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KYLT[®] SALMONELLA SPP. 2.0

**REAL-TIME PCR DETECTION KIT
FOR SALMONELLA SPP.**

i For *in vitro* diagnostics only.

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Passion for Innovation



KYLT[®]

SALMONELLA SPP. 2.0

Real-Time PCR Detection of *Salmonella* spp.

1. GENERAL

- Kylt[®] Salmonella spp. 2.0 Real-Time PCR Detection Kit (in the following Kylt[®] Salmonella) detects *Salmonella* spp. in food samples (raw meat and poultry and ready to cook meat and poultry products), environmental samples and primary production samples including veterinary samples such as boot swabs, feces, dust samples, tissue samples from affected organs, swabs from such tissues and sampling material derived from cultural processes with the aforementioned samples (e.g. cultural harvest) of poultry, swine and cattle. Please refer to chapter 6. "Supplementary information" or the validation report for detailed information about the categories and sample matrices that have been covered in the validation.
- Kylt[®] Salmonella comprises all reagents and controls for subsequent detection of bacterial DNA of *Salmonella* spp.. Reagents necessary for sample preparation from cultured materials (optionally) can be purchased individually. After bacterial pre-enrichment and DNA preparation, *Salmonella* spp. is qualitatively detected via specific and real-time detected amplification of a *Salmonella* spp. target gene. This way, results can be achieved within a few hours after bacterial enrichment.
- The qualitative testing of Kylt[®] Salmonella is based on a duplex Real-Time PCR system: In one reaction setting the target genes for *Salmonella* spp. as well as for the included Internal Amplification Control are amplified in parallel by respective primer pairs in the Polymerase Chain Reaction (PCR). Amplified target gene fragments are detected via fluorescently labeled probes during the PCR reaction in real-time (Real-Time PCR). The probes specific for detection of amplified *Salmonella* spp. and Internal Amplification Control target genes are labeled with FAM and HEX, respectively, and their emitted fluorescence is separately optically measured by the Real-Time PCR thermal cycler. By means of both individual analyses in one reaction vessel per sample and the Negative and Positive Control per run the *Salmonella* spp.-specific state of a sample can be evaluated in the end.
- This kit was developed for use by trained laboratory personnel following standardized procedures.

2. REAGENTS AND MATERIALS

- Kylt[®] Salmonella spp. 2.0 Real-Time PCR Detection Kit contains the following reagents:

Reagent	Colour of Lid	100 Reactions Article no. 31302	Storage conditions
Reaction-Mix	● yellow	4× 450 µl	≤ -18 °C
Positive Control	● red	2× lyophilizate (final 50 µl each)	≤ -18 °C *
Negative Control	● blue	1× 1 ml	≤ -18 °C

* Alternative storage conditions: +2 °C to +8 °C lyophilised, +2 °C to +8 °C rehydrated (max. 2 days), ≤ -18 °C rehydrated (max. 6 months)

- Kylt® DNA Extractionmix II (art. no. 31398) as well as other Kylt® Purification Products can be ordered separately - see chapter "7. Related and Accessory Products".
- The kit is to be used within the indicated shelf life. If occasional processing of few samples is expected, you may prepare aliquots of the **Reaction-Mix** before storage at -18 °C to -20 °C. Avoid repeated freezing and thawing of the kit or its components. Recommendation: limit freeze-thaw cycles to 3. Reaction-Mix has to be stored dark, do not expose to direct sunlight!
- Before the first use the lyophilized **Positive Control** is rehydrated: 50 µl of the Negative Control are added to the Positive Control, briefly incubated at room temperature and mixed thoroughly by repeated vortexing. Storage of aliquots with 5 - 10 µl volume (depending on the expected number of Positive Control reactions per kit) at -18 °C to -20 °C is recommended.

3. EQUIPMENT AND REAGENTS NOT INCLUDED

- This kit can be used on all commercially available Real-Time PCR thermal cyclers that are able to detect the emitted fluorescence of fluorescent dyes FAM (emission 520 nm) and HEX (also JOE/VIC, emission 550 nm). Note that default normalization option against ROX (e.g. using ABI cyclers) must be deactivated.
- Apart from the disposables, the following devices are needed (not included in this kit):
 - DNA(/RNA) preparation kit incl. respective accessories, the Kylt® DNA Extractionmix II (art. no. 31398) as well as other Kylt® Purification Products can be ordered separately - see chapter "7. Related and Accessory Products"
 - Incubators for sample pre-enrichment ($+37 \pm 1 \text{ °C}$ & $+41.5 \pm 1 \text{ °C}$)
 - Table top microcentrifuge
 - Dry heating block ($+100 \text{ °C} \pm 3 \text{ °C}$)
 - Vortex
 - Magnetic stirrer
 - Micropipettes volume range 1 – 1000 µl
 - Centrifuge for PCR tubes or plates
 - Real-Time PCR thermal cycler
- We recommend to exclusively using certified Nuclease-free materials and powder free protective gloves. Pipette tips have to be changed between samples to avoid cross contamination. Gloves have to be changed frequently, especially after spillage or other suspected contamination.

4. CONTROL REACTIONS

- The **Positive Control** allows for control of the specificity and efficiency of the reagents and the reaction itself, including the performance of the Real-Time PCR and of the Real-Time PCR thermal cycler.
- The **Negative Control** allows for exclusion of contaminations. The sample testing is only valid if both, Positive and Negative Controls, are used and verified for validity in every Real-Time PCR run.
- The **Internal Amplification Control** is included in the Reaction-Mix in a defined copy number; it is co-amplified (channel HEX) with every single reaction to detect possible inhibitory effects of the DNA preparation on the Real-Time PCR itself and thus to verify true-negative results.

5. PROTOCOL (see also "Protocol At A Glance" at the end of this Direction For Use)

- The overall protocol of this *Salmonella* spp. analysis consists of the following steps:
 1. Cultural pre-enrichment
 2. DNA preparation
 3. PCR Setup and Amplification
 4. Analysis

5.1. Cultural Pre-Enrichment

- The sample preparation and handling should follow the rules of Good Laboratory Practice with sterile instruments to avoid external contamination. Country specific regulations regarding sampling, pooling and subsampling should be followed.
- Pre-enrichment should be performed according to ISO 6579, FDA-BAM or USDA. In general 25 g or 375 g of sample is analyzed in a 1:9 sample/broth ratio (25 g in 225 ml broth or 375 g in 3375 ml broth, e.g. Buffered Peptone Water, BPW). Pre-enrichment of environmental samples and primary production samples should be performed according to ISO 6887 (please refer to chapter 6. "Supplementary information" or the validation report for detailed information about the specific preparations used in the validation study).
- To enable sufficient propagation of potentially present *Salmonella* during incubation time, the used pre-enrichment medium has to be pre-warmed to 37°C prior to the start of incubation time. The pre-enrichment is incubated at $+37 \pm 1$ °C for 18 ± 2 hours without agitation.
- **Recommendation:** To retain the sample material for future microbiological or biomolecular analysis at least 3 ml of the supernatant of the pre-enrichment are transferred to a sterile test tube with sterile transfer pipettes and the initial vessel (e.g. Stomacher bag) is discarded. The 3 ml in the tube can be used for storing the sample at +2 °C to +8 °C for at most 72 hours. Storage in test tubes is just a recommendation, the 1 ml sample for DNA extraction may also be transferred directly from the initial vessel into a conical screw cap tube and the vessel may be retained.
- **Attention:** Mixing of the pre-enrichment after incubation by shaking or any other agitation must be avoided! Avoid transferring solid or greasy debris. An aliquot should be taken directly below the surface, avoiding floating particles. For samples that are too difficult to be separated from the debris after incubation, Stomacher bags with filters should be used for pre-enrichment.
- A 1 ml aliquot is transferred from the test tube into a sterile, conical screw cap microcentrifuge tube. The remaining pre-enrichment in the test tube or initial vessel (e.g. Stomacher bag) can be stored for potential subsequent cultural examination.
- Certain sample matrices, such as turf- or soil-containing sock swabs and feces with a high concentration of humic acids or polyphenolic substances (e.g. feces from cattle), are known to have a potential inhibitory effect on the Real-Time PCR. In such cases a second enrichment step is performed with the pre-enrichment and the entire process of DNA extraction and Real-Time PCR is repeated (see details below). Or, for samples known to have an inhibitory effect on the Real-Time PCR, the second enrichment step may directly be performed subsequently to the pre-enrichment step without Real-Time PCR-testing in between.
- For the second enrichment step the selective medium Rappaport-Vassiliadis-Soy Broth (RVS) is used. The readily incubated pre-enrichment (sample in e.g. BPW) is diluted at a ratio of 1:100 in RVS (e.g. 100 µl in 10 ml). The selective enrichment in RVS broth is incubated for at least 5 hours at $+41.5 \pm 1$ °C without agitation. Longer incubation times can be implemented if required or if more suitable for the daily lab routine.

COLONY MATERIAL FROM SOLID MEDIA

- To analyze pure or mixed cultures on solid media, a small portion of an individual colony or mixed colony material can be transferred to a sterile, conical screw cap tube.
 - The DNA Extractionmix II is stirred on a slow moving magnetic stirrer, so that aliquots of a homogenous suspension can be taken. The pellet is resuspended by repeated up-and-down pipetting in 500 µl of DNA Extractionmix II. Use a 1000 µl pipette with filtered tips. Avoid formation of bubbles and aerosols and proceed with step 5 in 5.2.A "DNA Preparation Using the optionally included DNA Extractionmix II".
 - Alternatively the purification of the isolate can be carried out using Kylt® RNA/DNA Purification (see also 5.2.B "DNA Preparation Using Kylt® RNA/DNA Purification") or using manually or (semi-)automated DNA-purification systems (see also 5.2.C "DNA Preparation Using Manual or (Semi-)Automated DNA-Purification Systems").

5.2. DNA Preparation

- DNA preparation can be carried out using either the optionally available DNA Extractionmix II (proceed with chapter 5.2.A), manual DNA-purification using Kylt® RNA/DNA Purification (proceed with chapter 5.2.B), or other manual or (semi-) automated DNA-purification systems in combination with respective DNA-purification kits (proceed with chapter 5.2.C).

A) DNA PREPARATION USING THE OPTIONALLY AVAILABLE DNA EXTRACTIONMIX II

1. Pre-heat the heating block to set temperature of +100 °C, the block must have an actual temperature of +100 ± 3 °C at use.
2. The aliquoted pre-enrichment in the conical screw cap tube is pelleted by centrifugation at 10.000 g to 12.000 g for five minutes.
3. Discard the supernatant using a 1000 µl pipette tip, not by decantation.
4. The DNA Extractionmix II is stirred on a slow moving magnetic stirrer, so that aliquots of a homogenous suspension can be taken. The pellet is resuspended by repeated up-and-down pipetting in 200 µl of DNA Extractionmix II. Use a 1000 µl pipette with filtered tips. Avoid formation of bubbles and aerosols.
5. Screw cap tight, vortex thoroughly and incubate for 10 min to 15 min at +100 ± 3 °C.
6. Vortex sample thoroughly and centrifuge at 10.000 g to 12.000 g for five minutes; the supernatant is the DNA extract and can be used for Real-Time PCR immediately. Short-term storage (few hours) of the DNA extract at +2 °C to +8 °C is possible. For longterm storage of the DNA extract at -18 °C to -20 °C, transfer supernatant to a new (screw cap) tube. Before next use in Real- Time PCR, incubate DNA extracts that were stored at -18 °C to -20 °C for few minutes at +100 ± 3 °C, vortex and spin down by pulse-centrifugation.

B) DNA PREPARATION USING KYLT® RNA/DNA PURIFICATION

- For detailed information, please refer to the Direction For Use of Kylt® RNA/DNA Purification.

C) DNA PREPARATION USING MANUAL OR (SEMI-)AUTOMATED DNA-PURIFICATION SYSTEMS

- The DNA preparation from samples can also be performed manually or on (semi-)automated DNA-purification systems using commercially available DNA-purification kits. For the specific purification protocols please refer to the manual of the respective DNA purification kit appropriate for the sample matrix which has to be analyzed.

5.3. PCR Setup and Amplification

- Reaction-Mix and Negative Control are vortexed and pulse-centrifuged before each use.
- Determine the number of PCR-reactions needed: number of samples + Positive and Negative Control.
- Pipette 16 µl of **Reaction-Mix** to each PCR-reaction tube / each PCR-plate well ("cavity"). Keep exposure of Reaction-Mix to light as short as possible!
- Add 4 µl of **Negative Control** to corresponding cavity and seal.
- Add 4 µl of **sample DNA preparation** to the corresponding cavity and seal. In case of manually prepared DNA-extract solely use clear supernatant, avoid transferring debris.
- Once all sample cavities are sealed, 4 µl of the **Positive Control** are added to the corresponding cavity and sealed. Vortex and pulsecentrifuge the rehydrated Positive Control before each use.
- Avoid formation of bubbles when pipetting Reaction-Mix, samples and controls. It is recommended to always briefly pulse-centrifuge cavities before the PCR run.
- Place cavities in Real-Time PCR thermal cycler and amplify using the following parameters:

Kylt® Profile II

Step No	Description	Temperature	Duration	
1	Activation of Polymerase	95 °C	10 min	
2	Denaturation	95 °C	15 sec	} 42 cycles
3	Annealing & Extension	60 °C	1 min	
4	Fluorescence Detection	channels FAM and HEX		

- Alternatively, the following standard Kylt® Profil I can be used to combine Kylt® Salmonella spp. 2.0 with other Kylt® Real-Time-(RT-) PCR Detection products.

Kylt® Profil I

Step No	Description	Temperature	Duration
1	Reverse Transcription	50 °C	10 min
2	Activation of Polymerase	95 °C	1 min
3	Denaturation	95 °C	10 sec
4	Annealing & Extension	60 °C	1 min
5	Fluorescence Detection	channels FAM and HEX	

} 42 cycles

- These amplification parameters allow the combination of this Kylt® Salmonella spp. 2.0 Real-Time PCR with other Kylt® Real-Time PCRs for detection of bacteria or viruses. When combining several detection tests, make sure all necessary channels are used!
- Please follow the specific instructions of your Real-Time PCR thermal cycler as recommended by the manufacturer.

5.4. Analysis

GENERAL

- The data of the amplification reactions can automatically be processed using specific software for the Real-Time PCR thermal cycler. Alternatively, the thresholds can be set manually using either the linear or the logarithmic view. The threshold should cross the HEX-curve of the Negative Control and the FAM-curve of the Positive Control, respectively, in the exponential area of its slope (see exemplary figures below). By setting the thresholds, the crossing points with the HEX- and FAM-curves determine the respective “cycle of threshold” (Ct), which is negatively correlated with the initial concentration of copies of the target genes in the Real-Time PCR reaction.

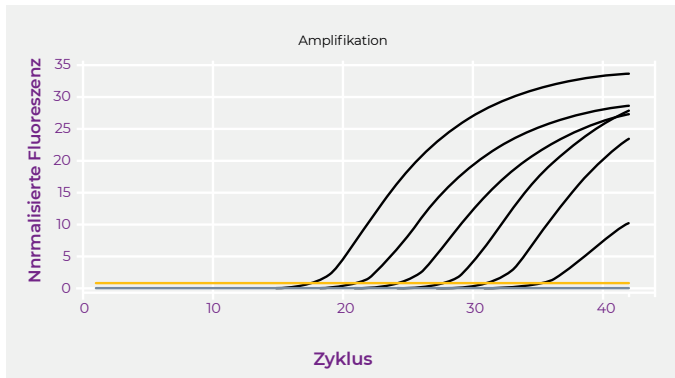


Figure 1: Linear view

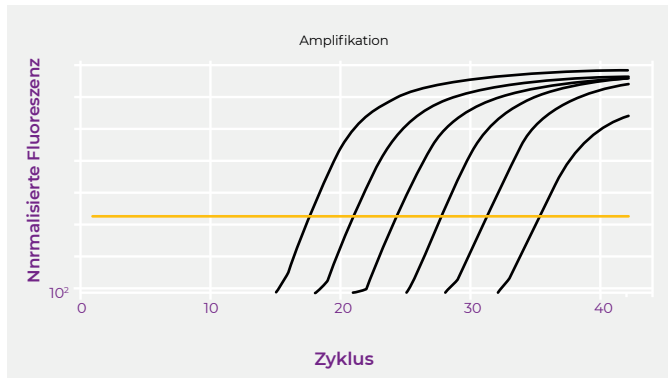


Figure 2: Logarithmic view

- Only curves with the typical exponential amplification, meaning the curve of the raw data shows a flat baseline at the beginning in the linear view, followed by a clear (exponential) slope in fluorescence and possibly reaching a plateau-phase, should be regarded as positive.
- For the test analysis the validity of the run is determined with the Negative and Positive Controls. After that the validity of each single sample is verified by the Internal Amplification Control reaction and finally their *Salmonella* spp.-specific status is checked.

TEST EVALUATION – CONTROL REACTIONS

- The Real-Time PCR test run is only valid if the curves of the control reactions can be evaluated as follows:

Control Reactions	Channel	
	HEX	FAM
Negative Control	positive	negative
Positive Control	positive	positive

- The FAM-Ct-value of the Positive Control has to be > 15 and ≤ 35.
- The HEX-Ct-values of the Positive and Negative Control have to be ≤ 40.

5.5. Test Evaluation – Samples

Target	Channel	Signal		
Internal Control	HEX	positive	positive / negative	negative
<i>Salmonella</i> spp.	FAM	negative	positive	negative
The sample is <i>Salmonella</i> spp.		negative	positive	inhibited

- A sample is **negative** for *Salmonella* spp. if its HEX-curve is positive, but its FAM-curve is negative.
- A sample is **positive** for *Salmonella* spp. if its FAM-curve is positive, independent of the HEX-curve.
- A sample is **inhibited** if neither the FAM-curve nor the HEX-curve are positive.
- **Recommendation:** In case of an inhibited sample the PCR can be repeated with a 1/10 dilution of the DNA extract. The Negative Control is used as the diluting agent. If the sample is still inhibited after running the PCR with the diluted sample or as a direct alternative, the pre-enrichment may be incubated for at least 5 hours in RVS (for details see chapter 5.1. "Cultural Pre-Enrichment"). By this step potentially present ingredients in the pre-enrichment that have inhibitory effects on the effectiveness of the Real-Time PCR are diluted and potentially present *Salmonella* are selectively propagated. After the second enrichment the entire process of DNA preparation and Real-Time PCR is repeated (see chapters 5.2. "DNA Preparation" and 5.3. "PCR Setup and Amplification").
- Presumptive results should be confirmed by methods other than the Kylt® *Salmonella* spp. 2.0 (DNA Extraction and) Real-Time PCR Detection kit. It is recommended to use the cultural reference method for *Salmonella* spp. (ISO 6579) to confirm the results. Briefly, a subculture of the pre-enrichment is prepared in RVS broth or on MSR/V agar as well as in MKTTn broth, depending on the sample type. Subsequently, the incubated samples are streaked on XLD agar and one additional selective agar. Typical colonies are confirmed by serological or biochemical methods. For details please refer to the current version of ISO 6579.

6. SUPPLEMENTARY INFORMATION

- Kylt® *Salmonella* spp. 2.0 has been validated in a harmonized MicroVal/AOAC RI/Nordval validation in combination with the Kylt® DNA Extractionmix II (Art. No. 31398) as well as with Kylt® RNA/DNA Purification (Art. No. 31315) as an alternative DNA purification method. The validation covers samples from raw meat and ready to cook meat products (25 g and 375 g), raw poultry and ready to cook poultry products (25 g and 375 g), environmental samples (such as process water or swab samples) as well as primary production samples (such as boot socks, faeces or rectal swabs). The performance of the kit was tested with the following Real-Time PCR instruments: ABI 7500 platform (Life Technologies), CFX 96 and CFX 384 (BioRad), Rotor-Gene Q (Qiagen), Rotor-Gene 3000 and 6000 (Corbett Research), AriaMx Pro (Agilent), LightCycler® 96 and 480 (Roche), Mic (BMS).

- The sample categories tested in the validation are listed in the table below:

Category	Type	Test portion size
Raw meat and ready to cook meat products	Fresh meats (unprocessed)	25 g & 375 g
	Ready to cook (processed)	
	Ready to eat and Ready to reheat products	
Raw poultry and ready to cook poultry products	Fresh meats (unprocessed)	25 g & 375 g
	Ready to cook (processed)	
	Ready to eat and Ready to reheat products	
Environmental	Dust and residues	25 g
	Cleaning and process waters	
	Surface samples	
Primary production	Poultry, Swine/ Cattle	25 g
	Environmental samples	
	Ready to cook (processed)	
	Ready to eat and Ready to reheat products	

- The sample categories and test portion sizes tested during validation with the respective extraction methods and temperature profiles:

Alternate method	Raw and ready to cook poultry (combined category)	Raw and ready to cook meat (combined category)	Environmental samples	Primary production samples
Kylt® DNA Extractionmix II profile I	25 g	25 g	25 g	25 g
Kylt® DNA Extractionmix II profile II	25 g & 375 g	25 g & 375 g	25 g	25 g
Kylt® RNA/DNA Purification profile I	25 g	25 g	25 g	25 g (after 5h RVS only)
Kylt® RNA/DNA Purification profile II	25 g & 375 g	25 g & 375 g	25 g	25 g

- Specific sample preparations used in the validation study:

Sample type	Category	Appropriate part of ISO 6887 to be used	Preparation used in validation
All food products within the category	Raw meat and ready to cook meat products, Raw poultry and ready to cook poultry products	1 and 2	For 25 g samples Add 225 ml BPW to 25 g samples, dilution ratio = 1:10 For 375 g samples Alternative method 1500ml BPW to 375 g samples, dilution ratio 1:5
Carcass swabs	Raw meat and ready to cook meat products	1 and 2	Add swab to 50 ml tube containing 40 ml BPW and make sure that the whole sample is submerged. Mix/ shake well before enrichment
Process water poultry, cattle and swine	Environmental	1 and 2	Samples <100 ml added to an equal volume of double strength BPW
Dust wipe samples	Environmental	N/A	Make sure that the whole sample is submerged in BPW. Mix/ shake well before enrichment
Sponge samples	Environmental	N/A	Make sure that the whole sample is submerged in BPW. Mix/ shake well before enrichment
Dust and residue swabs	Environmental	N/A	Add swab to 50 ml tube containing 40 ml BPW and make sure that the whole sample is submerged. Mix/ shake well before enrichment

Sample type	Category	Appropriate part of ISO 6887 to be used	Preparation used in validation
Swabs stainless steel, plastic surface, ceramic and rubber	Environmental	6	Add swab to 50 ml tube containing 40 ml BPW and make sure that the whole sample is submerged. Mix/ shake well before enrichment
Rectal swabs	Primary production	6	Add swab to 50 ml tube containing 40ml BPW and make sure that the whole sample is submerged. Mix/ shake well before enrichment
Faeces pig, chicken and turkey, as well as gut contents	Primary production	6	Mix gently and add 225 ml BPW to 25 g samples
Bootsocks	Primary production	6	Add at least 225 ml BPW and make sure that the whole sample is submerged
Hatchery samples – basket liners	Primary production	6	Samples should be at least 1 m surface area. Add 1 to 2 L of BPW (pre- warmed to at least room temperature, but preferably 37°C)
Transport truck sampling, waiting area swab, transport cage debris	Primary production	6	Add at least 225 ml BPW and make sure that the whole sample is submerged. Mix/ shake well before enrichment
Organs lymph nodes	Primary production	6	Macerate lymph nodes by hammering a strong sterile plastic bag containing the samples. Add 9 ml per g of sample

- In the validation study, meat, poultry and environmental samples were incubated at 37°C for 16h which is the shortest incubation time within the tolerance of 18h ±2h required for ISO 6579-1:2017.
- For more information about the validation including cyclers and samples tested, please refer to **MicroVal Certificate No. 2017LR78**.

7. RELATED AND ACCESSORY PRODUCTS

Product name	Article no.	Content	Description
Kylt® DNA Extractionmix II	31398	100	Simplified and economic DNA extraction.
Kylt® RNA/DNA Purification	31315	50	Combined RNA and DNA purification from veterinary samples (spin-column based).
Kylt® RNA/DNA Purification HTP	31575	4 x 96	Magnetic bead based combined RNA and DNA purification kit for veterinary diagnostic samples. Suitable for Kylt® Purifier and Kylt® Purifier 48.
Kylt® Purifier	31436	1	Purification system for magnetic bead based kits. Up to 96 samples are processed in under 30 minutes. Intended for high-throughput laboratories.
Kylt® Purifier 48	31748	1	Purification system for magnetic bead based kits. Up to 48 samples are processed in under 30 minutes. Intended for low to medium throughput laboratories.
Kylt® Purifier Spin Tips	31434	5	Plate with 96 separate spin tips, used by the Kylt® Purifier to mix the well contents by stirring. Sufficient for 480 samples.
Kylt® Purifier Plates	31435	20	Plates to be used for the several reactions and reagents during automated nucleic acid purification. Sufficient for 320 to 480 samples (depending on device and protocol).

8. ORDERING INFORMATION

General terms and conditions of SAN Group Biotech Germany GmbH apply (www.anicon.eu). For a fast and efficient service please send your order to orders.kylt-de@san-group.com and please provide the following information:

- Delivery and invoice address
- Purchaser contact telephone number
- End user name and telephone number (if different)
- Purchase order number
- Product name and catalogue number
- Quantity and size of products
- Indicate if your account is VAT exempt

9. REVISION HISTORY

Revision	Status	Amendments
Rev005	Jan 2026	<ul style="list-style-type: none">- New layout / design, changed manufacturer name and TÜV-logo;- Chapter 1. "General": Specification of the intended use and validated sample matrices;- Chapter 2. "Reagents and Materials": adaptation of the table and changed recommended storage conditions, added recommendation for freeze-thaw cycles;- Chapter 5.5 "Test Evaluation – Samples": addition of the diluting agent in the "Recommendation" section; excluded note about the software used for sample data entry and documentation;- Chapter 6. "Supplementary information": Detailed specification of sample categories tested and sample preparations used in the validation;- addition of chapter 7., 8. and 9., simplified "Protocol at a glance".

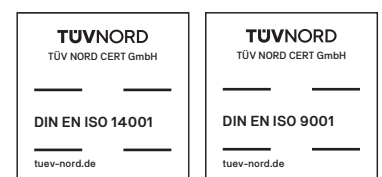
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Development, production and distribution of in vitro diagnostics is certified to ISO 14001:2015 and ISO 9001:2015.

Kylt® is a registered trademark.

For veterinary use and food testing applications only. For *in vitro* use only. Regulatory requirements vary by country, not all of the products described herein may be available in your geographic area.

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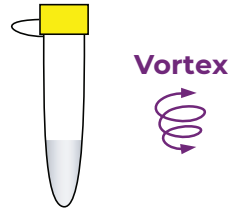


PROTOCOL AT A GLANCE

Real-Time PCR Setup

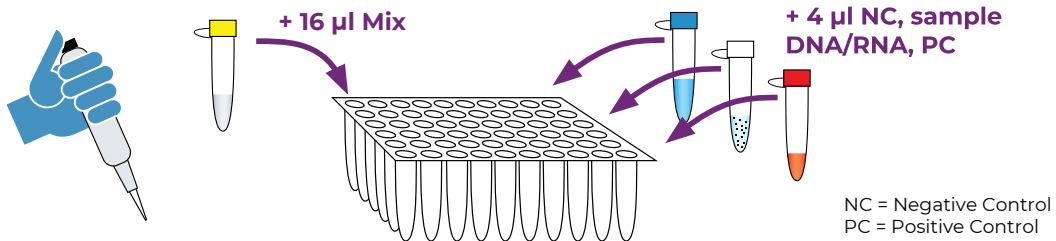
1

Pulse-vortex and spin down



2

Dispense Reaction-Mix (or RTU-Mix) and add 4 μ l NC, sample DNA/RNA, PC



3

Seal cavities, spin down (recommended), and start cycler



4

Analysis

