



# Genomic DNA from blood

## User manual

NucleoSpin<sup>®</sup> 8 Blood QuickPure

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**MACHEREY-NAGEL**

[www.mn-net.com](http://www.mn-net.com)



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# 1 Components

## 1.1 Kit contents

| NucleoSpin® 8 Blood QuickPure                                  |              |              |
|----------------------------------------------------------------|--------------|--------------|
| REF                                                            | 12 x 8 preps | 60 x 8 preps |
|                                                                | 740666       | 740666.5     |
| Lysis Buffer BQ1                                               | 25 mL        | 125 mL       |
| Wash Buffer BQ2<br>(Concentrate) <sup>1</sup>                  | 20 mL        | 2 x 50 mL    |
| Elution Buffer BE <sup>2</sup>                                 | 13 mL        | 60 mL        |
| Proteinase K (lyophilized) <sup>1</sup>                        | 60 mg        | 2 x 126 mg   |
| Proteinase Buffer PB                                           | 8 mL         | 15 mL        |
| NucleoSpin® Blood QuickPure<br>Binding Strips (dark red rings) | 12           | 60           |
| Rack of Tube Strips <sup>3</sup>                               | 2            | 5            |
| Cap Strips                                                     | 24           | 60           |
| Self-adhering PE Foil                                          | 12           | 60           |
| User manual                                                    | 1            | 1            |

## 1.2 Reagent to be supplied by user

- 96–100 % ethanol

For more detailed information regarding special hardware required for centrifuge or vacuum processing, please see section 2.3.

<sup>1</sup> For preparation of working solutions and storage conditions, see section 3.

<sup>2</sup> Elution Buffer BE: 5 mM Tris/HCl, pH 8.5

<sup>3</sup> Set of 1 rack, 12 strips with 8 tubes each, Cap Strips included

## 2 Product description

### 2.1 The basic principle

With the **NucleoSpin® 8 Blood QuickPure** method, genomic DNA is prepared from whole blood, cultured cells, serum, plasma, or other body fluids. Lysis is achieved by incubation of whole blood in a solution containing large amounts of chaotropic ions and Proteinase K. Appropriate conditions for binding of DNA to the silica membrane of the corresponding **NucleoSpin® Blood QuickPure Binding Strips** are created by addition of ethanol to the lysate. The binding process is reversible and specific to nucleic acids. Contaminants are efficiently washed away with only one step for washing and drying. Pure genomic DNA is finally eluted under low ionic strength conditions in a slightly alkaline elution buffer.

### 2.2 Kit specifications

- **NucleoSpin® 8 Blood QuickPure** is designed for ultra-fast small-scale purification of highly pure genomic DNA from whole blood, serum, plasma, or other body fluids.
- Blood treated either with EDTA, citrate, heparin, or CPDA can be used. If leukocyte rich materials like buffy coat are used, apply smaller volumes and dilute the samples with sterile PBS (dissolve 8 g NaCl, 0.2 g KCl, 1.44 g Na<sub>2</sub>HPO<sub>4</sub>, and 0.24 g KH<sub>2</sub>PO<sub>4</sub> in 800 mL H<sub>2</sub>O. Adjust pH to 7.4 with HCl. Add H<sub>2</sub>O to 1 liter).
- The kit allows purification of highly pure genomic DNA with an A<sub>260</sub>/A<sub>280</sub>-ratio between 1.6 and 1.9 and a typical concentration of 40–60 ng per µL.
- The obtained DNA is ready to use for subsequent reactions like PCR, Southern blotting, or any kind of enzymatic reactions.

**Table 1: Kit specifications at a glance**

| Parameters        | NucleoSpin® 8 Blood QuickPure                                 |
|-------------------|---------------------------------------------------------------|
| Technology        | Silica-membrane technology                                    |
| Format            | 8-well strips                                                 |
| Processing        | Manual, centrifugation                                        |
| Sample material   | 200 µL (default volume)<br>300 µL (support protocol)          |
| Fragment size     | 300 bp–approx. 50 kbp                                         |
| Typical DNA yield | 4–6 µg                                                        |
| $A_{260}/A_{280}$ | 1.6–1.9                                                       |
| Elution volume    | 100 µL (default volume;<br>75 µL increases DNA concentration) |
| Preparation time  | 60 min/12 strips                                              |
| Binding capacity  | 60 µg                                                         |

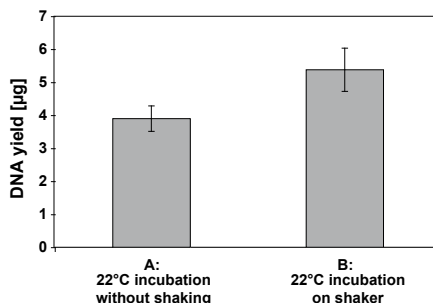
## 2.3 Required hardware

**Centrifuge:** For centrifugation, a microtiterplate centrifuge which is able to accommodate NucleoSpin® 8 Blood QuickPure Binding Strips stacked on an MN Square-well Block or Rack of Tube Strips and which reaches accelerations of  $5,600 \times g$  is required (bucket height: 85 mm).

**Shaker:** For higher convenience (ease of handling, pipette tip savings) and most consistent results (high DNA yield and purity, high yield uniformity) a 96-well plate shaker with small spinning radius and high mixing frequency is recommended ( $\sim 2\text{--}4$  mm radius;  $> 1000$  rpm; see Figure 1). Incubators with a larger spinning radius and lower speeds (e.g., bacterial culture incubators with  $\sim 19\text{--}30$  mm spinning radius and  $< 500$  rpm) are not recommended due to their inefficient mixing effect. Alternatively, lysates can be set up in 1.5 mL tubes and mixing can be performed with a vortexer.

If no shaker is available, mixing blood, Proteinase K, and Buffer BQ1 by pipetting 3–5 times up and down is highly recommended (lysis step). Pipette tips with small diameter are recommended. Do not use serological pipet tips with large outlet diameter as marginal mixing of the components may lead to incomplete blood lysis and reduced kit performance.

**Starter Set C** (see ordering information) is required for first time users of NucleoSpin® 8 Blood QuickPure kits. The Starter Set C contains Column Holders C, MN Square-well Blocks for waste collection, and Rack of Tube Strips, which serve as support strips during elution.



**Figure 1: Lysis procedure: Shaking during incubation increases yield**

200 µL frozen, human EDTA whole blood was subjected to DNA isolation with **NucleoSpin® 96 Blood QuickPure**. **A:** Blood, Proteinase K, and lysis buffer were mixed by pipetting 3x up and down. **B:** MN Square-well Block containing blood, Proteinase K, and lysis buffer were incubated on a Thermomixer™ Comfort (Eppendorf®). Median yield of DNA and average error are indicated (sample number = 32).

## 2.4 Storage of blood samples

For the isolation of genomic DNA from blood treated with anticoagulant (heparin, citrate, EDTA, CPDA) using a **NucleoSpin® 8 Blood QuickPure** kit, the blood samples can be stored at room temperature, 4 °C, or frozen.

Blood samples stored at room temperature or 4 °C for several days or weeks, will still allow DNA isolation. However, DNA yield and quality will slowly decrease due to prolonged storage of blood samples under these conditions (depending on blood sample).

Blood stored frozen for years is well suited for DNA isolation.

Highest yields and quality of DNA is obtained from fresh blood.

## 2.5 Elution procedures

It is possible to adapt the elution method and the volume of elution buffer to the subsequent application of interest. In addition to the standard method (recovery rate about 70–90 %) several modifications are possible:

- **High yield:** Perform two elution steps with the volume indicated in the individual protocol. Alternatively, heat the elution buffer to 70 °C before applying it onto the membrane. About 96–100 % of bound nucleic acid can be eluted.
- **High concentration:** Perform one elution step with 75 % of the volume indicated in the individual protocol. Concentration of DNA will be approximately 40 % higher compared to standard elution. Maximal yield of bound nucleic acid is about 80 %.

- **High yield and high concentration:** Apply half the volume of elution buffer as indicated in the individual protocol, incubate for 3 min and centrifuge. Apply a second aliquot of elution buffer, incubate, and centrifuge again. Thus, about 85–100 % of bound nucleic acid is eluted in the standard elution volume with high concentration.
- **Convenient elution:** For convenience, elution buffer may be used at room temperature (default procedure). This will result in a little lower yield (approximately 20 %) compared to elution with heated elution buffer.

Elution may also be performed with Tris-EDTA-buffer (TE) of pH 8 to 9. This will increase DNA stability, especially during long term storage at 4 °C or ambient temperature, by inhibiting DNases. However, EDTA interferes with certain downstream applications depending on the final concentration. DNA isolated with **NucleoSpin® 8 Blood QuickPure** from human EDTA whole blood, eluted in Elution Buffer BE, has been shown to be stable at 20–37 °C for several weeks without observable DNA degradation according to gel electrophoresis.

For optimal performance of isolated DNA in downstream applications, we recommend elution with the supplied elution buffer and storage at -20 °C. Several freeze-thaw cycles will not interfere with most downstream applications.

Performance of long-range PCR (e.g., > 10 kbp) or detection sensitivity of trace amounts of DNA might be reduced after multiple freeze-thaw cycles or prolonged storage of eluted DNA at 4 °C or room temperature due to degradation of DNA or adsorption to surfaces.

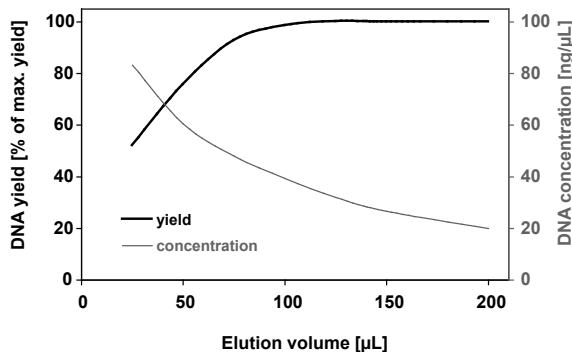


Figure 2: Influence of elution volume on DNA yield and concentration

### 3 Storage conditions and preparation of working solutions

#### Attention:

*Buffer BQ1 contains guanidine hydrochloride! Wear gloves and goggles when handling them!*

*CAUTION: Buffers BQ1 contains guanidine hydrochloride which can form highly reactive compounds when combined with bleach (sodium hypochlorite). DO NOT add bleach or acidic solutions directly to the sample-preparation waste.*

#### Storage conditions:

- All components of the **NucleoSpin® 8 Blood QuickPure** kit should be stored at room temperature and are stable for at least one year.
- Upon storage at lower temperature, a white precipitate may form in Buffer BQ1. Dissolve such precipitates by incubation of the bottle at 70 °C before use.

Before starting with any **NucleoSpin® 8 Blood QuickPure** kit procedure, prepare the following:

- **Wash Buffer BQ2:** Add the indicated volume of 96–100% ethanol to the **Buffer BQ2 Concentrate**. Mark the label of the bottle to indicate that ethanol is added. Store Wash Buffer BQ2 at room temperature (18–25 °C) for up to one year.
- Before first use of the kit, add the indicated volume of **Proteinase Buffer PB** to lyophilized **Proteinase K**. Proteinase K solution is stable at -20 °C for at least 6 months.

| NucleoSpin® 8 Blood QuickPure    |                                       |                                                            |
|----------------------------------|---------------------------------------|------------------------------------------------------------|
| REF                              | 12 x 8 preps                          | 60 x 8 preps                                               |
|                                  | 740666                                | 740666.5                                                   |
| Wash Buffer BQ2<br>(Concentrate) | 20 mL<br>Add 80 mL ethanol            | 2 x 50 mL<br>Add 200 mL ethanol<br>to each bottle          |
| Proteinase K<br>(lyophilized)    | 60 mg<br>Add 2.7 mL Proteinase Buffer | 2 x 126 mg<br>Add 6.6 mL Proteinase Buffer<br>to each vial |



## 4 Safety instructions

The following components of the **NucleoSpin® 8 Blood QuickPure** kit contain hazardous contents.

*Wear gloves and goggles and follow the safety instructions given in this section.*

### GHS classification

Only harmful features need not be labeled with H and P phrases up to 125 mL or 125 g.

*Mindergefährliche Eigenschaften müssen bis 125 mL oder 125 g nicht mit H- und P-Sätzen gekennzeichnet werden.*

| Component    | Hazard contents                                                           | GHS symbol                                                                                                                                                                                     | Hazard phrases     | Precaution phrases                                                               |
|--------------|---------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------|----------------------------------------------------------------------------------|
| Inhalt       | Gefahrstoff                                                               | GHS Symbol                                                                                                                                                                                     | H-Sätze            | P-Sätze                                                                          |
| BQ1          | Guanidine hydrochloride<br>50–66 %<br><i>Guanidinhydrochlorid 50–66 %</i> |  Warning<br><i>Achtung</i>                                                                                    | 302, 315, 319      | 280, 301+312, 302+352, 305+351+338, 330, 332+313, 337+313                        |
| Proteinase K | Proteinase K, lyophilized<br><i>Proteinase K, lyophilisiert</i>           | <br> Danger<br><i>Gefahr</i> | 315, 319, 334, 335 | 261, 280, 302+352, 304+340, 305+351+338, 312, 332+313, 337+313, 342+311, 403+233 |

### Hazard phrases

|       |                                                                                                                                                                          |
|-------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| H 302 | Harmful if swallowed.<br><i>Gesundheitsschädlich bei Verschlucken.</i>                                                                                                   |
| H 315 | Causes skin irritation.<br><i>Verursacht Hautreizungen.</i>                                                                                                              |
| H 319 | Causes serious eye irritation.<br><i>Verursacht schwere Augenreizung.</i>                                                                                                |
| H 334 | May cause allergy or asthma symptoms or breathing difficulties if inhaled.<br><i>Kann bei Einatmen Allergie, asthmaartige Symptome oder Atembeschwerden verursachen.</i> |
| H 335 | May cause respiratory irritation.<br><i>Kann die Atemwege reizen.</i>                                                                                                    |

### Precaution phrases

|           |                                                                                                                                                            |
|-----------|------------------------------------------------------------------------------------------------------------------------------------------------------------|
| P 261     | Avoid breathing dust.<br><i>Einatmen von Staub vermeiden.</i>                                                                                              |
| P 280     | Wear protective gloves / eye protection.<br><i>Schutzhandschuhe / Augenschutz tragen.</i>                                                                  |
| P 301+312 | IF SWALLOWED: Call a POISON CENTER/ doctor/.../if you feel unwell.<br><i>BEI VERSCHLUCKEN: Bei Unwohlsein GIFTINFORMATIONSZENTRUM / Arzt /... anrufen.</i> |

### Precaution phrases

- P 302+352 IF ON SKIN: Wash with plenty of water/...  
*BEI KONTAKT MIT DER HAUT: Mit viel Wasser/... waschen.*
- P 304+340 IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing.  
*BEI EINATMEN: An die frische Luft bringen und in einer Position ruhigstellen, die das Atmen erleichtert.*
- P 305+351+338 IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do – continue rinsing.  
*BEI KONTAKT MIT DEN AUGEN: Einige Minuten lang behutsam mit Wasser spülen. Vorhandene Kontaktlinsen nach Möglichkeit entfernen. Weiter spülen.*
- P 312 Call a POISON CENTER/ doctor/.../if you feel unwell.  
*Bei Unwohlsein GIFTINFORMATIONSZENTRUM / Arzt /... anrufen.*
- P 330 Rinse mouth.  
*Mund ausspülen.*
- P 332+313 If skin irritation occurs: Get medical advice / attention.  
*Bei Hautreizung: Ärztlichen Rat einholen / ärztliche Hilfe hinzuziehen.*
- P 337+313 Get medical advice / attention.  
*Bei anhaltender Augenreizung: Ärztlichen Rat einholen / ärztliche Hilfe hinzuziehen.*
- P 342+311 If experiencing respiratory symptoms: Call a POISON CENTER/ doctor/...  
*Bei Symptomen der Atemwege: GIFTINFORMATIONSZENTRUM /Arzt/... anrufen.*
- P 403+233 Store in a well ventilated place. Keep container tightly closed.  
*Behälter dicht geschlossen an einem gut belüfteten Ort aufbewahren.*

For further information please see Material Safety Data Sheets ([www.mn-net.com](http://www.mn-net.com)).  
Weiterführende Informationen finden Sie in den Sicherheitsdatenblättern ([www.mn-net.com](http://www.mn-net.com)).

## 5 Protocols

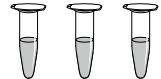
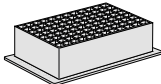
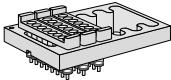
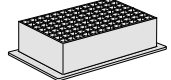
### 5.1 NucleoSpin® 8 Blood QuickPure – standard procedure

- For hardware requirements, refer to section 2.3.
- For detailed information on each step, see page 15.

#### Before starting the preparation:

- Check if Buffer BQ2 and Proteinase K were prepared according to section 3.

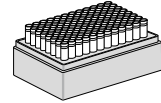
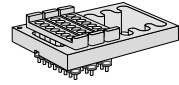
### Protocol-at-a-glance

|                                        |                                                                                                                                                                 |                                                                                                                                                                               |
|----------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>1 Lyse samples</b>                  | <b>200 µL blood</b><br><br><b>25 µL Proteinase K</b><br><br><b>200 µL BQ1</b><br><br><b>Incubate on shaker</b><br><b>(1.200–1.500 rpm)</b><br><b>RT, 10 min</b> | <br>or<br>  |
| <b>2 Adjust DNA binding conditions</b> | <b>200 µL ethanol</b><br><br><b>Mix</b>                                                                                                                         | e.g., reaction tubes,<br>MN Square-well Block                                                                                                                                 |
| <b>3 Bind DNA to silica membrane</b>   | <b>Transfer samples to</b><br><b>NucleoSpin® Blood QuickPure</b><br><b>Binding Strips</b><br><br><b>5,600 x g,</b><br><b>3 min</b>                              | <br><br> |
| <b>4 Wash and dry silica membrane</b>  | <b>500 µL BQ2</b><br><br><b>5,600 x g,</b><br><b>5 min</b>                                                                                                      | Column Holder C with<br>NucleoSpin® Blood<br>QuickPure Binding Strips on<br>MN Square-well Block                                                                              |

**5 Elute DNA**

**100 µL BE**

**5,600 x g,  
2 min**



Column Holder C with  
NucleoSpin® Blood  
QuickPure Binding Strips on  
Rack of Tube Strips

---

## Detailed protocol

- For hardware requirements, refer to section 2.3.

### Before starting the preparation:

- Check if Wash Buffer BQ2 and Proteinase K are prepared according to section 3.

---

## 1 Lyse samples

Pipette **25 µL Proteinase K** and **200 µL blood**, buffy coat, or body fluid sample (equilibrated to room temperature) into a 1.5 or 2.0 mL reaction tube (not supplied) or MN Square-well Block (not supplied).

*For sample volumes less than 200 µL, add PBS to adjust the volume to 200 µL. If purifying DNA viruses, we recommend starting with 200 µL serum or plasma. If cultured cells are used, resuspend up to  $5 \times 10^6$  cells in a final volume of 200 µL PBS.*

Add **200 µL Buffer BQ1** to each sample. Close reaction tubes.

Incubate reaction tubes at ambient temperature (18–25 °C) for **10 min** on a **shaker** at high shaking speed (1,200–1,500 rpm).

*See section 2.3 for recommended shaker. If no shaker is available, pipette the lysate up and down 3–5 times to ensure thorough mixing of the solution (also read recommendations in section 2.3). The lysate will turn brownish during incubation with Buffer BQ1 and Proteinase K. Increase incubation time with Proteinase K (up to 30 min) if processing older or clotted blood samples.*

---

## 2 Adjust DNA binding conditions

Add **200 µL ethanol (96–100 %)** to each reaction tube. Mix solution by pipetting up and down three times.

*Note: Upon addition of ethanol to the lysate, mix them immediately (within approximately 1 min). Do not let the ethanol float on top of the lysate for an extended time (> 1 min). Extended ethanol floating before mixing can reduce DNA yield and purity.*

*Short incubation on a shaker is usually not sufficient for thorough mixing of the less dense and thus floating ethanol into the blood lysate. Always mix solutions by pipetting up and down.*

### 3 Bind DNA to silica membrane

Insert an appropriate number of **NucleoSpin® Blood QuickPure Binding Strips** into the Column Holder C and place the assembly onto an MN Square-well Block.

**Transfer lysates** into the wells of the NucleoSpin® Blood QuickPure Binding Strips. Cover all strips with Self-adhering PE Foil.

Centrifuge **3 min** at **5,600 x g**.

*Column Holder C and reusable MN Square-well Blocks are provided in the Starter Set C. If the samples are not drawn through the matrix completely, repeat the centrifugation for a 10 min period. It is not necessary to discard the flow-through.*

---

### 4 Wash and dry silica membrane

Remove Self-adhering PE Foil and add **500 µL Buffer BQ2** to each well of the strips. Cover all strips with new Self-adhering PE Foil.

Centrifuge **5 min** at **5,600 x g**.

*Optional: Repeat washing step, especially if difficult blood samples (old or clotted) are processed or if extremely demanding downstream applications are performed. Make sure to remove flow-through of the first washing step if applying this optional washing step.*

*The membrane is washed and residual ethanol is removed during this step. Discard flow-through, clean and sterilize MN Square-well Block for reuse as flow-through collecting device.*

---

### 5 Elute DNA

Place NucleoSpin® Blood QuickPure assembly onto a Rack of Tube Strips. The rack must be completely filled with 8-well strips to ensure stable support of the column holder.

Remove Self-adhering PE Foil and add **100 µL Buffer BE** to each well. Cover all strips with new Self-adhering PE Foil.

Centrifuge **2 min** at **5,600 x g**.

Seal Tube Strips with Cap Strips for DNA storage.

*For alternative elution procedures, see section 2.5. If elution in small volume tubes is desired, place a 96 PCR plate (not supplied) on top of the Rack of Tube Strips and elute into the PCR plate.*

---

## 5.2 NucleoSpin® 8 Blood QuickPure – protocol for 300 µL samples

This protocol allows DNA purification from 300 µL samples without additional loading steps.

- For hardware requirements, refer to section 2.3.

### Before starting the preparation:

- Check if Wash Buffer BQ2 and Proteinase K were prepared according to section 3.
- Additional Buffer BQ1 and Proteinase K is necessary. Please see ordering information.

---

### 1 Lyse samples

Pipette **37 µL Proteinase K** and **300 µL blood** or body fluid sample (equilibrated to room temperature) into a 1.5 or 2.0 mL reaction tube (not supplied) or MN Square-well Block (not supplied).

Add **300 µL Buffer BQ1** to each sample. Cover MN Square-well Block with Self-adhering PE Foil, close reaction tube (whatever applicable).

Incubate reaction tubes at ambient temperature (18–25 °C) for **10 min** on a **shaker** at high shaking speed.

*See section 2.3 for recommended shaker and for further information if no shaker is available. The lysate will turn brownish during incubation with Buffer BQ1 and Proteinase K. Increase incubation time with Proteinase K (up to 30 min) if processing older or clotted blood samples.*

---

### 2 Adjust DNA binding conditions

Add **300 µL ethanol (96–100 %)** to each 1.5 or 2.0 mL reaction tube. Mix solution by pipetting up and down 3 times.

***Note:** Upon addition of ethanol to the lysate, mix them immediately (within approximately 1 min). Do not let the ethanol float on top of the lysate for an extended time (> 1 min). Extended ethanol floating before mixing can reduce DNA yield and purity.*

*Short incubation on a shaker is usually not sufficient for thorough mixing of the less dense and thus floating ethanol into the blood lysate. Always mix solutions by pipetting up and down.*

---

Continue procedure with step 3 (Bind DNA to silica membrane, see section 5.1) of the NucleoSpin® 8 Blood QuickPure standard protocol. The lysate can be loaded in one step.

*Due to the superior properties of the binding matrix and effective lysis 300 µL samples may be routinely processed with very low risk of membrane clogging compared to competitor products.*

---



## 6 Appendix

### 6.1 Troubleshooting

| Problem              | Possible cause and suggestions                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
|----------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| No or poor DNA yield | <i>Low concentration of leukocytes in sample</i> <ul style="list-style-type: none"><li>• Prepare buffy coat from the blood sample: Centrifuge whole blood containing anticoagulant at room temperature (3,300 x g; 10 min). Three different layers will be visible after centrifugation. Leukocytes are concentrated in the intermediate layer (= buffy coat).</li></ul>                                                                                                                                                                                                                                                                                                                                               |
|                      | <i>Incomplete cell lysis</i> <ul style="list-style-type: none"><li>• Inhomogeneous blood sample or blood clots within the sample: Make sure that blood samples are collected following the instructions of the manufacturer of the blood collection tube. Make sure that only blood is used as sample material which can easily be transferred by pipetting. If necessary, homogenize the blood sample before use.</li><li>• Sample not thoroughly mixed with lysis buffer/Proteinase K. The three components have to be mixed vigorously after addition of lysis buffer.</li><li>• Proteinase K digestion not optimal. Never add Proteinase K directly to lysis buffer. Increase incubation time to 30 min.</li></ul> |
|                      | <i>Reagents not properly applied</i> <ul style="list-style-type: none"><li>• Prepare buffers and Proteinase K solution according to instructions (section 3). Add ethanol to lysate before loading onto the columns.</li></ul>                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
|                      | <i>Suboptimal elution of DNA from the column</i> <ul style="list-style-type: none"><li>• Elution efficiencies decrease significantly if elution is performed with buffers of pH &lt; 7. Use slightly alkaline elution buffer like Buffer BE (pH 8.5).</li></ul>                                                                                                                                                                                                                                                                                                                                                                                                                                                        |
|                      | <i>Too long resting time of ethanol floating on top of the lysate</i> <ul style="list-style-type: none"><li>• Upon addition of ethanol to the lysate, mix immediately. Do not let the ethanol float on top of the lysate for longer than ~ 1 min.</li></ul>                                                                                                                                                                                                                                                                                                                                                                                                                                                            |
|                      |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        |

| Problem                                                      | Possible cause and suggestions                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                   |
|--------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Poor DNA quality                                             | <p><i>Reagents not properly applied</i></p> <ul style="list-style-type: none"> <li>• Prepare buffers and Proteinase K solution according to instructions (section 3). Add ethanol to lysate and mix thoroughly before loading onto the columns.</li> </ul>                                                                                                                                                                                                                                                                                                       |
|                                                              | <p><i>Incomplete cell lysis</i></p> <ul style="list-style-type: none"> <li>• Sample not thoroughly mixed with lysis buffer/Proteinase K. The three components have to be mixed vigorously after addition of lysis buffer.</li> <li>• Proteinase K digestion not optimal. Do not add Proteinase K directly to lysis buffer. Increase incubation time to 30 min.</li> </ul>                                                                                                                                                                                        |
|                                                              | <p><i>RNA in sample</i></p> <ul style="list-style-type: none"> <li>• If DNA free of RNA is desired, add 20 µL of an RNase A solution (20 mg/mL) before adding the lysis buffer.</li> </ul>                                                                                                                                                                                                                                                                                                                                                                       |
|                                                              | <p><i>Old or clotted blood samples processed</i></p> <ul style="list-style-type: none"> <li>• For isolation of DNA from older or clotted blood samples, we recommend extending the Proteinase K incubation to 30 min.</li> </ul>                                                                                                                                                                                                                                                                                                                                 |
| Suboptimal performance of genomic DNA in enzymatic reactions | <p><i>Carry-over of ethanol</i></p> <ul style="list-style-type: none"> <li>• Be sure to remove all ethanolic Buffer BQ2 before eluting the DNA. If the level of Buffer BQ2 after the wash has reached the column outlet for any reason, discard flow-through, place the column holder with NucleoSpin® 8 QuickPure Binding Strips back onto the collecting MN Square-well Block, and centrifuge again.</li> </ul>                                                                                                                                                |
|                                                              | <p><i>Contamination of DNA with inhibitory substances</i></p> <ul style="list-style-type: none"> <li>• If DNA has been eluted with Tris/EDTA-buffer (TE), make sure that EDTA does not interfere with downstream applications or repurify DNA and elute in Buffer BE.</li> <li>• If preparing DNA from older or clotted blood samples, extend Proteinase K incubation to 30 min.</li> <li>• If the <math>A_{260}/A_{280}</math>-ratio of the eluate is below 1.6, repeat the purification procedure or apply the optional second wash step next time.</li> </ul> |

| Problem                 | Possible cause and suggestions                                                                                                                                                                                                                                   |
|-------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| High variation in yield | <i>Possible reasons</i>                                                                                                                                                                                                                                          |
|                         | <ul style="list-style-type: none"><li>• DNA-isolation from 96 different and independent samples (e.g., blood samples from 96 different persons) will result in higher yield variation (measured and actual) than from 96 aliquots of one blood sample.</li></ul> |
|                         | <ul style="list-style-type: none"><li>• Elution with smaller volumes (e.g., 50 µL vs. 150 µL) will result in larger (measured and actual) yield variation compared to elution in larger volumes.</li></ul>                                                       |
|                         | <ul style="list-style-type: none"><li>• If only a few single wells are used as global blank for a complete 96-well plate measurement this will likely cause larger <u>measured</u> yield variation than well to well blank correction measurements.</li></ul>    |

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## 6.2 Ordering information

| Product                                                                                                 | REF                               | Pack of                                       |
|---------------------------------------------------------------------------------------------------------|-----------------------------------|-----------------------------------------------|
| NucleoSpin® 8 Blood QuickPure                                                                           | 740666<br>740666.5                | 12 x 8 preps<br>60 x 8 preps                  |
| NucleoSpin® 96 Blood QuickPure                                                                          | 740667.2<br>740667.4<br>740667.24 | 2 x 96 preps<br>4 x 96 preps<br>24 x 96 preps |
| Buffer BQ1                                                                                              | 740923                            | 125 mL                                        |
| Proteinase K                                                                                            | 740506                            | 100 mg                                        |
| RNase A                                                                                                 | 740505                            | 100 mg                                        |
| Rack of Tube Strips<br>(set consists of 1 Rack, 12 Tube Strips<br>with 8 tubes each, and 12 Cap Strips) | 740477<br>740477.24               | 4 sets<br>24 sets                             |
| MN Square-well Block                                                                                    | 740476<br>740476.24               | 4 sets<br>24 sets                             |
| Round-well Block, Low                                                                                   | 740487<br>740487.24               | 4<br>24                                       |
| Starter Set C<br>(for use of 8-well strips under<br>centrifugation)                                     | 740684                            | 1 set                                         |
| Self-adhering PE Foil                                                                                   | 740676                            | 50                                            |
| NucleoCard®                                                                                             | 740403.10<br>740403.100           | 10 cards<br>100 cards                         |

Visit **[www.mn-net.com](http://www.mn-net.com)** for more detailed product information.

## 6.3 Product use restriction/warranty

**NucleoSpin® 8 Blood QuickPure** components are intended, developed, designed, and sold FOR RESEARCH PURPOSES ONLY, except, however, any other function of the product being expressly described in original MACHEREY-NAGEL product leaflets.

MACHEREY-NAGEL products are intended for GENERAL LABORATORY USE ONLY! MACHEREY-NAGEL products are suited for QUALIFIED PERSONNEL ONLY! MACHEREY-NAGEL products shall in any event only be used wearing adequate PROTECTIVE CLOTHING. For detailed information please refer to the respective Material Safety Data Sheet of the product! MACHEREY-NAGEL products shall exclusively be used in an ADEQUATE TEST ENVIRONMENT. MACHEREY-NAGEL does not assume any responsibility for damages due to improper application of our products in other fields of application. Application on the human body is STRICTLY FORBIDDEN. The respective user is liable for any and all damages resulting from such application.

DNA/RNA/PROTEIN purification products of MACHEREY-NAGEL are suitable for *IN-VITRO*-USES ONLY!

ONLY MACHEREY-NAGEL products specially labeled as IVD are also suitable for *IN-VITRO*-diagnostic use. Please pay attention to the package of the product. *IN-VITRO*-diagnostic products are expressly marked as IVD on the packaging.

IF THERE IS NO IVD SIGN, THE PRODUCT SHALL NOT BE SUITABLE FOR *IN-VITRO*-DIAGNOSTIC USE!

ALL OTHER PRODUCTS NOT LABELED AS IVD ARE NOT SUITED FOR ANY CLINICAL USE (INCLUDING, BUT NOT LIMITED TO DIAGNOSTIC, THERAPEUTIC AND/OR PROGNOSTIC USE).

No claim or representations is intended for its use to identify any specific organism or for clinical use (included, but not limited to diagnostic, prognostic, therapeutic, or blood banking). It is rather in the responsibility of the user or - in any case of resale of the products - in the responsibility of the reseller to inspect and assure the use of the DNA/RNA/protein purification products of MACHEREY-NAGEL for a well-defined and specific application.

MACHEREY-NAGEL shall only be responsible for the product specifications and the performance range of MN products according to the specifications of in-house quality control, product documentation and marketing material.

This MACHEREY-NAGEL product is shipped with documentation stating specifications and other technical information. MACHEREY-NAGEL warrants to meet the stated specifications. MACHEREY-NAGEL's sole obligation and the customer's sole remedy is limited to replacement of products free of charge in the event products fail to perform as warranted. Supplementary reference is made to the general business terms and conditions of MACHEREY-NAGEL, which are printed on the price list. Please contact us if you wish to get an extra copy.

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