

Instructions for Use

Life Science Kits & Assays



innuSPEED Soil DNA Kit Advanced - PP Mini



Innuscreen
innovative
Sensor
Technology

Order No.:

845-PS-0150016 16 reactions
845-PS-0150016 96 reactions

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1 Introduction

1.1 Intended use

The innuSPEED Soil DNA Kit Advanced – PP Mini is designed for the automated isolation of DNA from soil samples using the PurePrep Mini instrument.

The extraction procedure is based on new patented chemistry that delivers high-quality DNA from wide range of soil types. Optimized buffer systems and advanced purification steps ensure exceptional DNA purity, stable and reproducible PCR performance, and reliable yields – even from highly inhibitory soil matrices.

The extraction procedure takes place on the magnetic particle processor PurePrep Mini and allows the parallel and flexible extraction of 1 up to 16 samples. The procedure begins with a highly efficient external homogenization step. Afterwards, the homogenized samples are transferred into a DW Strip or DW Plate (available separately). The following extraction process runs automatically on the PurePrep Mini.

The extraction is based on binding DNA to surface-modified magnetic particles. After several washing steps the nucleic acids are eluted from the magnetic particles with RNase-free water and are ready for different downstream applications like PCR, qPCR, Digital PCR or Nanopore Sequencing.

The extraction chemistry in combination with the PurePrep Mini is optimized to get maximum yield and quality. The kit is intended for use by professional users.

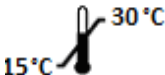
CONSULT INSTRUCTION FOR USE



This package insert must be read carefully before use. Package insert instructions must be followed accordingly. Reliability of results cannot be guaranteed if there are any deviations from the instructions in this package insert.

1.2 Notes on the use of this manual and the kit

For easy reference and orientation, the manual and labels use the following warning and information symbols as well as the shown methodology:

Symbol	Information
	REF Catalogue number.
	Content Contains sufficient reagents for <N> reactions.
	Storage conditions Store at room temperature or shown conditions respectively.
	Consult instructions for use This information must be observed to avoid improper use of the kit and the kit components.
	Expiry date
	Lot number The number of the kit charge.
	Manufactured by Contact information of manufacturer.
	For single use only Do not use components for a second time.
	Note / Attention Observe the notes marked in this way to ensure correct function of the kit and to avoid operating errors for obtaining correct results.

The following systematic approach is introduced in the manual:

- The chapters and figures are numbered consecutively.
- A cross reference is indicated with an arrow (e.g. → „Notes on the use of this manual and the kit“ p. 3).
- Working steps are numbered.

2 Safety precautions

NOTE

Read through this chapter carefully before use to guarantee your own safety and a trouble-free operation.

Follow all the safety instructions explained in the manual, as well as all messages and information which are shown.

All due care and attention should be exercised in handling the materials and reagents contained in the kit. Always wear gloves while handling these reagents and avoid any skin contact! In case of contact, flush eyes or skin with a large amount of water immediately.



FOR SINGLE USE ONLY!

This kit is made for single use only!

ATTENTION!

Don't eat or drink components of the kit!

The kit shall only be handled by educated personnel in a laboratory environment!

If the buffer bottles are damaged or leaking, wear gloves and protective goggles when discarding the bottles in order to avoid any injuries. This kit can be used with potentially infectious samples. Therefore, all liquid waste must be considered as potentially infectious and must be handled and discarded according to local safety regulation.

Please observe the federal, state and local safety and environmental regulations. Follow the usual precautions for applications using extracted nucleic acids. All materials and reagents used for DNA or RNA isolation should be free of DNases or RNases.

ATTENTION!

Do not add bleach or acidic components to the waste after sample preparation!

NOTE

Emergency medical information in English and German can be obtained 24 hours a day from:

Poison Information Center, Freiburg / Germany

Phone: +49 (0)761 19 240.

For more information on GHS classification and the Safety Data Sheet (SDS) please contact sds.innu@ist-ag.com.

3 Storage conditions

All kit components are shipped at ambient temperature.

Upon arrival, store **MAG Suspension F** at 4 °C to 8 °C.

All other components of the kit should be stored dry at room temperature (15 °C to 30 °C). When stored at room temperature, the kit is stable until the expiration date printed on the label on the kit box.

If there are any precipitates within the provided solutions solve these precipitates by careful warming. Before every use make sure that all components have room temperature.

4 Functional testing and technical assistance

The IST Innuscreen GmbH guarantees the correct function of the kit for applications as described in the manual. This product has been produced and tested in an ISO 13485 certified facility.

We reserve the right to change or modify our products to enhance their performance and design. If you have any questions or problems regarding any aspects of the **innuSPEED Soil DNA Kit Advanced - PP Mini** or other IST Innuscreen GmbH products, please do not hesitate to contact us. For technical support or further information in Germany please contact info.innu@ist-ag.com. For other countries please contact your local distributor.

5 Product use and warranty

The kit is not designed for the usage of other starting materials or other amounts of starting materials than those referred to in the manual (→ "Product specifications" p. 7). Since the performance characteristics of IST Innuscreen GmbH kits have only been validated for the application described above, the user is responsible for the validation of the performance of IST Innuscreen GmbH kits using other protocols than those described below. IST Innuscreen GmbH kits may be used in clinical diagnostic laboratory systems after the laboratory has validated the complete diagnostic system as required by CLIA' 88 regulations in the U.S. or equivalents in other countries.

All products sold by IST Innuscreen GmbH are subjected to extensive quality control procedures and are warranted to perform as described when used correctly. Any problems should be reported immediately.

NOTE

This kit is for research use only!

6 Kit components

6.1 Components included in the kit

	Σ 16	Σ 96
REF	845-PS-0150016	845-PS-0150096
Lysis Tube B 2.0	16 pcs	96 pcs
Grinding Buffer S	15 ml	80 ml
Grinding Enhancer S	2 ml	9 ml
Precipitation Buffer S	3 ml	15 ml
MAG Suspension F	0.25 ml	1.1 ml
Binding Solution V	10 ml	60 ml
Washing Buffer S (conc.)	30 ml	125 ml
Washing Solution ER	17 ml	85 ml
RNase-free Water	2 x 2 ml	25 ml
Manual	1	1

6.2 Components not included in the kit

- 1.5 ml tubes
- 96 %–99.8 % ethanol (molecular biology grade, undenatured)
- DW Strip / DW Plate / DW Tip Comb (compatible with the PP Mini device)

7 Product specifications

1. Starting material:

- Soil Sample (150 - 250 mg)

2. Time for automated extraction protocol on PurePrep Mini:

- Approx. 31 minutes (excluding external Homogenization and precipitation step)

8 Initial steps before starting

- Add the indicated volume of absolute ethanol to **Washing Buffer S (conc.)** and mix thoroughly. Always keep the bottle firmly closed!

845-PS-0150016	Add 30 ml ethanol to 30 ml Washing Solution S.
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845-PS-0150096	Add 125 ml ethanol to 125 ml Washing Solution S.
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- Avoid freezing and thawing of starting material.
- Centrifugation steps should be carried out at room temperature.

9 Sample Homogenization

1. Weigh up to 250 mg of Soil sample and transfer into a Lysis Tube B 2.0.
2. Add 750 µl Grinding Buffer S and 80 µl Grinding Enhancer S, mix shortly by pulsed vortexing for 5 sec. Incubate at ambient temperature for 2 minutes.
3. Place the Lysis Tube B 2.0 in the Homogenizer and start the homogenization for 1 min.

NOTE

For Homogenization process use commercially available Homogenizers (e.g. SpeedMill, Precellys, Fastprep, Bead Raptor etc.).

The homogenization process can be changed and optimized depending on the homogenizer used. The optimal duration and intensity of homogenization depends on which kind of homogenizer is used.

4. Remove the Lysis Tube B 2.0 from the Homogenizer and centrifuge the Lysis Tube B 2.0 at max. speed for 2 min
5. Open the Lysis Tube B 2.0 and transfer the supernatant carefully into a new 2.0 ml reaction tube. Add 120 µl Precipitation Buffer S and vortex shortly for 5 s. Leave the tube for 3 min at ambient temperature.
6. Centrifuge at maximum speed for 2min. Carefully transfer 500 µl of the supernatant into the first cavity of a DW Plate or the DW Strip.
7. Proceed with "Automated extraction using PurePrep Mini" on p.10.

10 Automated extraction using PurePrep Mini

10.1 Prefilling of the DW Plate or the DW Strips

	1	2	3	4	5	6	7	8	9	10	11	12
A	Sample 1	→				Eluate 1	Sample 9	→				Eluate 9
B	Sample 2	→				Eluate 2	Sample 10	→				Eluate 10
C	Sample 3	→				Eluate 3	Sample 11	→				Eluate 11
D	Sample 4	→				Eluate 4	Sample 12	→				Eluate 12
E	Sample 5	→				Eluate 5	Sample 13	→				Eluate 13
F	Sample 6	→				Eluate 6	Sample 14	→				Eluate 14
G	Sample 7	→				Eluate 7	Sample 15	→				Eluate 15
H	Sample 8	→				Eluate 8	Sample 16	→				Eluate 16

Fig. 1: Schematic illustration of DW Plate

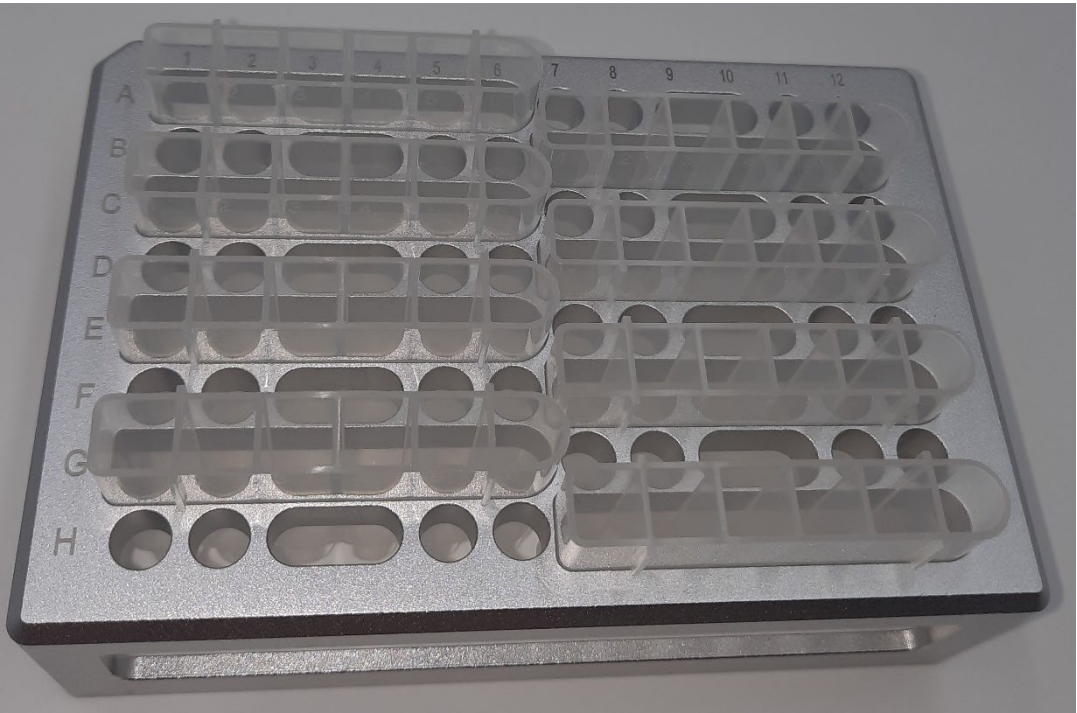


Fig. 2: Arrangement of the DW Strips in Tray

Cavity of 96 DW Plate/ DW Strip	Content
Cavity 1 + 7	500 µl sample supernatant + 10 µl MAG Suspension F (vortex the Beads intensively before use) + 490 µl Binding Solution V
Cavity 2 + 8	800 µl Washing Buffer S
Cavity 3 + 9	800 µl Washing Buffer S
Cavity 4 + 10	800 µl Washing Buffer S
Cavity 5 + 11	800 µl Washing Solution ER
Cavity 6 + 12	100 µl – 200 µl H ₂ O

The prefilling is carried out from left to right as shown in the illustration, Fig. 1. The DW Strips located in the tray are filled in the same way.

10.2 Loading filled Deep Well Plate/Strips to the PurePrep Mini and plug in the Tip Combs

NOTE

- When using a strip (strips), the strip is inserted into the metal tray. In total, a maximum of 8 strips can be used in one extraction-run.
- When working with strips, only every other tip of the tip comb will be used for extraction.

Left tray side: Tip 1, 3, 5, 7

Right tray side: Tip 2, 4, 6, 8.

- It is recommended to mark the tips used for the extraction so that they are not used more than once.

1. Select the protocol

“SOIL1” and start the run.

2. After finishing the extraction protocol, the Cavities 6 and 12 contain the isolated DNA.
3. Transfer the DNA into a fresh 1.5 ml Tube.

IMPORTANT NOTE

After finishing the extraction protocol, the last cavity of the Plate/Strip contains the isolated DNA. Store the DNA under adequate conditions. We recommend storing the extracted DNA at +4 °C to +8 °C. For long-term storage –22 °C to –18 °C is recommended.

11 Troubleshooting

Problem / probable cause	Comments and suggestions
Low amount of extracted DNA	
Insufficient homogenization	Increase homogenization time and/or speed.
Eluate volume too high	Decrease the eluate volume. The suggested eluate volume is 100–200 µl. Please note that lowering the eluate volume will not increase the yield proportionally!
Inadequate extraction	Presence of inhibiting substances in the starting material. Please use the kit only for samples that match the requirements declared in "Product specifications".
Carryover of Magnetic particles	
Eluate contains carryover of magnetic particles	Place the plate on a magnet or centrifuge the plate at maximum speed for 3 minutes. Pipet the supernatant with DNA into a new plate or Elution vessels

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