



DNA from food

User manual NucleoMag[®] DNA Food

January 2020 / Rev. 02

MACHEREY-NAGEL

www.mn-net.com



DOMINIQUE DUTSCHER SAS

Contact MN

Germany and international

MACHEREY-NAGEL GmbH & Co. KG

Neumann-Neander-Str. 6-8 · 52355 Düren · Germany

Tel.: +49 24 21 969-0

Toll-free: 0800 26 16 000 (Germany only)

Fax: +49 24 21 969-199

E-mail: info@mn-net.com

Technical Support Bioanalysis

Tel.: +49 24 21 969-270

E-mail: tech-bio@mn-net.com

USA

MACHEREY-NAGEL Inc.

2850 Emrick Blvd. · Bethlehem, PA 18020 · USA

Tel.: +1 484 821 0984

Toll-free: 888 321 6224 (MACH)

Fax: +1 484 821 1272

E-mail: sales-us@mn-net.com

France

MACHEREY-NAGEL SARL à associé unique

1, rue Gutenberg · 67722 Hoerd · France

Tel.: +33 388 68 22 68

Fax: +33 388 51 76 88

E-mail: sales-fr@mn-net.com

Switzerland

MACHEREY-NAGEL AG

Hirsackerstr. 7 · 4702 Oensingen · Switzerland

Tel.: +41 62 388 55 00

Fax: +41 62 388 55 05

E-mail: sales-ch@mn-net.com

www.mn-net.com

DOMINIQUE DUTSCHER SAS

Table of contents

1 Components	4
1.1 Kit contents	4
1.2 Reagents, consumables and equipment to be supplied by user	4
1.3 About this user manual	5
2 Product description	6
2.1 The basic principle	6
2.2 Kit specifications	6
2.3 Magnetic separation systems	6
2.4 Adjusting the shaker settings	7
2.5 Handling of beads	8
2.6 Elution procedures	8
3 Storage conditions and preparation of working solutions	9
4 Safety instructions	10
5 General remarks	12
5.1 Important information and advice	12
6 Protocol for the isolation of DNA from food samples	14
6.1 Protocol at a glance	14
6.2 Detailed protocol	16
7 Appendix	19
7.1 Troubleshooting	19
7.2 Ordering information	21
7.3 Product use restriction / warranty	22

1 Components

1.1 Kit contents

NucleoMag® DNA Food		
REF	1 x 96 preps 744945.1	4 x 96 preps 744945.4
NucleoMag® B-Beads	2 x 1.5 mL	12 mL
Lysis Buffer CF	100 mL	500 mL
Binding Buffer CB	100 mL	300 mL
Wash Buffer CMW	100 mL	300 mL
Wash Buffer CQW	125 mL	2 x 250 mL
Elution Buffer CE	30 mL	125 mL
Liquid Proteinase K	1 x 1250 µl	4,5 mL
User manual	1	1

1.2 Reagents, consumables and equipment to be supplied by user

Reagents

- 80 % Ethanol
- Personal protection equipment (e.g., lab coat, gloves, goggles)

Product	REF	Pack of
Magnet for magnetic beads separation e.g. NucleoMag® SEP	744900	1
NucleoMag® SEP Mini	744901	1
NucleoMag® SEP Maxi	744902	1
NucleoMag® SEP 24	744903	1
Separation plate for magnetic beads separation, e.g. Square-well Block (96-well block with 2.1 mL square-wells)	740481 740481.24	4 24
Lysis tubes for incubation of samples and lysis, e.g., Rack of Tubes Strips (1 set consists of 1 Rack, 12 Strips with 8 tubes (1.2 mL wells) each, and 12 Cap Strips)	740477 740477.24	4 sets 24 sets

Product	REF	Pack of
Elution plate for collecting purified nucleic acids, e.g., Elution Plate U-bottom (96-well 0.3 mL microtiterplate with 300 µL u-bottom wells)	740486.24	24
For use of kit on KingFisher® instruments: e.g., KingFisher® 96 Accessory Kit A (Square- well Blocks, Deep-well tip combs, Plates for 4 x 96 NucleoMag® DNA Food preps using KingFisher® 96/Flex platform)	744950	1 set
KingFisher® DUO Accessory Kit (Square-well Blocks, Deep-well tip combs, Plates for 8 x 12 NucleoMag® DNA Food preps using KingFisher® DUO/DUO Prime platform.	744952	1 set

1.3 About this user manual

It is strongly recommended that first time users of the NucleoMag® DNA Food kit read the detailed protocol sections of this user manual. Experienced users, however, may refer to the Protocol at a glance instead. The Protocol at a glance is designed to be used as a tool for quick referencing while performing the purification procedure.

All technical literature is available at www.mn-net.com.

Please contact Technical Service regarding information about changes of the current user manual compared to previous revisions.

2 Product description

2.1 The basic principle

The **NucleoMag® DNA Food** kit is designed for the isolation of genomic DNA from food and feed samples. The procedure is based on reversible adsorption of nucleic acids to paramagnetic beads under appropriate buffer conditions. Sample lysis is achieved by incubation of samples with Proteinase K at 65 °C. Lysis mixtures should be cleared by centrifugation or filtration in order to remove contaminants and residual cellular debris. For binding of nucleic acids to the paramagnetic beads, Binding Buffer CB and the NucleoMag® B-Beads are added to the clear lysate. After magnetic separation, the paramagnetic beads are washed to remove contaminants and salts using Wash Buffers CMW, CQW and 80% ethanol. Residual ethanol from previous wash steps is removed by air drying. Next, highly purified DNA is eluted with low-salt Elution Buffer (CE) and can be used directly for downstream applications. The **NucleoMag® DNA Food** kit can be used either manually, or automated on standard liquid handling instruments and automated magnetic separators.

2.2 Kit specifications

- **NucleoMag® DNA Food** is designed for rapid manual and automated small scale preparation of DNA from food or feed samples. For the isolation of DNA from bacteria within the food or feed samples please follow the recommendations in section 5.1 page 12.
- **The NucleoMag® DNA Food** kit can be used for the identification of GMO DNA or animal components in food and feed.
- The kit provides reagents for the purification of up to 200 mg material. Depending on the individual sample, typical yields for NucleoMag® DNA Food are in the range of 0.1–10 µg DNA.
- The eluted DNA is ready to use for subsequent reactions like real-time PCR, GMO detection, etc.
- **The NucleoMag® DNA Food** kit is designed for use with NucleoMag® SEP magnetic separator plate (see ordering information) or any other magnetic separation systems (see section 2.3 page 6). Manual time for the preparation of 96 samples is about 120 minutes.
- **NucleoMag® DNA Food** allows easy automation on common liquid handling instruments or automated magnetic separators. The actual processing time depends on the configuration of the instrument and the magnetic separation system used. Typically, 96 samples can be purified in less than 120 minutes using the NucleoMag® SEP on an automation platform. For more information about the automation process and the availability of ready to run scripts for certain platforms, please contact your local distributor or MN directly.

2.3 Magnetic separation systems

For use of **NucleoMag® DNA Food**, the use of the magnetic separator NucleoMag® SEP is recommended. Separation is carried out in a Square-well Block (see ordering information). The kit can also be used with other common separators.

Magnetic separator	Separation plate or tube
NucleoMag® SEP (MN REF 744900)	Square-well Block (MN REF 740481)
NucleoMag® SEP Mini (MN REF 744901)	1.5 mL or 2 mL reaction tubes (Sarstedt)
NucleoMag® SEP Maxi (MN REF 744902)	50 mL tubes (Falcon)
NucleoMag® SEP 24 (MN REF 744903)	24-Square-well Block U-bottom (MN REF 740448.4/.24)
Tecan Te-MagS™	1.5 mL tubes without lid (Sarstedt)

Static magnetic pins

Separators with static magnetic pins, for example, NucleoMag® SEP or other common magnetic separators are suitable for manual use and for use on liquid handling workstations: This type of separator is recommended in combination with a suitable microplate shaker for optimal resuspension of the beads during the washing and elution steps. Alternatively, beads can be resuspended in the buffer by pipetting up and down several times. For fully automated use on liquid handling workstations, a gripper tool is required; the plate is transferred to the magnetic separator for separation of the beads and transferred to the shaker module for resuspension of the beads.

Movable magnetic systems

Separators with moving magnetic pins: Magnetic pins/rods are moved from one side of the well to the other and vice versa. Beads follow this movement and are thus pulled through the buffer during the wash and elution steps. Separation takes place when the system stops.

Automated separators

Separators with moving magnets: Magnetic beads are transferred into suitable plates or tubes. Beads are resuspended from the rod-covered magnets. Following binding, washing or elution beads are collected again with the rod-covered magnets and transferred to the next plate or tube.

2.4 Adjusting the shaker settings

When using a plate shaker for the washing and elution steps, the speed settings have to be adjusted carefully for each specific separation plate and shaker to prevent cross contamination from well to well. Proceed as follows:

Adjusting shaker speed for binding and wash steps:

- Load 600 µL dyed water to the wells of the separation plate. Place the plate on the shaker and start shaking with a moderate speed setting for 30 seconds. Turn off the shaker and check the plate surface for small droplets of dyed water.
- Increase speed setting, shake for an additional 30 seconds, and check the plate surface for droplets again.
- Continue increasing the speed setting until you observe droplets on top of the separation plate. Reduce speed setting, check again, and use this setting for the washing step.

Adjusting shaker speed for the elution step:

- Load 100–200 μL dyed water to the wells of the collection plate and proceed as described above.

2.5 Handling of beads**Distribution of beads**

A homogeneous distribution of the magnetic beads to the individual wells of the separation plate is essential for a high well-to-well consistency. Therefore, before distributing the beads, make sure that the beads are completely resuspended. Shake the storage bottle well or place it on a vortexer shortly. During automation, a premix step before aspirating the beads from the reservoir is recommended to keep the beads resuspended.

Magnetic separation time

Attraction of the magnetic beads to the magnetic pins depends on the magnetic strength of the magnetic pins, the selected separation plate, distance of the separation plate from the magnetic pins, and the volume to be processed. The individual times for complete attraction of the beads to the magnetic pins should be checked and adjusted on each system. It is recommended using the separation plates or tubes specified by the supplier of the magnetic separator.

Washing the beads

Washing the beads can be achieved by shaking or mixing. In contrast to mixing by pipetting up and down, mixing by shaker or magnetic mixing allows simultaneous mixing of all samples. This reduces the time and number of tips needed for the preparation. Resuspension by pipetting up and down, however, is more efficient than mixing by a shaker or magnetic mix.

Method	Resuspension efficiency	Speed	Small elution volume possible	Number of tips needed
Magnetic mix	+	++	+	Low
Shaker	++	++	+++	Low
Pipetting	+++	+*	++	High

+: acceptable, ++: good, +++: excellent, * 8-channel pipetting device

2.6 Elution procedures

Purified DNA can be eluted directly with the supplied Elution Buffer CE. Elution can be carried out in a volume of $\geq 50 \mu\text{L}$. It is essential to cover the NucleoMag® B-Beads completely with elution buffer during the elution step. The volume of dispensed elution buffer depends on the magnetic separation system (e.g., the position of the pellet inside the separation plate). For efficient elution, the magnetic bead pellet should be resuspended completely in the elution buffer. For some separators, higher elution volumes might be necessary to cover the whole pellet.

3 Storage conditions and preparation of working solutions

Attention: Buffers CB, CMW and CQW contain chaotropic salt! Wear gloves and goggles!

All components of the **NucleoMag® DNA Food** kit should be stored at room temperature (18–25 °C) and are stable for at least one year.

All buffers are delivered ready to use.







4 Safety instructions

The following components of the **NucleoMag® DNA Food** kits contain hazardous contents.

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

GHS classification

Only harmful features do not need to 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
CB	2-propanol 35–50 % 2-propanol 35–50 % CAS 67-63-0	  WARNING ACHTUNG	226, 319, 336	210, 260D, 280sh
CMW	Guanidine hydrochloride 24–36 % and Ethanol 35–55 % Guanidinhydrochlorid 24–36 % und Ethanol 35–55 % CAS 50-01-1, 64-17-5	  WARNING ACHTUNG	226, 302	210, 264W, 301+312, 330
CQW	Guanidine hydrochloride 24–36 % and Ethanol 35–55 % Guanidinhydrochlorid 24–36 % und Ethanol 35–55 % CAS 50-01-1, 64-17-5	  WARNING ACHTUNG	226, 302	210, 264W, 301+312, 330

Hazard phrases

H226	Flammable liquid and vapour. <i>Flüssigkeit und Dampf entzündbar.</i>
H302	Harmful if swallowed. <i>Gesundheitsschädlich bei Verschlucken.</i>
H 319	Causes serious eye irritation. <i>Verursacht schwere Augenreizung.</i>
H 336	May cause drowsiness or dizziness. <i>Kann Schläfrigkeit und Benommenheit verursachen.</i>

Precaution phrases

- P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
Von Hitze, heißen Oberflächen, Funken, offenen Flammen sowie anderen Zündquellenarten fernhalten. Nicht rauchen.
- P260D Do not breathe vapours.
Dampf nicht einatmen.
- P264W Wash with water thoroughly after handling.
Nach Gebrauch mit Wasser gründlich waschen.
- P280sh Wear protective gloves/eye protection.
Schutzhandschuhe/Augenschutz tragen.
- P301+312 IF SWALLOWED: Call a POISON CENTER/ doctor if you feel unwell.
BEI VERSCHLUCKEN: Bei Unwohlsein GIFTINFORMATIONSZENTRUM/Arzt anrufen.
- P330 Rinse mouth.
Mund ausspülen.

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

The symbol shown on labels refers to further safety information in this section.
Das auf Etiketten dargestellte Symbol weist auf weitere Sicherheitsinformationen dieses Kapitels hin.

5 General remarks

5.1 Important information and advice

- Due to the low DNA content in processed food, this protocol should be started with up to 0.2 g of material.
- Lysis buffer was tested (see list on the next page) for extraction of DNA from various types of samples including food of plant or animal origin, and bacteria. To detect bacterial DNA in food samples, we recommend an overnight pre-culture of the sample in an appropriate culture medium. Centrifuge an aliquot of the culture and start the preparation with the bacterial pellet.
- RNase A (not included in the kit, see ordering information) addition may be recommended for RNA-rich samples. Add 10 μL (20 mg/mL stock solution) per 550 μL lysis buffer in step 2 of the protocol or perform an RNase A digestion in the eluate before further use.
- Ketchup, sauce, and similar fluid samples (0.2 g equivalents) can be mixed with lysis buffer (500–1000 μL each) and incubated with Liquid Proteinase K as described in the protocol (see ordering information for additional Lysis Buffer CF).
- For powdered hygroscopic samples, more lysis buffer than indicated in the protocol can be used until the lysis solution is at least semi fluid and can be pipetted (see ordering information for Lysis Buffer CF). Extraction can be improved by preincubation of sample with lysis buffer for 1–2 h.
- According to local law regulations, different amounts of sample have to be analyzed for GMO detection, for example, up to 1–2 g of sample can be used with upscaled lysis buffer volumes. We recommend using a single aliquot of up to 400 μL (step 3) of the clear supernatant for further processing with the NucleoMag[®] DNA Food kit. Otherwise, increase the respective amount of binding and wash buffers proportionally.
- For processing larger sample volumes an upscale of the lysis buffer volume and the amount of Proteinase K is required. Per each 0.1 g of sample add 275 μL Buffer CF and 5 μL Liquid Proteinase K. After incubation and clearance of the lysate, proceed with step 3 in the protocol and proceed with up to 400 μL of the cleared lysate.
- For fruit juice or similar samples with a low pH-value, an adjustment of the pH to 8 (e.g. by NaOH) can enhance the nucleic acid recovery significantly.
- For processing complex or difficult sample materials, more lysis buffer than indicated in the protocol can be used. Transfer clear supernatant after centrifugation into a suitable reaction container and add 1.5 vol of binding buffer and the respective of NucleoMag[®] B-Beads

Positively tested samples (PCR)

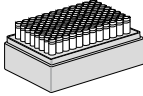
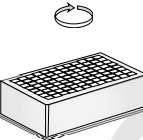
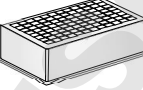

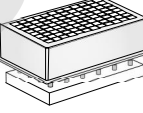


Food (plant origin)	Raw products: maize, soja, rape etc. (powder or oil) Chocolate products: cocoa, nougat products Breakfast cereals, muesli, nut/ chocolate spread Jam and fruit concentrates Pollen / honey Lecithine Spices Bread Various seeds
Food (animal origin)	Raw and processed products (meat, sausage, pie)
Cosmetics	Plant and animal ingredients (e.g., in crème or powder)
Bacteria	Starter cultures, etc.

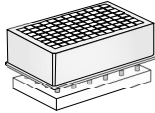

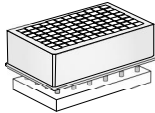

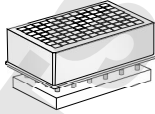
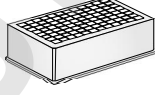

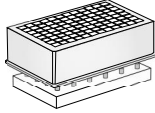
6 Protocol for the isolation of DNA from food samples

6.1 Protocol at a glance

- For additional equipment and hardware requirements, refer to section 1.2 and 2.3, respectively.
- For detailed information on each step, see page 16.

Before starting the preparation:

1	Homogenize samples	0.2 g sample		
		550 µL preheated (65 °C) CF 10 µL Liquid Proteinase K		
		Mix 65 °C, 30 min		
2	Clear lysate	5,600 x g, 20 min		
3	Bind DNA to NucleoMag® B-Beads	Up to 400 µL clear lysate		
		25 µL NucleoMag® B-Beads 600 µL CB		
		Mix by shaking for 10 min at RT <i>(Optional: Mix by pipetting up and down)</i>		
		Remove supernatant after 5 min separation		
4	Wash with CMW	Remove Square-well Block from NucleoMag® SEP 600 µL CMW		
		Resuspend: Shake 2 min at RT <i>(Optional: Mix by pipetting up and down)</i>		

		Remove supernatant after 2 min separation	
5 Wash with CQW		Remove Square-well Block from NucleoMag® SEP 600 µL CQW	
		Resuspend: Shake 2 min at RT <i>(Optional: Mix by pipetting up and down)</i>	↔
		Remove supernatant after 2 min separation	
6 Wash with 80 % Ethanol		Remove Square-well Block on NucleoMag® SEP 600 µL 80 % Ethanol	
		Resuspend: Shake 2 min at RT <i>(Optional: Mix by pipetting up and down)</i>	↔
		Remove supernatant after 2 min separation	
7 Dry the beads		15 min at RT	
8 Elute DNA		Remove Square-well Block from NucleoMag® SEP 50–100 µL CE <i>(Optional: Elute at 56 °C)</i>	
		Shake 5 min at RT <i>(Optional: Mix by pipetting up and down)</i>	↔
		Separate 2 min and transfer DNA into elution plate	

6.2 Detailed protocol

This protocol is designed for magnetic separators with static pins (e.g., NucleoMag® SEP) and suitable plate shakers (see section 2.3 page 6). It is recommended using a Square-well Block for separation (see section 1.2 page 4). Alternatively, isolation of DNA can be performed in reaction tubes with suitable magnetic separators. This protocol is for manual use and serves as a guideline for adapting the kit to robotic instruments.

- For hardware requirements, refer to section 2.3 page 6.

Before starting the preparation:

- Preheat Buffer CF to 65 °C.

For each preparation, collect up to 200 mg of sample into an appropriate lysis vessel (e.g., Rack of Tube Strips)

1 Homogenize samples

Homogenize up to 200 mg sample using a commercial homogenizer. Transfer homogenized samples into Rack of Tube Strips and add 550 µL Lysis Buffer CF preheated to 65 °C. Add 10 µL of Liquid Proteinase K. Close the Tube Strips using Cap Strips and mix by vigorous shaking for 15–30 s. Spin briefly for 30 s at 1,500 x g to collect any sample at the bottom of the Tube Strips.

If the lysis buffer volume is not large enough to dissolve the sample completely add more buffer (and Liquid Proteinase K proportionally) until sample has been totally resuspended.

Incubate at 65 °C for 30 min. For optimal lysis incubate with shaking.

Optional: If RNA-free DNA is crucial for downstream applications an RNase digest may be performed: After incubation at 65 °C for 30 min, add 10 µL RNase A (10 mg/mL stock solution, not provided, see ordering information) per 550 µL lysis buffer, mix well, and incubate at RT (18–25 °C) for 30 min. Proceed with protocol with the centrifugation step.

2 Clear lysate

Centrifuge the samples for 20 min at full speed (5,600–6,000 x g) to pellet contaminations and cell debris. Remove Cap Strips. After homogenization and treatment of the sample with lysis buffer, mixtures can be cleared easily and effectively by either centrifugation or with NucleoSpin® 96 Filter Plate (see ordering information).

3 Bind DNA to NucleoMag® B-Beads

Transfer **up to 400 µL clear lysate to a Square-well block**. Add **25 µL of NucleoMag® B-Beads** and **600 µL of Binding Buffer CB**. Mix by pipetting up and down 6 times and **shake for 5 min at room temperature**. Alternatively, when processing the kit without a shaker, pipette up and down 10 times and incubate for 5 min at room temperature.

Be sure to resuspend the NucleoMag® B-Beads before removing them from the storage bottle. Vortex storage bottle briefly until a homogenous suspension has formed.

Separate the magnetic beads against the side of the wells by placing the Square-well Block on the NucleoMag® SEP magnetic separator. Wait at least 5 min until all the beads have been attracted to the magnets. Remove and discard supernatant by pipetting.

Note: Do not disturb the attracted beads while aspirating the supernatant. The magnetic pellet might not be visible in this step. Remove supernatant from the opposite side of the well.

4 Wash with CMW

Remove the Square-well Block from the NucleoMag® SEP magnetic separator.

Add **600 µL Buffer CMW** to each well and resuspend the beads by shaking until the beads are resuspended completely (**1–3 min**). Alternatively, resuspend beads completely by repeated pipetting up and down.

Separate the magnetic beads by placing the Square-well Block on the NucleoMag® SEP magnetic separator. Wait at least **2–5 min** until all the beads have been attracted to the magnet. Remove and discard supernatant by pipetting.

5 Wash with CQW

Remove the Square-well Block from the NucleoMag® SEP magnetic separator.

Add **600 µL Buffer CQW** to each well and resuspend the beads by shaking until the beads are resuspended completely (**1–3 min**). Alternatively, resuspend beads completely by repeated pipetting up and down.

Separate the magnetic beads by placing the Square-well Block on the NucleoMag® SEP magnetic separator. Wait at least **2 min** until all the beads have been attracted to the magnet. Remove and discard supernatant by pipetting.

6 Wash with 80 % Ethanol

Remove the Square-well Block from the NucleoMag® SEP magnetic separator.

Add **600 µL 80 % ethanol** to each well and resuspend the beads by shaking until the beads are resuspended completely (**1–3 min**). Alternatively, resuspend beads completely by repeated pipetting up and down.

Separate the magnetic beads by placing the Square-well Block on the NucleoMag® SEP magnetic separator. Wait at least **2 min** until all the beads have been attracted to the magnet. Remove and discard supernatant by pipetting.

7 Air dry magnetic beads

Air dry the magnetic beads for 10–15 min at room temperature.

8 Elute DNA

Remove the Square-well Block from the NucleoMag® SEP magnetic separator.

Add desired volume of **Buffer CE (50–100 µL)** to each well of the Square-well Block and resuspend the beads by shaking **5 min** at room temperature. Alternatively, resuspend beads completely by repeated pipetting up and down and incubate for **5–10 min at room temperature** or **56 °C**.

Separate the magnetic beads by placing the Square-well Block on the NucleoMag® SEP magnetic separator. Wait at least **2 min** until all the beads have been attracted to the magnets. Transfer the supernatant containing the purified DNA to either elution plates or tube stripes (see ordering informations).

7 Appendix

7.1 Troubleshooting

Problem	Possible cause and suggestions
Poor DNA yield	<p><i>Homogenization of food materials was not sufficient</i></p> <ul style="list-style-type: none"> For most species, we recommend grinding with steel beads (see section 2.5 page 8) or with commercial bead mills, mixers or homogenizers.
	<p><i>Extraction of DNA from food material during lysis was not sufficient</i></p> <ul style="list-style-type: none"> To obtain higher yields of DNA, the incubation time in lysis buffer can be prolonged (up to overnight).
	<p><i>Sample contains too much RNA</i></p> <ul style="list-style-type: none"> Add 10–20 µL RNase A solution (10 mg/mL) to the lysis buffer before heat incubation. If this is not successful, add the enzyme to the cleared lysate and incubate for 30 min at 37 °C.
	<p><i>Insufficient Elution buffer volume</i></p> <ul style="list-style-type: none"> Beads pellet must be covered completely with elution buffer.
	<p><i>Insufficient performance of elution buffer during elution step</i></p> <ul style="list-style-type: none"> Remove residual buffers during the separation steps completely. Remaining buffers decrease efficiency of following wash steps and elution step.
	<p><i>Aspiration of attracted bead pellet</i></p> <ul style="list-style-type: none"> Do not disturb the attracted beads while aspirating the supernatant, especially when the magnetic pellet is not visible in the lysate.
	<p><i>Aspiration and loss of beads</i></p> <ul style="list-style-type: none"> Time for magnetic separation was too short or aspiration speed was too high.

Insufficient washing procedure

Low purity / Low sensitivity

- Use only the appropriate combinations of separator and plate, for example, Square-well Block in combination with NucleoMag[®] SEP.
- Make sure that beads are resuspended completely during the washing procedure. If shaking is not sufficient to resuspend the beads completely mix by repeated pipetting up and down.
- Repeat washing step with 80 % ethanol.

Insufficient washing procedure

Suboptimal performance of DNA in downstream applications

- Use only the appropriate combinations of separator and plate, for example, Square-well Block in combination with NucleoMag[®] SEP.
- Make sure that beads are resuspended completely during the washing procedure. If shaking is not sufficient to resuspend the beads completely mix by repeated pipetting up and down.
- Repeat washing step with 80 % ethanol.

Carry-over of ethanol from wash buffers

- Be sure to remove all of the ethanolic wash solution, as residual ethanol interferes with downstream applications.

Ethanol evaporation from wash buffers

- Close buffer bottles tightly, avoid ethanol evaporation from buffer bottles as well as from buffer filled in reservoirs. Do not reuse buffers from buffer reservoirs.

Time for magnetic separation too short

Carry-over of beads

- Increase separation time to allow the beads to be completely attracted to the magnetic pins before aspirating any liquid from the well.

Aspiration speed too high (elution step)

- High aspiration speed during the elution step may cause bead carry-over. Reduce aspiration speed for elution step.

Contamination of the rims

Cross contamination

- Do not moisten the rims of the Square-well Block when transferring the lysate. If the rim of the wells is contaminated, seal the Square-well Block with Self-adhering PE Foil (see ordering information) before starting the shaker.

DNA quality is low

Sample contains DNA-degrading contaminants (e.g., phenolic compounds, metabolites)

- Repeat washing step with Buffer CMW.

7.2 Ordering information

Product	REF	Pack of
NucleoMag® DNA Food	744945.1	1 x 96 preps
	744945.4	4 x 96 preps
NucleoSpin® 96 Filter Plate	740663.2	20
Buffer CF	740946	1 L
Proteinase K (lyophilized)	740506	100 mg
Liquid Proteinase K	740396	5 mL
RNase A	740505.50	50 mg
	740505	100 mg
NucleoSpin® Trace Filter Plate	740677	20
NucleoMag® SEP	744900	1
NucleoMag® SEP Mini	744901	1
NucleoMag® SEP Maxi	744902	1
NucleoMag® SEP 24	744903	1
Square-well Blocks	740481	4
	740481.24	24
Self-adhering PE Foil	740676	50 sheets
Rack of Tube Strips	740477	4 sets
(set consists of 1 Rack, 12 Tube Strips with 8 tubes each, and 12 Cap Strips)	740477.24	24 sets
Cap Strips	740638	30 strips

Product	REF	Pack of
Elution Plate U-bottom (96-well 0.3 mL microtiterplate with 300 µL u-bottom wells)	740486.24	24
KingFisher® 96/Flex Accessory Kit A (set consists of Square-well Blocks, Deep-well tip combs, Elution Plates; for 4 x 96 NucleoMag® DNA Food preps using KingFisher® 96/Flex platform)	744950	1 set
KingFisher® DUO Accessory Kit (Square-well Blocks, Deep-well tip combs, Plates for 8 x 12 NucleoMag® DNA Food preps using KingFisher® DUO/DUO Prime platform.	744952	1 set

Visit www.mn-net.com for more detailed product information.

7.3 Product use restriction / warranty

NucleoMag® DNA Food kit 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.

There is no warranty for and MACHEREY-NAGEL is not liable for damages or defects arising in shipping and handling (transport insurance for customers excluded), or out of accident or improper or abnormal use of this product; defects in products or components not manufactured by MACHEREY-NAGEL, or damages resulting from such non-MACHEREY-NAGEL components or products.

MACHEREY-NAGEL makes no other warranty of any kind whatsoever, and SPECIFICALLY DISCLAIMS AND EXCLUDES ALL OTHER WARRANTIES OF ANY KIND OR NATURE WHATSOEVER, DIRECTLY OR INDIRECTLY, EXPRESS OR IMPLIED, INCLUDING, WITHOUT LIMITATION, AS TO THE SUITABILITY, REPRODUCTIVITY, DURABILITY, FITNESS FOR A PARTICULAR PURPOSE OR USE, MERCHANTABILITY, CONDITION, OR ANY OTHER MATTER WITH RESPECT TO MACHEREY-NAGEL PRODUCTS.

In no event shall MACHEREY-NAGEL be liable for claims for any other damages, whether direct, indirect, incidental, compensatory, foreseeable, consequential, or special (including but not limited to loss of use, revenue or profit), whether based upon warranty, contract, tort (including negligence) or strict liability arising in connection with the sale or the failure of MACHEREY-NAGEL products to perform in accordance with the stated specifications. This warranty is exclusive and MACHEREY-NAGEL makes no other warranty expressed or implied.

The warranty provided herein and the data, specifications and descriptions of this MACHEREY-NAGEL product appearing in MACHEREY-NAGEL published catalogues and product literature are MACHEREY-NAGEL's sole representations concerning the product and warranty. No other statements or representations, written or oral, by MACHEREY-NAGEL's employees, agent or representatives, except written statements signed by a duly authorized officer of MACHEREY-NAGEL are authorized; they should not be relied upon by the customer and are not a part of the contract of sale or of this warranty.

Product claims are subject to change. Therefore please contact our Technical Service Team for the most up-to-date information on MACHEREY-NAGEL products. You may also contact your local distributor for general scientific information. Applications mentioned in MACHEREY-NAGEL literature are provided for informational purposes only. MACHEREY-NAGEL does not warrant that all applications have been tested in MACHEREY-NAGEL laboratories using MACHEREY-NAGEL products. MACHEREY-NAGEL does not warrant the correctness of any of those applications.

Last updated: 07/2010, Rev. 03

Please contact:
MACHEREY-NAGEL GmbH & Co. KG
Tel.: +49 24 21 969-270
tech-bio@mn-net.com

Trademarks:

KingFisher® is a registered trademark of Thermo Fisher Scientific

NucleoMag® is a registered trademark of MACHEREY-NAGEL GmbH & Co KG

Te-MagS™ is a trademark of Tecan Group Ltd., Switzerland

All used names and denotations can be brands, trademarks, or registered labels of their respective owner – also if they are not special denotation. To mention products and brands is only a kind of information (i.e., it does not offend against trademarks and brands and can not be seen as a kind of recommendation or assessment). Regarding these products or services we can not grant any guarantees regarding selection, efficiency, or operation.



www.mn-net.com

MACHEREY-NAGEL



MACHEREY-NAGEL GmbH & Co. KG · Neumann-Neander-Str. 6-8 · 52355 Düren · Germany

DE/International:

Tel.: +49 24 21 969-0

Fax: +49 24 21 969-199

E-mail: info@mn-net.com

CH:

Tel.: +41 62 388 55 00

Fax: +41 62 388 55 05

E-mail: sales-ch@mn-net.com

FR:

Tel.: +33 388 68 22 68

Fax: +33 388 51 76 88

E-mail: sales-fr@mn-net.com

US:

Tel.: +1 484 821 0984

Fax: +1 484 821 1272

E-mail: sales-us@mn-net.com

A058404/0100.2