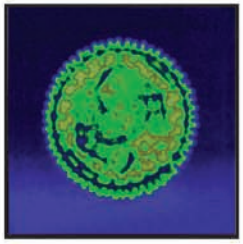

Viral RNA Isolation

User Manual

NucleoSpin® RNA Virus

NucleoSpin® RNA Virus F

Visit our website for more details! click here...

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A Takara Bio Company
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PT4028-1
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May 2009 / Rev. 07

MACHEREY-NAGEL

Viral RNA Isolation

Protocol-at-a-glance (Rev. 07)

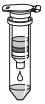
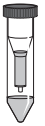
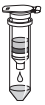
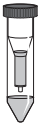
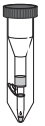
		Mini	Funnel
		NucleoSpin® RNA Virus	**NucleoSpin® RNA Virus F**
1	**Lysis of viruses**	 150 µl sample volume 600 µl RAV1 70°C 5 min	 1 ml sample volume 4 ml RAV1 70°C 5 min
2	**Adjust DNA binding condition**	 600 µl ethanol	 4 ml ethanol
3	**Bind viral RNA**	 Load sample stepwise  8,000 x *g* 1 min	 Load sample  3,000 x *g* 3 - 5 min
4	**Wash and dry silica membrane**	 1st wash 500 µl RAW 2nd wash 600 µl RAV3 3rd wash 200 µl RAV3  1st and 2nd 8,000 x *g* 1 min  3rd 11,000 x *g* 5 min	 1st wash 5 ml RAW 2nd wash 8 ml RAV3 3rd wash 2 ml RAV3  1st and 2nd 3,000 x *g* 3 min  3rd 3,000 x *g* 10 min
5	**Elute highly pure RNA**	 50 µl RNase-free H_2O (70°C) RT 1 - 2 min  11,000 x *g* 1 min	 50 - 100 µl RNase-free H_2O (70°C) RT 1 - 2 min  3,000 x *g* 3 min

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Table of contents

1 Components

1.1 Kit contents

	NucleoSpin® RNA Virus		
	10 preps	**50 preps**	**250 preps**
Cat. No.	**740956.10**	**740956.50**	**740956.250**
Lysis Buffer RAV1	10 ml	35 ml	5 x 35 ml
Wash Buffer RAW	6 ml	30 ml	2 x 75 ml
Wash Buffer RAV3 (Concentrate)*	12.5 ml	12.5 ml	3 x 25 ml
RNase-free H_2O	5 ml	5 ml	25 ml
Elution Buffer RE**	5 ml	5 ml	25 ml
Carrier RNA (lyophilized)	300 µg	1 mg	5 x 1 mg
NucleoSpin® RNA Virus Columns (dark blue rings, plus Collection Tubes)	10	50	250
Collection Tubes (2 ml)	30	150	750
User Manual	1	1	1

* For preparation of working solutions and storage conditions see section 3.

** Composition of Elution Buffer RE: 5 mM Tris/HCl, pH 8.5

1.1 Kit contents *continued*

	NucleoSpin® RNA Virus F
	25 preps
Cat. No.	**740958**
Lysis Buffer RAV1	2 x 120 ml
Wash Buffer RAW	2 x 80 ml
Wash Buffer RAV3 (Concentrate)*	3 x 25 ml
RNase-free H_2O	6 ml
Elution Buffer RE**	5 ml
Carrier RNA (lyophilized)	2 x 300 µg
NucleoSpin® RNA Virus F Columns (plus Collection Tubes)	25
Collection Tubes (50 ml)	25
Collection Tubes (0.5 ml)	25
User Manual	1

* For preparation of working solutions and storage conditions see section 3.

** Composition of Elution Buffer RE: 5 mM Tris/HCl, pH 8.5

1.2 Reagents, consumables, and equipment to be supplied by user

Reagents

- 96 - 100% ethanol

Consumables

- 1.5 ml microcentrifuge tubes (NucleoSpin® RNA Virus) or 50 ml tubes (NucleoSpin® RNA Virus F)
- Disposable tips

Equipment

- Manual pipettors
- Centrifuge for microcentrifuge tubes (NucleoSpin® RNA Virus) or 50 ml tubes (NucleoSpin® RNA Virus F)
- Vortex mixer
- Heating block or water bath for 70°C incubation
- Personal protection equipment (lab coat, gloves, goggles)

1.3 About this User Manual

It is strongly recommended reading the detailed protocol sections of this User Manual if the **NucleoSpin® RNA Virus/RNA Virus F** kit is used for the first time. Experienced users, however, my refer to the Protocol-at-a-glance instead. The Protocol-at-a-glance is designed to be used only as a supplemental tool for quick referencing while performing the purification procedure.

All technical literature is available on the internet at *www.mn-net.com*.

2 Product description

2.1 The basic principle

With the **NucleoSpin® RNA Virus** method, RNA viruses are lysed quickly and efficiently by Lysis Buffer RAV1 which is a highly concentrated solution of GITC. DNA viruses (e.g. HBV) are usually more difficult to lyse and require Proteinase K digestion (see support protocol, section 5.2). Lysis buffer and ethanol create appropriate conditions for binding of nucleic acids to the silica membrane of the **NucleoSpin® RNA Virus Columns**. Carrier RNA improves binding and recovery of low-concentrated viral RNA. Contaminations (potential PCR inhibitors) like salts, metabolites and soluble macromolecular cellular components are removed in simple washing steps with ethanolic buffers **RAW** and **RAV3**. The nucleic acids can be eluted in low salt buffer or water and are ready-for-use in subsequent reactions.

2.2 Kit specifications

- **NucleoSpin® RNA Virus/Virus F** kits are designed for the rapid preparation of highly pure viral nucleic acids (e.g. HCV, HIV, CMV) from fluid biological samples e.g. plasma, serum, urine, but not blood (see remarks in section 2.1).
- No cross contamination due to closed systems.
- The **NucleoSpin® RNA Virus** kit works with 150 µl serum, **NucleoSpin® RNA Virus F** funnel columns allow the processing of 1 ml serum.
- The funnel column of the **NucleoSpin® RNA Virus F** kit allows a high loading capacity by simultaneously small elution volume.
- The prepared nucleic acids are suitable for applications like automated fluorescent DNA sequencing, RT-PCR*, or any kind of enzymatic reaction. The detection limit for certain viruses depends on the individual procedures e.g. in-house nested (RT-) PCR. We highly recommend using internal (low-copy) standards as well as positive and negative controls to monitor the purification, amplification, and detection processes.

* PCR is patented by Roche Diagnostics

Table 1: Kit specifications at a glance		
Parameter	**NucleoSpin® RNA Virus (Mini)**	**NucleoSpin® RNA Virus F (Funnel)**
Sample size	Up to 150 µl	Up to 1 ml
Typical recovery rates	> 90%	> 90%
Typical analysis limit	30 - 60 cp/ml*	30 - 60 cp/ml*
Elution volume	50 µl	50 µl
Binding capacity	40 µg	30 µg
Time/prep	30 min/4 - 6 preps	45 min/2 - 4 preps
Column type	Mini spin column	Funnel column

2.3 Remarks regarding sample quality and preparation

All kinds of biological fluids or semi-fluid samples can be processed e.g. serum, urine, or BAL. For successful nucleic acid purification, it is important to obtain a homogeneous, clear and non-viscous sample before loading onto the corresponding **NucleoSpin® RNA Virus Columns**. Therefore, check all samples (especially old or frozen ones) for precipitates. Avoid clearing samples by centrifugation/filtration before the RAV1-lysis step, because viruses may be associated with particles or aggregates. Incubation with Buffer RAV1 may be prolonged to dissolve and digest residual cell structures, precipitates and virus particles. However, RNA is sensitive to autolysis and prolonged incubation may cause degradation and decreased yields.

This User Manual contains support protocols for isolation of viral DNA including a Proteinase K (not included, see ordering information) digestion step.

* Nested PCR

2.4 Remarks regarding elution

- Pure nucleic acids are finally eluted under low ionic strength conditions with RNase-free H_2O (pH about 7 - 8) or slightly alkaline Buffer RE (5 mM Tris-HCl, pH 8.5).

- Elution can be performed in a single step with water/elution buffer as indicated in the protocol, obtaining at least 80% of the bound nucleic acids. To improve sensitivity, this eluate can be used in a second elution step increasing the efficiency of elution and concentration of viral nucleic acids slightly. Alternatively, a second elution step can be performed with an additional volume of water/elution buffer releasing practically all bound nucleic acids but resulting in a lower concentrated, combined eluate.

- RNA should be eluted with the water supplied and DNA with Elution Buffer RE. Buffer RE provides better storage conditions for DNA. To elute both types of nucleic acids together, use the pH proofed (pH 6 - 8), RNase-free H_2O preheated to 70°C.

2.5 Remarks regarding quality control

- Buffers and **NucleoSpin® RNA Virus/RNA Virus F Columns** have been tested with rRNA and MS2 phage RNA. The absence of RNases, and the yield and efficiency of purification have been investigated with RT-PCR.

3 Storage conditions and preparation of working solutions

Attention:

Buffers RAV1 and RAW contain guanidine salts! Wear gloves and goggles!

- All kit components can be stored at room temperature (20 - 25°C) and are stable up to one year.
- **Carrier RNA** has a limited shelf life in Buffer RAV1. For this reason, some kits contain several bottles of lyophilized Carrier RNA that should be used successively as required, to avoid degradation of Carrier RNA.
- Before use, add 1 ml **Lysis Buffer RAV1** to the Carrier RNA tube. Dissolve the RNA and transfer it back to the RAV1 bottle.

Storage of Carrier RNA in Buffer RAV1:

- **Lysis Buffer RAV1 including Carrier RNA** can be stored at **room temperature** for **1 - 2 weeks**. Storage at room temperature prevents salt precipitation.
- **Lysis Buffer RAV1 including Carrier RNA** can be stored at **4°C** for up to **4 weeks** or aliquoted and stored at **-20°C for longer periods**. Storage at 4°C or below may cause salt precipitation. Therefore, the mixture must be preheated at 40 - 60°C for a maximum of 5 min in order to redissolve salts.
- **Do not warm Buffer RAV1 containing Carrier RNA more than 4 times!** Frequent warming, temperatures > 80°C, and extended heat incubation will accelerate the degradation of Carrier RNA. This leads to reduced recovery of viral RNA and eventually false negative RT-PCR results, in particular if low-titer samples are used.

Before starting any **NucleoSpin® RNA Virus/RNA Virus F** protocol prepare the following:

- **Wash Buffer RAV3:** Add the indicated volume of ethanol (96 - 100%) to Wash Buffer RAV3 Concentrate. Mark the label of the bottle to indicate that the ethanol is added. Store Wash Buffer RA3 at room temperature (20 - 25°C) for up to one year.

	NucleoSpin® RNA Virus		
	10 preps	**50 preps**	**250 preps**
Cat. No.	**740956.10**	**740956.50**	**740956.250**
Wash Buffer RAV3 (Concentrate)	12.5 ml Add 50 ml ethanol	12.5 ml Add 50 ml ethanol	3 x 25 ml Add 100 ml ethanol to each vial

	NucleoSpin® RNA Virus F
	25 preps
Cat. No.	**740958**
Wash Buffer RAV3 (Concentrate)	3 x 25 ml Add 100 ml ethanol to each vial

4 Safety instructions – risk and safety phrases

The following components of the **NucleoSpin® RNA Virus** kits contain hazardous contents.

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

Component	Hazard contents	Hazard symbol		Risk phrases	Safety phrases
RAV1	Guanidine thiocyanate	✖ Xn*	Harmful by inhalation, in contact with the skin and if swallowed	R 20/21/22	S 13
RAW	Guanidine hydrochloride + ethanol <50%	✖ Xn*	Flammable - Harmful if swallowed - Irritating to eyes and skin	R 10-22-36/38	S 7-16

Risk phrases

R 10	Flammable
R 20/21/22	Harmful by inhalation, in contact with the skin and if swallowed
R 22	Harmful if swallowed
R 36/38	Irritating to eyes and skin

Safety phrases

S 7	Keep container tightly closed
S 13	Keep away from food, drink, and animal feedstuffs
S 16	Keep away from sources of ignition - No Smoking!

* Hazard labeling not neccessary if quantity per bottle below 125 g or ml (certificate of exemption according to 67/548/EEC Art. 25, 1999/45/EC Art. 12 and German GefStoffV § 20 (3) and TRGS 200 7.1). For further information see Material Safety Data Sheet.

5 Protocols

5.1 Viral RNA isolation from cell-free biological fluids with NucleoSpin® RNA Virus

Before starting the preparation:

- Check if Wash Buffer RAV3 was prepared according to section 3.
- Preheat an aliquot of Elution Buffer RE/RNase-free H_2O to 70°C.

1 Lysis of viruses

Add **600 µl Buffer RAV1** containing Carrier RNA to **150 µl of the sample**. Pipette mixture up and down and vortex well. Incubate for **5 min** at **70°C**.

Incubation time and temperature are critical for lysis as well as RNA stability (see troubleshooting for further hints, section 6.1).

*If the resulting solution is still turbid, centrifuge the mixture for **1 min** at **11,000 x g** (to pellet particles and to prevent clogging of the NucleoSpin® RNA Virus Columns). Take off the supernatant and proceed with step 2.*

150 µl sample

+ 600 µl RAV1

70°C
5 min

2 Adjust binding condition

Add 600 µl ethanol (96 - 100%) to the clear lysis solution and mix by vortexing (10 - 15 s).

+ 600 µl EtOH

3 Bind viral RNA

Place NucleoSpin® RNA Virus Columns in Collection Tubes (2 ml) and load **700 µl** lysed sample. Centrifuge for **1 min** at **8,000 x *g***.

The use of new Collection Tubes (2 ml) is recommended if infectious material has to be prepared.

Load sample stepwise

8,000 x *g*
1 min

Load the residual lysis solution onto the NucleoSpin® RNA Virus Column. Centrifuge for **1 min** at **8,000 x *g***. Discard flow-through and place the NucleoSpin® RNA Virus Column into another new Collection Tube (2 ml). More than two loading steps are not recommended.

4 Wash and dry silica membrane

1st wash

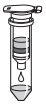
Add **500 µl Buffer RAW** to the NucleoSpin® RNA Virus Column. Centrifuge for **1 min** at **8,000 x *g***. Discard flow-through.

This washing step removes contaminants and PCR inhibitors.

+ 500 µl RAW

8,000 x *g*
1 min

2nd wash



Add **600 µl Buffer RAV3** to the NucleoSpin® RNA Virus Column. Centrifuge for **1 min** at **8,000 x *g***. Discard flow-through with Collection Tube.

+ 600 µl RAV3

8,000 x *g*
1 min

3rd wash

Place the NucleoSpin® RNA Virus Column in a new Collection Tube (2 ml) and add **200 µl Buffer RAV3**. Centrifuge for **2 - 5 min** at **11,000 x *g*** to remove ethanolic Buffer RAV3 completely.

Optional: Residual Buffer RAV3 may inhibit subsequent reactions. Therefore, for subsequent reactions which are extremely ethanol-sensitive, we recommend repeating the centrifugation with a new Collection Tube (2 ml). Or alternatively, incubate the NucleoSpin® RNA Virus Columns for 1 min at 70°C to remove any remaining traces of ethanol.

+ 200 µl RAV3

11,000 x *g*
5 min

5 Elute viral RNA

Place the NucleoSpin® RNA Virus Column into a new, sterile 1.5 ml microcentrifuge tube (not provided). Add **50 µl RNase-free H_2O (preheated to 70°C)** and incubate for **1 - 2 min**. Centrifuge for **1 min** at **11,000 x *g***.

50 µl
RNase-free H_2O
(70°C)

RT
1 - 2 min

To elute viral DNA which was prepared according to the support protocol 4.2, we recommend using Buffer RE, preheated to 70°C (also see section 2.4).

8,000 x *g*
1 min

5.2 Support protocol: Isolation of viral RNA and DNA from cell-free biological fluids with NucleoSpin® RNA Virus

This protocol is recommended for the purification of viral RNA and viral DNA for all types of DNA viruses like HBV and CMV from small samples of up to 150 µl.

Before starting the preparation:

- Check if Wash Buffer RAV3 was prepared according to section 3
- Check if Proteinase K solution (not included, see ordering information) was prepared.
- Preheat an aliquot of Elution Buffer RE/RNase-free H_2O to 70°C.

1 Lysis of viruses

Add **600 µl Buffer RAV1** containing Carrier RNA to **150 µl of the sample**. Add **20 µl Proteinase K** (20 mg/ml stock solution), to the lysis mixture. Pipette mixture up and down and vortex for 10 - 15 s. Incubate for **5 min** at **70°C**.

Incubation time and temperature are critical for lysis as well as RNA stability (see troubleshooting).

*Proteinase K is not included in this kit, but can be ordered separately (see ordering information). For the isolation of viral DNA and genomic DNA from other matrices (**not cell-free)** like blood we recommend the **NucleoSpin® Blood** or **NucleoSpin® Tissue** kit (see ordering information).*

*If the resulting solution is still turbid, centrifuge the mixture for **1 min** at **11,000 x g** to pellet particles and to prevent clogging of the NucleoSpin® RNA Virus Columns. Take off the supernatant and continue with step 2 of protocol 5.1.*

150 µl sample

+ 600 µl RAV1

+ 20 µl Prot. K

70°C
5 min

5.3 Viral RNA isolation from cell-free biological fluids with NucleoSpin® RNA Virus F

Before starting the preparation:

- Check if Wash Buffer RAV3 was prepared according to section 3.
- Preheat an aliquot of Elution Buffer RE/RNase-free H_2O to 70°C.

1 Lysis of viruses

Add **4 ml Buffer RAV1** containing Carrier RNA to **1 ml of the sample**. Pipette mixture up and down and vortex well. Incubate for **5 min** at **70°C**.

Incubation time and temperature are critical for lysis as well as RNA stability (see troubleshooting for further hints).

*If the resulting solution is still turbid, centrifuge the mixture for **1 min** at **11,000 x g** (to pellet particles and to prevent clogging of the NucleoSpin® RNA Virus F Columns). Take off the supernatant and proceed with step 2.*

1 ml sample

+ 4 ml RAV1

70°C
5 min

2 Adjust binding condition

Add **4 ml ethanol** (96-100%) to the clear lysis solution and mix by vortexing (10-15 s).

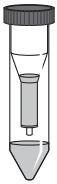
+ 4 ml EtOH

3 Bind viral RNA

Take the NucleoSpin® RNA Virus F Column placed in a Collection Tube and load lysed sample. Centrifuge for **3-5 min** at **3,000 x *g***.

The use of new 50 ml tubes for every step is recommended if infectious material has to be prepared. This avoids cross-contamination and contamination of centrifuge units. For non-infectious samples, we recommend discarding the flow-through and reusing the 50 ml tube for loading and washing steps. Additional 50 ml tubes have to be ordered separately.

The maximum loading capacity of the NucleoSpin® RNA Virus F Column is about 10 ml in order to work cross-contamination free. If more sample has to be loaded discard flow-through and put the NucleoSpin® RNA Virus F Column into a new 50 ml tube.



Load sample

3,000 x *g*
3-5 min

*Load the residual lysis solution onto the NucleoSpin® RNA Virus F Column. Centrifuge for **3 - 5 min** at **3,000 x g**. Discard flow-through and place the NucleoSpin® RNA Virus F Column into another new 50 ml tube. More than two loading steps are not recommended.*

4 Wash and dry silica membrane

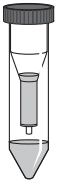
1st wash

Add **5 ml Buffer RAW** to the NucleoSpin® RNA Virus F Column. Centrifuge for **3 min** at **3,000 x *g***. Discard flow-through.

+ 5 ml RAW

3,000 x *g*
3 min

This washing step removes contaminants and PCR inhibitors.



2nd wash

Add **8 ml Buffer RAV3** to the NucleoSpin® RNA Virus F Column. Centrifuge for **3 min** at **3,000 x *g***. Discard flow-through with Collection Tube.

+ 8 ml RAV3

3,000 x *g*
3 min

3rd wash

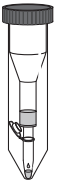
Place the NucleoSpin® RNA Virus F Column in a new Collection Tube (50 ml) and add **2 ml Buffer RAV3**. Centrifuge for **10 min** at **3,000 x *g*** to remove ethanolic Buffer RAV3 completely.

+ 2 ml RAV3

3,000 x *g*
10 min

<u>Optional:</u> Residual Buffer RAV3 may inhibit subsequent reactions. Therefore, for subsequent reactions, which are extremely ethanol-sensitive, we recommend repeating the centrifugation with a new Collection Tube (50 ml). Or alternatively, incubate the NucleoSpin® Virus F Columns for 1 min at 70°C to remove any remaining traces of ethanol.

5 Elute viral RNA



50 - 100 µl
RNase-free
H_2O (70°C)

Attach the supplied Collection Tube (0.5 ml) with the adaptor to the NucleoSpin® RNA Virus F Column. Place the assembly in a 50 ml tube (not provided). Add **50 - 100 µl RNase-free H_2O (preheated to 70°C)** and incubate for **1 - 2 min** at **room temperature**. Centrifuge for **3 min** at **3,000 x *g***.

RT
1 - 2 min

3,000 x *g*
3 min

5.4 Support protocol: Isolation of viral RNA and DNA from cell-free biological fluids with NucleoSpin® RNA Virus F

This protocol is recommended for the purification of viral RNA and viral DNA for all types of DNA viruses like HBV and CMV for samples of up to 1 ml.

Before starting the preparation:

- Check if Wash Buffer RAV3 was prepared according to section 3.
- Check if Proteinase K solution (not included, see ordering information) was prepared.
- Preheat an aliquot of Elution Buffer RE/RNase-free H_2O to 70°C.

1 Lysis of viruses

Add **4 ml Buffer RAV1** containing Carrier RNA to **1 ml of the fluid sample**. Add **133 µl Proteinase K** (20 mg/ml stock solution), to the lysis mixture. Pipette mixture up and down and vortex for 10 - 15 s. Incubate for **5 min** at **70°C**.

Incubation time and temperature are critical for lysis as well as RNA stability (see troubleshooting).

*Proteinase K is not included in this kit, but can be ordered separately (see ordering information). For the isolation of viral DNA and genomic DNA from other matrices (**not cell-free)** like blood we recommend the **NucleoSpin® Blood** or **NucleoSpin® Tissue** kit (see ordering information).*

*If the resulting solution is still turbid, centrifuge the mixture for **1 min** at **11,000 x g** to pellet particles and to prevent clogging of the NucleoSpin® RNA Virus Columns. Take off the supernatant and continue with step 2 of protocol 5.1.*

1 ml sample

+ 4 ml RAV1

+ 133 µl Prot. K

70°C
5 min

6 Appendix

6.1 Troubleshooting

Problem	Possible cause and suggestions
Small amounts or no viral nucleic acids in the eluate	*Problems with Carrier RNA* • Carrier RNA not added. • See remarks concerning storage of Buffer RAV1 with Carrier RNA (section 3). *Proteinase K digestion may be necessary* • Use and compare protocols with and without Proteinase K digestion or prolong incubation time to 10 min. *Viral nucleic acids degraded* • Samples should be processed immediately. If necessary, add RNase inhibitor to the sample and ensure appropriate storage conditions up to the processing. • Check that all buffers have been prepared and stored correctly. If in doubt, use new aliquots of Buffer RAV1, Carrier RNA and Elution Buffer RE.
Problems with subsequent detection	*Reduced sensitivity* • Change the volume of eluate added to the PCR/RT-PCR. • Incubation time and temperature are critical for lysis as well as RNA stability. For sensitive RNA preparations, incubation at room temperature is sufficient without significant loss of sensitivity. For parallel isolation of viral RNA and DNA incubation time (5 - 15 min) and temperature (RT/56°C/72°C) may be adapted in order to get optimal recovery rates for both species. *Ethanol carry-over* • Prolong centrifugation steps in order to remove Buffer RAV3 completely.
General problems	*Clogged membrane* • Centrifuge plasma lysate before the addition of ethanol and subsequent loading onto the corresponding NucleoSpin® RNA Virus Columns.

6.2 Ordering information

Product	Cat. No.	Pack of
NucleoSpin® RNA Virus	740956.10/.50/.250	10/50/250
NucleoSpin® RNA Virus F	740958	25
NucleoSpin® Funnel Columns	740959	30 sets
Proteinase K	740506	100 mg
rDNase Set (Recombinant DNase and Reaction Buffer for rDNase; sufficient for 50 mini preps)	740963	1 set
NucleoSpin® Blood	740951.10/.50/.250	10/50/250
NucleoSpin® Tissue	740952.10/.50/.250	10/50/250
Collection Tubes (2 ml)	740600	1000

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

6.3 References

M.L. Villahermosa, M.Thomson, E.Vazques de Parga, M.T. Cuevas, G. Contreas, L.Perez-Alvarez, E.Delgado, N.Manjon, L.Medrano and R.Najera. Improved Conditions for Extraction and Amplification of Human Immunodeficiency Virus Type 1 RNA from Plasma samples with low viral load. Journal of Human Virology 3: 27-34, (2000)

6.4 Product use restriction / warranty

NucleoSpin® RNA Virus kit components were developed, designed, distributed, and sold **FOR RESEARCH PURPOSES ONLY.** They are suitable **FOR *IN-VITRO* USES ONLY**. No claim or representation is intended for its use to identify any specific organism or for clinical use (diagnostic, prognostic, therapeutic, or blood banking).

It is rather the responsibility of the user to verify the use of the **NucleoSpin® RNA Virus** kit for a specific application range as the performance characteristic of this kit has not been verified to a specific organism.

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