

Sapphire Chip for the naica® system

Instructions for Use

For Research Use Only. Not for use in diagnostic procedures.

Product Name

Sapphire Chip (Ref.C14012)

Resources

Depending on the naica® system configuration, the technical resources for Instructions for Use for the Sapphire Chip are available:

3-color naica® system / Prism3 N10001.3

<https://www.stillatechnologies.com/technical-resources/naica-system-prism3/>

6-color naica® system / Prism6 N20001.6

<https://www.stillatechnologies.com/technical-resources/naica-system-prism6/>



Intended Use

The Sapphire Chip is a component of the naica® system, a high-resolution nucleic acid analysis system based on digital PCR. The Sapphire Chip is intended to partition nucleic acid samples and to contain the partitioned samples on the chip for amplification and detection of nucleic acids. Partitioning is performed by generating water-in-oil droplets. The Sapphire Chip is intended for use by laboratory personnel trained in the techniques of Crystal Digital PCR™.

Composition

Sapphire Chip consumables are packed in specific well-sealed shipping boxes containing in total 12 chips per box (4 plastic trays of 3 chips each).

Each secondary packaging contains:

- 12 Sapphire Chip consumables pre-loaded with proprietary oil and closed with flat white and blue caps;
- at least 12x4 white vented Sapphire Chip (PCR) caps.

Consumables Required but Not Provided

- Reagents or consumables for nucleic acid purification.
- Standard consumables and equipment for PCR mix preparation
- Assay-specific digital PCR reagents, primers and probes.
- Antistatic wetted wipes (ACL Staticide®, Reference: SW12)
 - ➔ The specific product can be ordered directly from the supplier Digi-Key using the reference ST1059-ND. [SW12 ACL Staticide Inc | Static Control, ESD, Clean Room Products | DigiKey](#)
- Precision Wipes (Kimtech™ Science, Reference: 7552, 1 ply, 213x114 mm) can be ordered from standard laboratory suppliers.

Specifications and Performances

General characteristics

The layout of the chips is detailed below (**Figure 1**). Each Sapphire Chip consists of 4 chambers (A to D) and each chamber has one inlet (white flat caps) and one outlet (blue flat caps). Sapphire Chip consumables are identified with a unique serial number (ID number) and this ID can be scanned using a barcode reader. In addition, each chamber can be annotated by writing directly on the sticker.

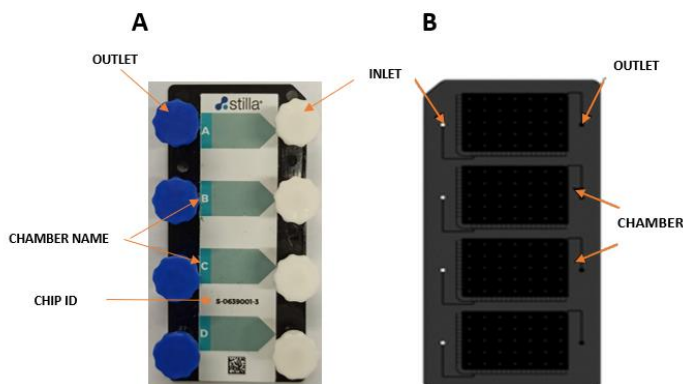


Figure 1: A) Opaque front view, B) and transparent back view of the Sapphire Chip.

Table 1: General characteristics

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General characteristics Sapphire Chip	
Capacity	Up to 4 samples/chip
Input volume	25 μ L/chamber
Droplets per sample	12 900 – 25 800**
Average droplet volume	~ 0,62** nL
Dynamic range*	~ 5,0 log
LOD min (95%)*	down to 0,20**cp/ μ L
LOD max (95%)*	up to 13.000**cp/ μ L
Altitude	Operating at max. 1000m above sea level
naica® system compatibility	Geode (PN H13000-H15000) Prism3 (PN H22000) Prism6 (PN H24000.6)

(*) Analytical specifications are assay dependant; values are theoretical, calculated from the droplet size and droplet volume.

(**) naica® multiplex PCR MIX and naica® PCR MIX.

Note: For more details about the recommended mastermixes for the naica® system, refer to the Geode User Manual available at <https://www.stillatechnologies.com/technical-resources/>.

Caution: Input material for the Crystal Digital PCR™ workflow includes extracted nuclei acid. The purity of the extracted sample may vary depending on the raw material and the implemented extraction protocol. It is recommended to perform validation tests to select a compatible extraction protocol for optimal performance of Crystal Digital PCR™.

DNA samples with ≥ 10 kb average length (e.g., genomic DNA) should be fragmented by restriction digestion. Care must be taken to use restriction enzymes that do not cut within the amplified sequence.

Preventing DNA Contamination

Due to the extraordinary sensitivity of most Taq polymerases, exogenous DNA amplification can occur and ruin an entire experiment (Kwok and Higuchi, 1989). Since many sources of contamination exist such as cross-contamination and previous PCR amplification, adherence to a strict set of protocols combined with some precautions is essential. To limit DNA contamination, the following conventional recommendations for PCR should be followed (not exhaustive):

- Separate areas and dedicate equipment/supplies for sample preparation, PCR setup, PCR amplification, and analysis of PCR products. Wear a clean lab coat and gloves during each step of the experiment.
- Clean laboratory benches and equipment periodically with water/ethanol, or with DNase/RNase away depending on the application (avoid aerosols of cleaning reagents to prevent negative contamination). For the naica® system instruments, please refer to the cleaning sections of the respective user manuals.
Note: Bleach is not recommended; if its use is unavoidable, be sure to abundantly rinse with water after the bleach treatment.
- Keep tubes capped as much as possible.
- Centrifuge tubes after vortexing to limit aerosols.
- Monitor potential contaminants by including a no template control.
- It could be useful to aliquot some reagents to prevent contamination and, at the same time, to reduce repetitive freezing/thaw cycles that might affect their efficiency

Principle of the Procedure

Crystal Digital PCR™ is performed in the Sapphire Chip which contains the samples to be analyzed.

To load the Sapphire Chip, the steps are as follows:

- 1) Open the plastic blister containing 3 chips pre-loaded with proprietary PCR oil and closed with flat caps and remove one Sapphire Chip.
- 2) Apply the antistatic coating:
 - a. Open a pouch of the pre-wetted antistatic wipe (ACL Staticide®).
 - b. Wrap the antistatic wipe around a finger and thoroughly wipe the underside and the edges of one Sapphire Chip to fully coat it with the antistatic treatment.
 - c. Without turning the Sapphire Chip upside down, by lifting the Sapphire Chip visually check that the entire bottom surface of the Sapphire Chip has been fully covered with anti-static solution.
 - d. After applying the antistatic treatment, wipe off excess product with a piece of Precision Wipe (Kimtech™ Science, Reference: 7552).

- e. One ACL Staticide® antistatic wipe can be used for up to 3 chips, if they are all processed directly one after another. If the antistatic wipe appears to be dried out, take a new antistatic wipe to prepare additional chips.
- f. Place the Sapphire Chip back in the plastic blister and apply the antistatic treatment to the next chip.

3) When all the Sapphire Chip consumables from a plastic blister are to be used, keep them inside the blister to proceed to sample loading. The plastic blister can be used to transport the Sapphire Chip consumables. Besides, when less than 3 chips are needed, lay a Precision Wipe on the bench to protect the Sapphire Chip from scratches and dust particles.

4) Disconnect the white flat inlet caps from the inlets of the Sapphire Chip by twisting a ¼ of a turn in any direction, the white flat inlet cap should become loose and easily come off (**Figure 2**). Then, discard the white flat inlet caps.

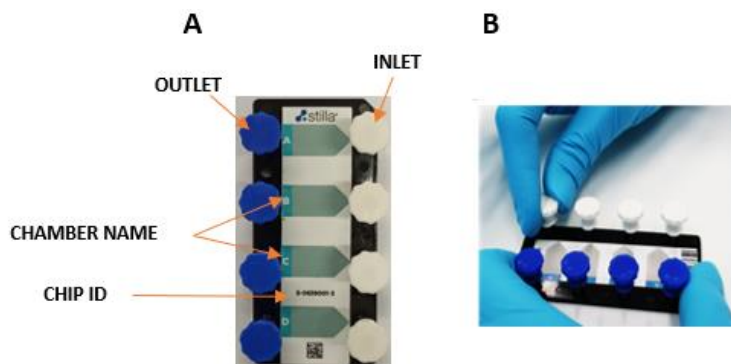


Figure 2: Disconnect the flat caps from the inlets (A) of the chips (B).

- 5) Pipet 25 µL of reaction mix into each inlet.
 - a. During pipetting the reaction mix stays above the oil phase. Do not insert the tip in the oil phase.
 - b. Make sure to change pipette tips after each pipetting step.
- 6) Place one tall white vented Sapphire Chip PCR cap on top of each inlet.
 - a. Tighten the tall white vented Sapphire Chip PCR cap enough to avoid disconnection from the inlets (**Figure 3**).
 - b. If all chambers of a Sapphire Chip are not used during an experiment, disconnect the white flat inlet caps from the inlets of these chambers and place one tall white vented Sapphire Chip PCR cap on each inlet. These chambers should not be selected for reading in the Crystal Reader software.

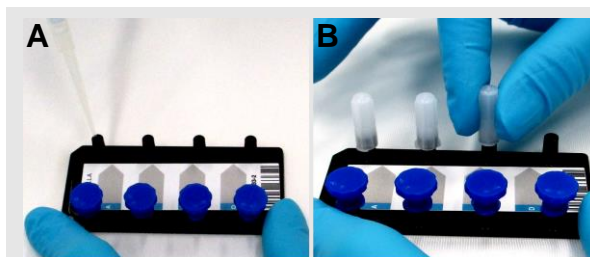


Figure 3: Add the PCR reaction mix (A) and place one tall cap onto each inlet (B).

Do not attempt to load more than 3 Sapphire Chip consumables at a time to prevent evaporation of the oil contained within the Sapphire Chip.

Hold the chips by the tall white vented Sapphire Chip PCR caps and transfer the Sapphire Chip to the plastic blister or a Petri dish containing a Precision Wipe to transfer the chips to the Geode.

For the subsequent steps, please refer to the Geode User Manual available the Technical Resources webpage.

Precautions and Warnings

Precautions

1. Wear protective clothing: nitrile or PVA (Polyethylene/Ethylene Vinyl Alcohol) gloves – safety glasses with side shield are recommended.
2. Avoid release of oil or components contaminated with oils in the environment.
3. Adsorb any spill with a cleaning wipe or an inert liquid-binding material, dispose according to precaution mentioned in 2.
4. Before the experiment, please be familiar with and master the operation methods and precautions of various instruments to be used and carry out quality control for each experiment.
5. Use general dilution ventilation and/or local exhaust ventilation.
6. Laboratory management should be strictly in accordance with the management regulations of PCR gene amplification laboratories.
7. All test samples are considered to be infectious. During the experiment, wear overalls, disposable gloves and change gloves frequently to avoid cross-contamination between samples. Sample handling and waste disposal must meet relevant regulatory requirements: General Guidelines on Biosafety of Microbial Biomedical Laboratories and Regulations on Medical Waste Management.

Warnings

1. In case of inhalation of emulsion oil: aerate with fresh air; if you feel unwell, seek medical help.
2. In case of skin contact with emulsion oil and surfactant: wash with soap and water. If signs/symptoms develop, seek medical help.
3. In case of eye contact with emulsion oil and surfactant: flush with large amounts of water. Remove contact lenses if easy to do. Continue rinsing. If signs/symptoms persist, seek medical help.
4. In case of emulsion oil and surfactant is swallowed: rinse the mouth. If you feel unwell, seek medical help.
5. In case of fire, use a fire-fighting agent suitable for ordinary combustible materials such as water or foam to extinguish.

Handling and Storage Requirements

Handling


1. Handle the Sapphire Chip by the edges to avoid any marks on the chamber. Do not touch the back side of the Sapphire Chip, as this may compromise further analysis. Do not hold the Sapphire Chip by the blue caps.

2. Handle the Sapphire Chip with caution and avoid shaking.
3. Carry the chips in the plastic blister provided or on a Petri dish lined with a Precision Wipe.
4. Avoid contact with hair, hands, clothes, and everything that might accumulate static energy.
5. While transferring the chips from the Geode to the Prism3 or Prism6 instrument, handle them by the white vented PCR caps and transfer them directly from the Geode to the Prism3 or Prism6 instrument. Discharge the tray holder of the Prism3 or Prism6 instrument by touching it before taking the chips from the Geode.
6. Handle the Sapphire Chip with the flat inlet port caps (blue) and white vented Sapphire Chip PCR caps pointing upwards.

Storage

1. Keep the shipping box in upright position in a clean area, at room temperature (+15°C to +25°C).
2. Store the Sapphire Chip in the original packaging and keep away from strong bases, strong alkali, and oxidizing agents.
3. Use the Sapphire Chip before the expiry date stated on the Sapphire Chip shipping box.

Disposal Considerations

	<p>Must not be disposed with household garbage. Do not allow product to reach sewage system.</p> <p>Dispose of waste product in a permitted industrial waste facility. Consult state local or national regulations for proper disposal.</p> <p>In some countries, the waste must be incinerated in a permitted incineration facility that can handle fluorine containing material.</p>
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Sapphire Chip Limitations

- Strict compliance with this document is required for optimal results.
- Appropriate collection, transport, storage, and processing procedures of samples are required for optimal results. Any improper handling and operation will cause inaccurate results.

Technical Support Contact Information

Online Technical Support is available at: www.stillatechnologies.com/technical-support/

For help and technical advice or any issue regarding the Sapphire Chip contact us:

For customers from Europe, China, and Africa:

Monday to Friday, 9:30 AM - 6:30 PM Central European Time (CET).

Closed on French bank holidays.

Phone: (+33) 9 82 27 47 47

Email: support@stilla.fr

For customers from America, Asia (without China) and Oceania

Monday to Friday, 8:00 AM – 6:00 PM, Eastern Standard Time (EST).

Closed on American bank holidays.

Phone: 1-833-888-0150 ext. 1

Email: support@stilla-inc.com

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