

Instructions for use

FTA® Cards Made by Whatman®

Description:

FTA cards are designed for room temperature collection, shipment. archiving, and purification of nucleic acids from a wide variety of biological samples for PCR analysis. These include (but are not limited to) blood, buccal cells, tissue, cultured cells, microorganisms, and plant tissue. FTA Cards are impregnated with a patented chemical formula that lyses cell membranes and denatures proteins upon contact. Nucleic acids are immobilized and protected from UV damage and microbial and fungal attack. Infectious pathogens in samples applied to FTA Cards are rendered inactive upon contact. FTA Cards are available in several formats such as the Classic Card, Mini Card, Micro Card, and Gene Card. Indicating FTA Cards turn from pink to white upon sample application and are recommended for colorless samples. To use FTA Cards, simply apply sample (liquid or pressed tissue), air dry at room temperature, then remove a small disc (1.2 mm or 2.0 mm, depending on application). The disc is then washed and used in PCR-based analysis.

Precautions:

Handling: Always wear gloves to avoid contamination of FTA Cards. Follow universal precautions when handling biological specimens.

Storage: Store unused cards in original packaging in a cool, dry clean environment. After applying samples, allow them to dry, then store at room temperature in a dry environment.

INSTRUCTIONS:

<u>Application of Blood Samples</u> (fresh whole blood, or with the anticoagulants: EDTA, sodium citrate, ACD, or heparin):

1. Label the FTA card with the appropriate sample identification.

- Drop the blood (< 125 μl per 1 inch circle, < 75 μl per ¾ inch circle) onto the card in a concentric circular motion within the printed circle area. Avoid "puddling" of the liquid sample, as it will overload the chemicals on the card. Also, do not rub or smear the blood onto the card.
- 3. Samples applied to FTA cards are ready for immediate room temperature storage.
 - **Note**: if samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room temperature prior to punching. Do not heat to shorten the drying period.
- 4. Dried blood spots will appear darker than freshly spotted ones.
- 5. The sample is now ready for downstream processing or archive (see reverse).

Collection and application of buccal cell samples:

- Place the FTA card (Indicating FTA is recommended) on a clean, dry, flat surface. Label the FTA Card with appropriate sample identification.
- 2. Remove one Foam Tipped Applicator from the protective packaging according to instruction.
- 3. Hold the plastic handle of the Applicator, place the foam tip in the mouth and rub one side of the foam tip on the inside of the cheek for 30 sec. Repeat using the opposite side of the foam tip for the other cheek. Run the foam tip along the gum-line and fold of the cheek and under the tongue, soaking up as much saliva as possible. Remove the Applicator from the mouth.
- 4. Lift the paper cover to the Indicating FTA Card to expose the pink sample area. Press the flat surface of the foam applicator tip within the sample circle area. Without lifting the foam tip from the card, squeeze the tip using a side-to-side rocking motion (90° in each direction) 3 times to completely saturate the sample area. Turn the Applicator over and repeat with the other side of the foam tip within the same circle. The sample area will turn white indicating the location of sample.
- 5. If not using the Indicating FTA Cards, circle the area of the sample location with a ballpoint pen or pencil.
- Discard the Applicator according to laboratory procedure.
 Do not place the foam swab into the mouth after it has touched the FTA card.
- 7. If buccal cells are to be applied to more than one FTA circle area, use a new Applicator and repeat steps 1-6.
- 8. Samples applied to FTA cards are ready for immediate room temperature storage.

Note: if samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room

- temperature prior to punching. Do not heat to shorten the drying period.
- 9. The sample is now ready for downstream processing or archive (see reverse).

Application of Tissue/Cell culture samples:

- Tissue culture cells should be applied to FTA Cards at a concentration of >100 cells/µl for DNA analysis and >1000 cells/µl for RNA analysis in media, trypsin, or PBS buffer. Approximately 65 µl of sample will fill a 1 inch printed circle on an FTA Card.
- 2. Samples applied to FTA cards are ready for immediate room temperature storage. Note: If samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room temperature prior to punching. Do not heat to shorten the drying period.
- 3. The sample is now ready for downstream processing or archive (see reverse).

Application of Plant samples:

Direct leaf press:

- Place leaf material directly onto the FTA card. Lay a piece of parafilm over the leaf.
- 2. Apply moderate pounding/pressure to the leaf area with a blunt object such as a tack hammer or pestle.
- 3. When the extract is drawn through to the back of the FTA card the collection process is complete.
- 4. Samples applied to FTA cards are ready for immediate room temperature storage.
 - **Note**: If samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room temperature prior to punching. Do not heat to shorten the drying period.
- The sample is now ready for downstream processing or archive.

Plant Tissue Homogenate:

- 1. Use about 10-20 mg of plant tissue for the homogenate.
- Add PBS buffer to plant tissue using an estimated ratio of 5 parts PBS buffer to 1 part plant tissue. Grind with a pestle unit it is apparent that some plant tissue is homogenized. The homogenate does not have to be smooth in consistency.

- 3. Using a pipette, apply about 25 µl of plant homogenate to each circle on an FTA card.
- 4. Samples applied to FTA cards are ready for immediate room temperature storage.

Note: if samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room temperature prior to punching. Do not heat to shorten the drying period.

The sample is now ready for downstream processing or archive (see below).

Application of Bacterial Samples (for bacterial genomic DNA)

Bacterial colonies:

- Pick one colony from agar, and suspend in 5-10 μl of bacterial culture medium, PBS, or TE¹ buffer (10 mM Tris-HCl, 0.1 mM EDTA, pH 8.0).
- 2. Apply 5-10 µl of bacterial suspension to FTA (indicating FTA Cards are recommended). If applied to non-indicating FTA cards, circle the area of application with a ballpoint pen or pencil.
- 3. Samples applied to FTA cards are ready for immediate room temperature storage.

Note: If samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room temperature prior to punching. Do not heat to shorten the drying period.

4. The sample is now ready for downstream processing or archive (see below).

Overnight Bacterial Cultures:

- Take about 65 µl of overnight culture and apply to FTA (Indicating FTA Card is recommended). If applying to non-Indicating FTA, circle the area of application with a ballpoint pen or pencil.
- 2. Samples applied to FTA cards are ready for immediate room temperature storage.

Note: If samples are to be processed shortly after application on the FTA card, allow the sample to dry for one hour at room temperature prior to punching. Do not heat to shorten the drying period.

The sample is now ready for downstream processing or archive (see below).

Archiving of samples on FTA Cards:

Biological samples applied to FTA cards should be archived at room temperature in a Multi-Barrier Pouch with a desiccant or stored in a humidity-controlled, cool, dry environment. Samples for RNA analysis should be stored at -20 °C or -70 °C for long term storage.

Preparation of Sample DNA for Downstream Analysis:

- Take a sample disk from the desired sample spot using a coring device. For blood samples and bacterial genomic DNA samples, a 1.2 mm disc is recommended. For all other sample types, use a 2.0 mm disc. Please sample disc in a PCR amplification tube.
- 2. Add 200 µl of FTA Purification Reagent (to PCR tube.
- 3. Incubate for 5 minutes at room temperature, (the tube may be given moderate manual mixing if desired).
- 4. Remove and discard all spent FTA Purification Reagent using a pipette.
- 5. Repeat steps 2-4 twice, for a total of three washes with FTA Purification Reagent*.
- Add 200 μl of TE¹ Buffer (10 mM Tris-HCl, 0.1 mM EDTA, pH 8.0) to PCR tube.
- 7. Incubate for 5 minutes at room temperature.
- 8. Remove and discard all spent TE-1 Buffer with a pipette.
- Repeat steps 6-8 once for a total of two washes with TE⁻¹ Buffer.
- 10. Allow disc to dry at room temperature for about one hour, or heat assist the drying of disc at 56 °C for 10 minutes. The FTA disc is now ready for PCR.

If purifying sample disc from a plant source or bacterial culture, only two washes with the FTA Purification Reagent are necessary.

Downstream PCR Applications:

The washes and dried disc is now ready for PCR analysis. Assuming a 25 μ l reaction, we recommend a 1.2 mm disc for blood and 2.0 mm disc for buccal and bacterial samples. The disc is included in the PCR reaction. No alterations in the PCR reaction mix or cycling conditions are required.

For additional protocol information, please contact us under info@carlroth.de, or visit www.whatman.com for applications quide.

FTA® Cards MINI		CL90.1
FTA® Cards MINI indicated		CL91.1
FTA® Pouch MINI		CL92.1
FTA® Cards CLASSIC		CL93.1
FTA® Cards CLASSIC indicated		CL94.1
FTA® Pouch MAXI		CL95.1
FTA® Kit		CL98.1
FTA® Purification Reagent		CL99.1
FTA® Foam Tipped Applicator		HP14.1
Harris Uni-Core Punches 1 mm	25 piec.	6729.1
Harris Uni-Core Punches 1.2 mm	4 piec.*	HP15.1
	25 piec.	HP15.2
Harris Uni-Core Punches 2 mm	4 piec.*	HP16.1
	25 piec.	HP16.2
Harris Uni-Core Punches 3 mm	4 piec.*	6798.1
	25 piec.	6798.2
Harris Uni-Core Punches 6 mm	4 piec.*	6799.1
	25 piec.	6799.2

*incl. 2 cutting mats

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