Hyris
NL Nucleic Acid extraction kit
Instructions for Use (IFU)

REF: HR043X025A
HR043X032A

Version 1.1

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INTENDED USE

Hyris **NL Nucleic Acid extraction kit** is intended for the isolation and purification of high-quality nucleic acids for molecular biology applications. This product is not intended for the diagnosis, prevention, or treatment of disease.

Hyris **NL Nucleic Acid extraction kit** is intended for use in conjunction with automated magnetic bead extraction systems. The system is intended for use by professional users trained in molecular biological techniques.

PRINCIPLE OF THE METHOD

Hyris **NL Nucleic Acid extraction kit** is an automated extraction system whose procedure is based on the isolation of nucleic acids by a lysis buffer and the reversible adsorption of nucleic acids.
Hyris **NL Nucleic Acid extraction kit** is based on magnetic bead technology. Nucleic acids bind to the surface of the magnetic spheres and are released using a buffer system.
After the lysis phase of the sample, the nucleic acids are bound to specific magnetic beads and purified with a series of washes aimed at eliminating proteins, contaminants, and impurities.
Finally, the nucleic acids are eluted with an Elution Buffer that induces the release of nucleic acid from the magnetic beads.
The high-quality, purified nucleic acids obtained from the application of the Hyris **NL Nucleic Acid extraction kit** are ready for use in applications such as Real Time PCR assays.

Hyris **NL Nucleic Acid extraction kit** is for general use and can be used for the isolation of DNA from plants (Olea europaea, Cannabis sativa), probiotics (Lactobacillus spp and Bifidobacterium spp) (1) (2) and other biological samples like for instance blood, urine and cells (3) (4)

PRODUCT CONTENT

KIT DESCRIPTION

REF	Trade Name	Contents	Size
HR043X025A	NL Nucleic Acid extraction kit	25 NL beads extraction strip 14 Tips comb	25 reactions 2 x 7 bags
HR043X032A	NL Nucleic Acid extraction kit	2 NL beads extraction plate 4 Tips comb	2 x 16 reactions 2 x 2 bags

HR043X025A – For Allsheng Auto-Pure Mini	
Components of an Extraction Strip	Well position
NL Solution	1
Magnetic beads	2
Washing Buffer 1	3
Washing Buffer 2	4
Washing Buffer 2	5
Elution Buffer	6

HR043X032A – For Allsheng Auto-Pure Mini		
Components of an Extraction Plate	Well position	
NL Solution	1	7
Magnetic beads	2	8
Washing Buffer 1	3	9
Washing Buffer 2	4	10
Washing Buffer 2	5	11
Elution Buffer	6	12

MATERIAL REQUIRED BUT NOT PROVIDED (available separately from HYRIS S.r.l.)

REF	Trade Name	Description	Notes
Sk.1x-Acc.01	Accessory Starter Kit	3 x Micro-pipettes (10 µL, 100 µL, 1000 µL) Vortex	Customizable according to the needs of the Laboratory
AS-17170-00	Auto-Pure Mini (Hangzhou Allsheng Instruments Co., Ltd.)	Miniature Automatic Extractor	Up to 16 samples per run
HR039X032P	NL Lysis buffer	NL Solution 32 extraction size	For plant and probiotics sample pretreatment
HR039X050P	NL Lysis buffer	NL Solution 50 extraction size	For plant and probiotics sample pretreatment

REQUIRED MATERIAL

- Vortex mixer
- Calibrated 100 µL and 1000 µL precision micropipettes.
- Sterile 100 µL and 1000 µL filter tips.
- PCR grade 1.5 mL tubes.
- Tube racks.
- DPC and PPE such as (but not limited to) full-length long-sleeved lab coats and disposable gloves.
- Refrigerator from 2 to 8°C for the storage of the whole sample.
- Freezer from -10 to -30 °C for storage of the extracted sample.

REAGENT STORAGE

Store Hyris **NL Nucleic Acid extraction kit** at the temperature indicated on the label.
The expiry date can be found on the label. Do not use the Product after the expiration date indicated on the label.
Pre-filled plates and individual tests should be stored with the aluminium foil side facing up.

SAMPLE COLLECTION AND STORAGE AND PRETREATMENT

Use freshly collected whole sample or dry grinded sample.
As plant tissue is very robust, the lysis procedure is most effective with well homogenized, powdered samples. Suitable methods include grinding or using steel beads.
Liquified samples can be directly loaded into the extraction plate/strip.
Steam-treated raw material, liquid, or powders are not matrixes suitable for DNA extraction. The steam process can reduce the yield or the quality of the nucleic acids.
Cells can be stored in transport media as well as cytological fluid. Cell suspended in culture media as well as cytological fluid must be washed in PBS and concentrated in 400uL prior the extraction.
To get help for the sample preparation contact support@hyris.net

Solid samples preparation (plant and probiotics)

- Take a new 2 mL tube
- Add stainless steel beads
- Add the samples (leaves)
- Add 1 – 1.5 mL of Hyris NL Lysis buffer (#HR039X032P) or Nuclease Free Water
- Vortex at maximum speed for 1 minute
- Load into the extraction plate/strip

WARNINGS AND PRECAUTIONS

- For *Research Use Only;*
- Read the instructions of this user manual carefully before use.
- Wear personal protective equipment such as (but not limited to) full-length long-sleeved lab coats and disposable gloves.
- Use the appropriate DPCs for the type of sample to be analyzed;
- Do not use the pipette by mouth.
- Do not smoke, drink, eat, handle contact lenses, or wear makeup in areas where kit reagents are used.
- Dispose of unused kit reagents and samples according to local, state, and federal regulations.
- Contamination of samples or reagents can produce incorrect results. Use good laboratory practices and control workflow;
- Use only the protocol described in this insert. Deviations from the protocol or the use of times or temperatures other than those specified may give incorrect results.
- The procedures described in this IFU should be performed at laboratory room temperature.
- Do not use the kit past the expiration date.
- Use calibrated micropipettes to transfer the sample.
- After removing the aluminium foil, do not expose the strips/plates to air for a long time to avoid evaporation and/or pH change and thus affect the purification efficiency.
- Safety Data Sheets (SDS) are provided on request for each reagent.
- Do not use the product if the primary or secondary packaging has been visually compromised. Immediate contact Hyris S.r.l. (support@hyris.net);
- When required by downstream applications, internal control can be added to the sample prior to the isolation and purification process.

OPERATING PROCEDURE

Before you start extraction, make sure that the extraction program, "**HR043vX**", is already set on the instrument. To get the latest version of the extraction protocol, visit the Hyris support centre https://support.hyris.net

Turn on the automatic extraction system referring to the manufacturer's manual.

Before each use of the strips or pre-filled plates, check the integrity of the package.

HR043X032A Plate Filling - Auto-Pure Mini

- Resuspend each pre-filled plate by turning it upside down three times, then give a sharp blow by holding the closing aluminum film up and making sure the reagents are at the bottom of the wells and not adhering to the bottom of the aluminum closure film.
- Swirl the liquid sample with the vortex for 30 seconds. Refer to the solid sample preparation paragraph for the solid sample preparation.
- Carefully remove aluminum foil to avoid liquid leakage.
- Place 2 Tips Comb in the appropriate slots on the instrument.
- Add 250 µL of sample into the wells in column 1 and column 7 of the plate.
- Place the plate in the appropriate housing of the instrument with the adhesive placed on the side of the plate facing the operator (the well A1 facing the top left of the instrument).
- Close the door.
- Select the "**HR034vX" protocol,** already present on the instrument, and start the procedure; for information on the program, refer to the "Programming the automatic extractor" section.
- At the end of the program, a short alarm will sound. Carefully pull out the griddle.
- Transfer the liquid containing purified nucleic acid from columns 6 and column 12 into clean tubes.
- The sample can be used immediately or stored at -20°C.
- Remove and discard the Tips Comb immediately.
- Condutct the sterilization program with U.V. for 10 min (suggested).

HR043X025A Strip Filling - Auto-Pure Mini

- Resuspend each pre-filled strip by turning it upside down three times, then give a sharp blow by holding the sealing foil film upwards and making sure the reagents are at the bottom of the wells and not adhering to the bottom of the foil sealing film.
- Place a strip for each sample to be analyzed on the strip rack supplied with the instrument and assign the sample identification code to each one.
- Swirl the liquid sample with the vortex for 30 seconds. Refer to the solid sample preparation paragraph for the solid sample preparation.
- Carefully remove the aluminium foil to avoid liquid leakage.
- Place 1 or 2 Tips comb (depending on the arrangement of the specimens in the strip rack) in the appropriate slot of the instrument.
- Add 250 µL of sample into the first well of each strip.
- Place the strip rack in the instrument holder bay with the well A1 facing the top left of the instrument.
- Close the door.
- Select the "**HR043vX" program,** already present on the instrument, and start the procedure; for information on the program, refer to the "Automatic extractor programming" section.
- At the end of the program, a short alarm will sound. Carefully pull out the strip rack.
- Transfer the liquid containing purified nucleic acid from the well 6 (last well) of each strip into clean tubes.
- The sample obtained can be used immediately or stored at -20°C.
- Remove and discard the Tips comb immediately.
- Conduct the sterilization program with U.V. for 10 min (suggested).

Automatic extractor programming

Allsheng Auto-pure mini

The last version of the extraction program is already set on the instrument. The extraction program can be set by QR code available on the Hyris support centre: https://support.hyris.net
For manual setup: set the extraction program on the All-sheng Auto-pure mini automatic magnetic bead extractor as below.

Step	1	2	3	4	5	6	7
Well	2	1	3	4	5	6	5
Name	Bind	Lysis	Wash 1	Wash 2	Wash 3	Elution	Recycle
Volume	200 µL	700 µL	400 µL	500 µL	500 µL	60 µL	200 µL
Mix time	0.2 min	5 min	1 min	0.5 min	0.5 min	5 min	0.3 min
Mix speed	5	7	7	7	7	5	5
Dry time	/	/	0 min	0 min	2 min	/	/
Temperatures	/	65°C	/	/	/	65°C	/
Segments	3	3	3	3	3	1	1
Segment time	20 s	20 s	20 s	20 s	20 s	60 s	0 s
Magnet time	15 s	15 s	30 s	15 s	15 s	40 s	0 s
Magnet speed	0.5 mm/s	0.5 mm/s	0.5 mm/s	0.5 mm/s	0.5 mm/s	0.5 mm/s	2.5 mm/s

To get QR code with the last version of the extraction program visit the Hyris support centre https://support.hyris.net

LIMITATIONS

- This product is for research use only and should only be used by trained professionals.
- The test result is related to the collection, handling, delivery, and storage of the sample. Any deviation from the indicated procedure will result in an inaccurate detection result.
- The result depends on the correctness of the collection and storage of the sample.
- The use of unsuitable tools may compromise the results.
- The use of PCR grade consumables may compromise the result.
- Refer to the operating instructions of the kits for downstream applications.

REFERENCES

1. *A rapid and cost-effective method for DNA extraction from archival herbarium specimens.* Krinitsina, A.A., Sizova, T.V., Zaika, M.A. 1478–1484, s.l. : Biochemistry Moscow, 2015, Vol. 80.
2. *A single protocol for extraction of gDNA from bacteria and yeast.* Vingataramin, L. e Frost, E.H. 120–125, s.l. : BioTechniques, 2015, Vol. 58.
3. *Long-Term Stability of Human Genomic and Human Papillomavirus DNA Stored in BD SurePath and Hologic PreservCyt Liquid-Based Cytology Media.* Agreda, Patricia M. s.l. : J Clin Microbiol., 2013.
4. *Rapid, simple alkaline extraction of human genomic DNA from whole blood, buccal epithelial cells, semen and forensic stains for PCR.* Rudbeck, L. e Dissing, J. 588–592, s.l. : BioTechniques, 1998, Vol. 25.
5. *A new DNA extraction method by controlled alkaline treatments from consolidated subsurface sediments.* Kouduka, M., et al. 47–54, s.l. : FEMS Microbiol. Lett, 2012, Vol. 326.

HAZARD SYMBOLS

Pictogram	
Warning	Danger
Hazard statements	
H225	Easily flammable liquid and vapours.
H319	Causes severe eye irritation
Precautionary statements	
P210	Keep away from heat sources, hot surfaces, sparks, open flames or other sources of ignition. Don't smoke
P233	Keep the container tightly closed
P240	Ground and ground the enclosure and device recipient
P241	Use electrical/ventilation/lighting systems explosion test.
P242	Use non-sparking utensils.
P305 + P351 + P338	IN CASE OF EYE CONTACT: Rinse thoroughly for several minutes. Remove any contact lenses if it is easy to do so. Continue rinsing.
Additional Risk Descriptions	None

DESCRIPTION OF SYMBOLS

SYMBOLS	DESCRIPTION
LOT	Batch number
REF	Catalog Number
	Expiry date
	Manufacturer
	Refer to the instructions for use
	Sufficient content for <n> tests
	Recommended Temperature Limits
	Do not use if the packaging is damaged
CE	European Conformity Mark

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