute responsabilité supérieure de ce montant est exclue.
- Le séparateur thermoMACS est garanti pour une durée de 12 mois à compter de la date de livraison.

## Avvertimenti e precauzioni

▲ Il separatore thermoMACS™ deve essere impiegato esclusivamente per applicazioni di ricerca. Leggere ed osservare attentamente tutte le istruzioni per l'uso prima di procedere all'installazione ed all'utilizzazione dell'apparecchiatura.

▲ L'unità di separazione thermoMACS é dotata di un magnete estremamente potente. Il suo campo magnetico può danneggiare orologi, floppy disc, monitor ed altri oggetti che risentono dell'influenza dei campi magnetici vicini. Non collocare il separatore thermoMACS a distanze inferiori ai 20 cm da ogni oggetto sensibile all'azione del campo magnetico.

▲ Come tutte le apparecchiature elettriche, il separatore thermoMACS, in caso di utilizzo non adeguato, potrebbe causare shock elettrici. Per ridurre tale rischio è necessario porre attenzione nel maneggiare liquidi ed assicurarsi che non penetrino all'interno dell'apparecchiatura. As-ciugare immediatamente gli eventuali liquidi versati accidentalmente. Staccare il cavo d'alimentazione prima di effettuare la pulizia del separatore thermoMACS.

▲ Controllare la tensione di rete, essa deve corrispondere al voltaggio indicato sull'unità d'alimentazione. Se l'alimentatore e la tensione di rete non dovessero essere compatibili, i componenti elettrici del separatore thermoMACS potrebbero subire danneggiamenti.

### Condizioni di validità della Garanzia

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- La garanzia non si estende ai danni diretti e/o indiretti conseguenti all'uso improprio del separatore thermoMACS.
- Le modalità per il corretto utilizzo del separatore thermoMACS sono specificate nelle avvertenze e precauzioni per l'uso e nel Manuale dell'operatore allegato allo stesso.
- La garanzia non si estende ai danni diretti e/o indiretti e/o conseguenti dovuti all'utilizzo di reagenti di separazione che non siano prodotti e distribuiti da Miltenyi Biotec.
- In ogni circostanza la responsabilità è limitata al valore del separatore thermoMACS: qualsiasi responsabilità oltre il valore del separatore thermoMACS è esclusa, salvo limiti inderogabili di legge.
- L'unità di separazione thermoMACS è garantita per 12 mesi dalla data di consegna.

## Advertencias y precauciones

- ▲ El separatore thermoMACS™ es sólo para aplicaciones de uso investigacional. Antes de instalarlo y ponerlo en marcha lea cuidadosamente las instrucciones de uso.
- ▲ La unidad de separación thermoMACS está equipada con un imán extremadamente potente. Su campo magnético puede dañar relojes, discos y otros objetos que reaccionen a campos magnéticos. No permita nunca que la unidad de separación thermoMACS esté a menos de 20 cm de distancia de cualquier objeto sensible magnéticamente.
- ▲ Debido a que el uso de sistemas eléctricos puede causar shock eléctrico debe prestarse especial atención cuando maneje líquidos cerca del separatore thermoMACS. Asegúrese de que los líquidos no entran en el interior del aparato. Desenchufe el aparato antes de limpiarlo.
- ▲ Chequee el voltaje de la línea eléctrica, debe corresponder al voltaje indicado en la unidad de suministro de energía. Si no son compatibles pueden dañarse los componentes eléctricos del separatore thermoMACS.

### Garantía Limitada

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- Si el daño es debido a mal uso del sistema separatore thermoMACS, se invalidarán todas las garantías. Esto incluye daños al sistema en sí, así como cualquier daño subsiguiente.
- El uso adecuado del sistema separatore thermoMACS está especificado en el manual de uso.
- Las garantías serán inválidas si el daño es debido al uso de reactivos de separación que no han sido producidos o distribuidos por Miltenyi Biotec. Esto incluye daños al sistema en sí, así como cualquier daño subsiguiente.
- Bajo cualquier circunstancia, la responsabilidad está limitada al valor del separatore thermoMACS. Se excluye cualquier responsabilidad a partir de ese valor.
- La unidad de separación thermoMACS está garantizada por 12 meses desde la fecha de envío.

# 1. Introduction

## 1.1 Description of the thermoMACS™ Separator

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The thermoMACS™ Separator is a heatable magnetic separator specifically developed for molecular biology applications. The thermoMACS Separator, in combination with  $\mu$  Columns and  $\mu$ MACS™ MicroBeads, is used for the isolation of highly pure biomolecules such as mRNA, sequence-specific DNA or proteins. In addition to applications at room temperature, the thermoMACS Separator allows molecular biological reactions to be carried out at increased temperatures of 37 °C or 42 °C such as enzymatic reactions. The elution volume from a  $\mu$  Column may be as low as 50–150  $\mu$ L and therefore allows a highly concentrated target fraction to be collected, which is convenient for further downstream applications. The thermoMACS Separator can hold up to four  $\mu$  Columns to process four samples simultaneously.

## 1.2 Applications

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Solid phase enzymatic reactions on  $\mu$  Columns are an easy and elegant method for purification of biomolecules. While the molecule is immobilized in the column, it is incubated with an enzymatic solution. To ensure optimal reaction conditions, the column can be heated to 37 °C and 42 °C, respectively, which covers the reaction conditions for most of the enzymes used in molecular biology applications. After the reaction, buffer and reaction components can be easily and efficiently washed out, thereby allowing serial reactions to be carried out in a single  $\mu$  Column. Expensive and time-consuming purifications between single enzyme reactions is not necessary. Consequently, there is no loss of biomolecules due to the transfer from one tube to the next. This can significantly improve sensitivity of downstream assays like (real-time) PCR or microarray analysis after mRNA isolation and in-column cDNA synthesis or labeling, respectively.

Furthermore, enzymes may be immobilized in the column. This allows the enzymatic modification of substrates and subsequent elution of these while the enzyme remains on the column.

Performing an enzymatic reaction using biomolecules in a column matrix – such as the MACS® Columns – is a new application and is filed for a patent.

## 1.3 The MACS® Technology

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For magnetic purification, biomolecules are labeled with target specific  $\mu$ MACS™ MicroBeads. Subsequently they are separated in a  $\mu$  Column that is placed in a strong magnetic field.  $\mu$ MACS MicroBeads are optimized for the isolation of biomolecules, such as RNA or proteins. They are conjugated to specific molecules e.g., proteins like Protein A, Protein G, epitope tag specific antibodies, Streptavidin or single-stranded oligonucleotides, and can be used for magnetic labeling of virtually any target molecule. The colloidal behavior of the MicroBeads facilitates and accelerates the binding of  $\mu$ MACS MicroBeads to the target molecules. The MicroBead labeled sample is then applied to the column which is placed in the magnet. While the labeled molecules are retained in the magnetic matrix, the non-target molecules are effectively rinsed out of the column. As the columns flow is by gravity, tedious centrifugation steps are eliminated from the procedure.

For elution of the target molecule an appropriate elution buffer which disrupts the binding between the target molecule and its immobilized ligand, is applied onto the column. While the target molecule is collected in the eluate, the probe remains within the column. A second option is the release of the target molecule–probe complex by removing the column from the magnetic field. In this case, the complete complex can be eluted using a buffer of choice.

## 2. Technical service

### Germany/Austria/Switzerland

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Miltenyi Biotec GmbH  
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51429 Bergisch Gladbach, Germany  
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### Australia

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### Benelux

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### UK

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E-mail: macs@miltenyibiotec.co.uk

For countries not listed, please contact  
Miltenyi Biotec GmbH, Germany.

## 3. The thermoMACS™ Separation Unit

### 3.1 Equipment supplied with the thermoMACS™ Separation Unit

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#### **thermoMACS™ Separator**

**μMACS™ Sealing Solution, 1 mL** (additional Sealing Solution can be purchased separately # 130-091-160)

#### **Power supply**

#### **Main power supply cable**

#### **Low voltage power supply cable with temperature sensor**

#### **User manual**

### 3.2 Additional materials required

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#### **MACS® MultiStand (# 130-042-303)**

The MACS® MultiStand is necessary for setting up μMACS™, MiniMACS™, QuadroMACS™, OctoMACS™, and thermoMACS™ Separation Units. MACS Separation Units are magnetically attached to the metal stand. The MultiStand can hold one thermoMACS Separator.

#### **μ Columns (# 130-042-701)**

μ Columns are specifically designed for small scale molecular biology applications in combination with μMACS MicroBeads. The magnetically labeled molecules are efficiently retained in the column matrix while unlabeled material is washed away. Elution of the target molecules can be performed in a convenient, small volume, e.g., 50–100 μL. μ Columns can be used for a large variety of applications such as RNA isolation, (co-)immunopurification, tagged protein isolation, etc.

#### **Reagents**

A complete list of MACS molecular products and protocols is available at [www.miltenyibiotec.com](http://www.miltenyibiotec.com)

#### **Rack**

Please use an appropriate rack for the collection tubes.

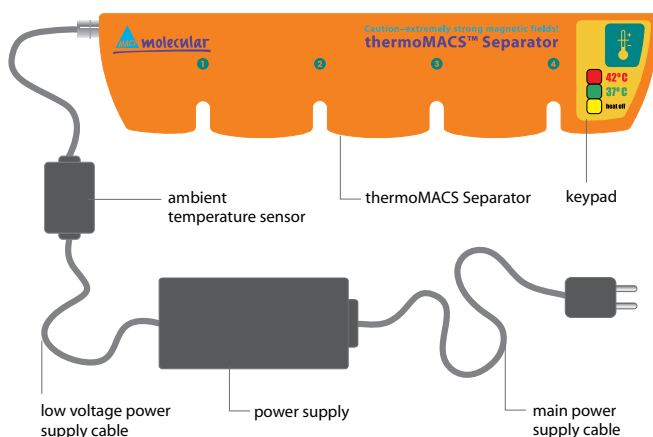
## 3. The thermoMACS™ Separation Unit

### 3.3 Key components of the thermoMACS™ Separation Unit

The key components of the thermoMACS™ Separation Unit are shown in figure 1.

1. The thermoMACS Separator generates a high gradient magnetic field to bind magnetically labeled biomolecules in the  $\mu$  Column. The magnetic field is identical to that in the  $\mu$ MACS™ Separator.
2. Heating elements are present in each column slot, heating up the reaction chamber in the  $\mu$  Column to pre-defined temperatures of 37 °C or 42 °C.
3. A microprocessor-based electronic board and thermocouples in the heating elements control the temperature of the reaction chamber in the  $\mu$  Column. This maintains a constant temperature throughout the whole separation process, which ensures stable and highly reproducible experimental results.
4. The temperature sensor integrated in the power supply cable of the thermoMACS Separator measures the room temperature. This allows precise amendment of the temperature in the column relative to variations in the local environment.

#### The thermoMACS™ Separation Unit (Figure 1)



## 4. Installation

1. Place the MACS® MultiStand on a clean and stable laboratory bench.
2. Fix the thermoMACS™ Separator to the MultiStand by holding the rear surface against the stand. It will remain attached to the MultiStand by internal magnets on the rear surface of the thermoMACS Separator.
3. Take the low voltage power supply cable and insert the output into the plug on the left side of the thermoMACS Separator. The power supply cable is correctly orientated, when the red dot on the plug meets the red dot on the connector.

▲ **Note:** Use the thermoMACS Separator only with the enclosed power supply. Using a non-specified power supply may damage the device.

4. Connect one end of the main power supply cable with the power supply and plug it into an appropriate electrical outlet.

The thermoMACS Separator is now ready for separation. The column temperature is turned to heat off (yellow LED) as the default setting.

▲ **Note:** The thermoMACS Separator and the temperature sensor should not be used in direct proximity to any heat source or strong illuminants.

## 5. Maintenance

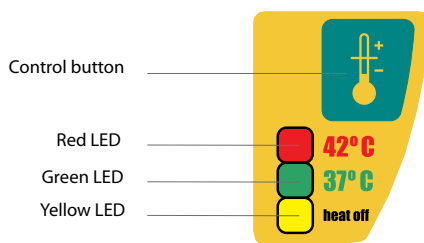
Clean up spills immediately. Unplug the power cable before cleaning the thermoMACS Separator. Use a detergent containing wash solution (e.g. 1% SDS solution) and/or alcohol (e.g. 70% ethanol solution) for disinfection and cleaning.

▲ **Note:** The thermoMACS Separation Unit should not be submerged under water, or cleaned in a dish washer, ultrasonic cleaner, autoclave, or similar appliances.

## 6. Instructions for use

The keypad (fig. 2) is used to select the temperature of the reaction chamber in the  $\mu$  Column. Gently touching the control button on the keypad changes the temperature as indicated by the three light emitting diodes (LED): the red LED for 42 °C, the green LED for 37 °C, and the yellow LED for heat off, i.e. for working at room temperature. When the target temperature is changed via the control button to 37 °C or 42 °C, the corresponding temperature LED blinks until the target temperature is reached. Then the LED is permanently illuminated.

### The keypad (Figure 2)



## 7. Status of the thermoMACS™ Separator

LED signals	Status of the thermoMACS™ Separator
<b>No LED is on</b>	The thermoMACS Separator may not be connected to the power supply.
<b>The yellow LED is on</b>	The thermoMACS Separator is in standby mode and the heating elements are deactivated.
<b>The yellow LED blinks fast</b>	The thermoMACS Separator has a hardware error. Please contact Technical Service.
<b>The green LED blinks slowly</b>	The 37 °C heating mode is on but the target temperature has not been reached.
<b>The green LED is on</b>	The target temperature of 37 °C has been reached.
<b>The red LED blinks slowly</b>	The 42 °C heating mode is on but the target temperature has not been reached.
<b>The red LED is on</b>	The temperature of 42 °C has been reached.

## 8. Isolation and processing of molecules

### 8.1 Tips & hints

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#### ▲ General considerations

Before starting an application on the thermoMACS™ Separator, consider whether the assay requires immediate heating, or heating after biomolecule separation:

#### **Immediate heating:**

Pre-heat the column in the thermoMACS Separator for 5–10 min before applying the buffer for equilibration. Use of pre-heated buffer is recommended. Apply the reaction solution directly after flow-through of the equilibration buffer. Immediately add 1 µL of µMACS™ Sealing Solution onto the µ Column.

#### **Heating after biomolecule separation:**

First, carry out the separation according to the standard µMACS protocol at room temperature (thermoMACS Separator „off“). Before starting the heated reaction, set the thermoMACS Separator to the required temperature. If not otherwise specified in the µMACS reagent user manual, wait for 5–10 minutes, then add the reaction solution. Use of pre-heated buffer for the reaction is recommended. Immediately add 1 µL of µMACS Sealing Solution.

#### ▲ Warm-up time

The thermoMACS Separator requires 5–10 minutes to reach 37 °C or 42 °C. Permanent illumination of the LED indicates that the target temperature is reached. If the µ Column is inserted in the thermoMACS Separator before it is switched on, the column matrix will reach 37 °C or 42 °C after 5–10 minutes. If the µ Column is inserted in a pre-heated thermoMACS Separator, allow an additional five minute adjustment period to ensure that the matrix in the column is heated to the correct temperature.

#### ▲ Buffer temperature

The use of cold buffer on a pre-warmed column is not recommended due to the possibility of air bubble formation in the column matrix.

#### ▲ µMACS Sealing Solution

The µMACS Sealing Solution is a unique solution that creates a closed reaction chamber in the µ Column, thereby preventing evaporation of the

## 8. Isolation and processing of molecules

reaction mixture at reaction temperatures above room temperature.

After the reaction solution is added on the heated  $\mu$  Column, 1  $\mu$ L of the  $\mu$ MACS Sealing Solution should be applied on top of the column matrix. Make sure to pipette the  $\mu$ MACS Sealing Solution directly onto the iron sphere matrix in the  $\mu$  Column so that it can spread and completely cover the surface of the matrix. If the  $\mu$ MACS Sealing Solution is applied only on the side wall of the column reservoir it will not run down to the matrix due to its small volume. The volume of the applied  $\mu$ MACS Sealing Solution should not exceed 2  $\mu$ L.

Generally, the  $\mu$ MACS Sealing Solution only needs to be applied once. It stays on top of the column matrix and does not enter the matrix, because its density is lower than the buffer densities. When buffer is applied on top of a sealed column, the  $\mu$ MACS Sealing Solution lets the buffer pass into the matrix and reseals the surface. However, the function of the  $\mu$ MACS Sealing Solution can be impaired when working with detergents or other amphipathic substances (see paragraph „If amphipathic substances...“ below).

The  $\mu$ MACS Sealing Solution does not interfere with the immobilized biomolecules in the column reaction chamber nor does it contaminate the elution fraction as long as working with detergents or other amphipathic substances is avoided (see paragraph „If amphipathic substances...“ below).

If amphipathic substances including fatty acids, phospholipids, and detergents, especially ionic detergents like SDS, are used after sealing the column with the  $\mu$ MACS Sealing Solution, the effectiveness of the sealing may be compromised. For additional incubation steps we recommend applying a further 1  $\mu$ L of the  $\mu$ MACS Sealing Solution.

If the current application is performed at room temperature (thermoMACS Separator switched to „heat off“), no  $\mu$ MACS Sealing Solution is necessary.

### 8.2 Applications & protocols

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For specific applications please refer to [www.miltenyibiotec.com](http://www.miltenyibiotec.com).

## 9. thermoMACS™ accessories

MACS® Product	Quantity	Order no.
MACS MultiStand	1 piece	130-042-303
μ Column	20 columns	130-042-701
μMACS Sealing Solution	1 mL	130-091-160

## 10. Related products

Please refer to [www.miltenyibiotec.com](http://www.miltenyibiotec.com) for a complete list of MACSmolecular products.

MACS Product	Application	No. of tests per kit	Order no.
<b>mRNA isolation and cDNA synthesis</b>			
μMACS mRNA Isolation Kit - Small Scale	mRNA isolation from cells, tissue and blood	20	130-075-201
		10	130-090-276
μMACS mRNA Isolation Kit - Large Scale	mRNA isolation from cells, tissue and blood	8	130-075-101
		4	130-090-277
μMACS mRNA Isolation Kit - For Total RNA	mRNA isolation from total RNA	8	130-075-102
μMACS One-step cDNA Kit	mRNA isolation and in-column cDNA synthesis	20	130-091-902
μMACS One-step cDNA Labeling Kit	mRNA isolation and in-column cDNA labeling	20	130-092-443
μMACS One-step T7 Template Kit	mRNA isolation and in-column amplification	20	130-092-866
<b>Protein and molecular complex isolation</b>			
μMACS Protein A MicroBeads	Immunopurification of proteins	20–40	130-071-001
μMACS Protein G MicroBeads	Immunopurification of proteins	20–40	130-071-101
μMACS Epitope Tag Protein Isolation Kits	Isolation of His-, HA-, c-myc-, GST-, and GFP-tagged proteins	40	
μMACS Streptavidin Kit	Isolation of biotinylated molecules and their targets	20	130-074-101
μMACS FactorFinder Kit	Isolation of transcription factors	20	130-092-317

## 11. Technical data and specifications

1. Size (thermoMACS Separator): 191×40×33 mm
2. Weight (thermoMACS Separator): 500 g
3. Input voltage of the thermoMACS Separator: 12 V  
Power consumption: 52 W
4. Input voltage of the power supply  
(enclosed with the thermoMACS Separator):  
100–240 VAC, ~ 50–60 Hz  
Power consumption: 52 W

▲ **Note:** Use the thermoMACS Separator only with the enclosed power supply. Use of a non-specified power supply may damage the device.

5. The thermoMACS Separator is labeled as a protection class III device.
6. Conditions of operation: 10 °C to 30 °C with 0% to 85% humidity.
7.  $\mu$  Column reaction temperatures: 37 °C, 42 °C, room temperature (heat off).
8. The thermoMACS Separator can process up to four samples in parallel.
9. Standards: The thermoMACS Separation Unit meets the intent of the EMC Directive 89/336/EEC and the Low Voltage Directive (LVD) 73/23/EEC. Compliance was demonstrated by conformance to the following specifications which have been listed in the Official Journal of the European Communities.

### thermoMACS™ Separator: Power Supply:

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**EMC:** EN 55022, Class B;  
ICE 61000-4-2  
ICE 61000-4-3  
**Safety:** EN 61010/A2

**EMC:** EN 55022, Class B  
EN 61000-3-2  
EN 61000-3-3  
EN 61000-4-2  
EN 61000-4-3  
EN 61000-4-4  
EN 61000-4-6  
EN 61000-4-11  
**Safety:** UL 1950  
CSA 22.2  
EN 60950



## **Warranty**

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