FOUNDATIONONE®CDx

# Specimen Instructions

If submitted material does not meet the standard requirements listed below, the test may result in a qualified report* or additional tumour material may be requested. **This can lengthen testing time or result in an insufficient specimen for FoundationOne®CDx analysis.**

* In the event of a qualified report, alterations detected will be listed however due to the specimen quality, there may be additional alterations present that could not be detected.

## Intended use

FoundationOne®CDx (F1CDx) is a next generation sequencing based in-vitro diagnostic device. The test detects substitutions, insertion and deletion alterations (indels), and copy number alterations (CNAs) in 324 genes. It also identifies select gene rearrangements, as well as genomic signatures including microsatellite instability (MSI), tumour mutational burden (TMB) and genomic loss of heterozygosity (gLOH – for selected tumour types), using DNA isolated from formalin-fixed paraffin embedded (FFPE) tumour tissue specimens.

The test is intended to identify patients who may benefit from treatment with therapies in accordance with approved therapeutic product labelling. Additionally, F1CDx is intended to provide tumour mutation profiling to be used by qualified health care professionals in accordance with professional guidelines in oncology for patients with solid malignant neoplasms.

## How to select the best specimen from multiple options

**Has the patient been treated with targeted therapy?**

NO

- Use the most recent available specimen.
- Choose the specimen with either the highest or largest tumour focus.
- Primary tumour or metastasis is acceptable.

YES

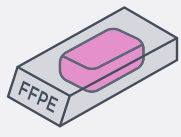
- It is **critical** to use a post-targeted therapy specimen.
- Submitting a FoundationOne®Liquid CDx liquid biopsy test for solid tumours can be considered if:
  - post-targeted therapy specimen is not available
  - or the tissue obtained is insufficient.

## Optimal specimen requirements

**Acceptable samples**

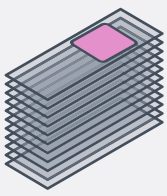
- Specimen types include tissue resections, small diagnostic biopsies, core-needle biopsies, fine-needle aspirations and effusion cytologies made into cell blocks.
- Tissue should be fixed using standard fixation methods to preserve nucleic acid integrity. The industry standard is **10% neutral-buffered formalin** for 6–72 hours. **Do not use other fixatives** (Bouins, B5, AZF, Holland's).
  **Please note:** Fresh tissues are not acceptable!
- **Do NOT decalcify** with strong acids (e.g. hydrochloric, sulfuric, or picric acid). Decalcification degrades DNA in samples making it unusable for comprehensive genomic profiling. Samples containing bone can be softened by EDTA chelation.
- Samples can be submitted as paraffin blocks or **unbaked** unstained slides cut at 4–5 microns thick.

### 1 SAMPLE SIZE




**FFPE block**

or



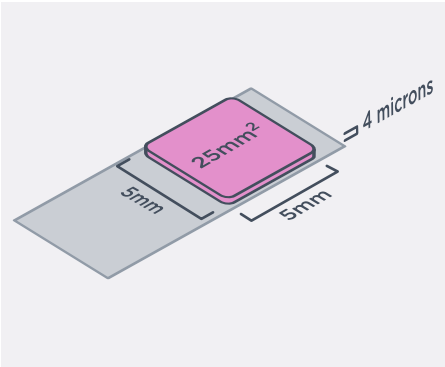
**at least 11 unstained slides**
(positively charged and **unbaked** at 4–5 microns thick).

**Use standard slides** (appr. 26mm x 76mm) and **standard tissue cassettes** (appr. 30mm x 25mm x 4mm).

For blocks or slides outside these standards please contact Roche Customer Service for additional guidance.

**Please note:** Foundation Medicine will attempt to return submitted paraffin blocks to the declared address on the order form depending on country of origin regulations.

## 2 SURFACE AREA AND VOLUME




**Optimal volume: 1mm³**

Surface area of tissue x thickness of section x number of slides.

**Example:**
5mm x 5mm (= 25mm²)
x 4 microns (= 0.1mm³)
x 10 slides = 1mm³

**Minimal acceptable volume: 0.6mm³**

For specimens with a smaller surface area (<25mm²) or impure samples, additional unstained slides may be needed to extract sufficient DNA for testing.

**Example:**
3mm x 5mm (= 15mm²)
x 4 microns (= 0.06mm³)
x 10 slides = 0.6mm³

**Specimens with tissue volume of 0.2mm³ to 0.6mm³**

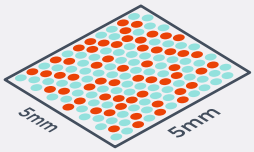
This tissue volume is accepted as conditional. The Foundation Medicine laboratory first reviews the specimen. If it is acceptable, the ordering physician is contacted for their approval to proceed with the available specimen.

## 3 TUMOUR NUCLEI PERCENTAGE

Percent tumour nuclei (%TN) = number of tumour cells divided by total number of all cells with nuclei.

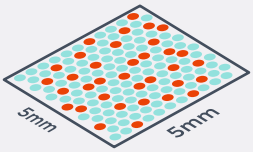
**Note:** Area of tissue occupied by tumour is not the same as percent tumour nuclei.

**Optimal**
(≥35%)



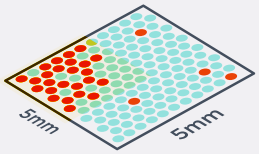

Nucleated tumour cells are dispersed over entire square with a ratio of more than 35:100 (diffuse).

**Acceptable**
(20%–35%)



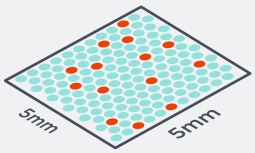

Nucleated tumour cells are dispersed over entire square with a ratio more than 1:5.

**Suboptimal**
(<20%)




Sparce distribution of cells with overall ratio less than 20:100 but all tumour cells are clustered together.

**Potentially acceptable specimen** with enrichment of the area containing tumour cells.



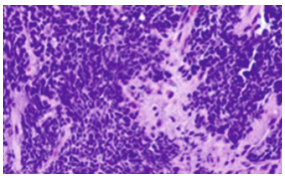

Nucleated tumour cells are dispersed over entire square with a ratio less than 1:5 (diffuse).

Cannot be enriched. Sample may be processed with approval of ordering physician if deemed acceptable after review at Foundation Medicine.

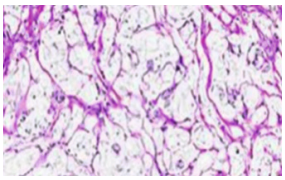
**Nucleated tumour cells**
**Normal cells**

**For liver specimens:** Minimum tumour content is ≥40%. Since polyploidy is a common characteristic of hepatocytes, twice as many tumour cells would be needed to obtain enough tumour DNA for analysis – higher tumour content may be required because hepatocyte nuclei have twice the DNA content of other somatic nuclei.

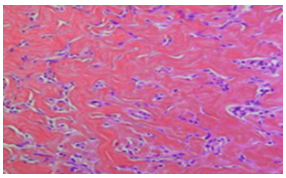
**Be aware: tissue nucleated density modifies volume of tissue required.** Areas of necrosis and fibrosis, extracellular mucin or other non-DNA containing material can decrease tissue density.



Hypercellular



Paucicellular mucin



Paucicellular

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**Shipping instructions:**

1. Please place the samples, FoundationOne CDx Test Requisition Form and any other documents into the FoundationOne CDx Specimen Shipping Kit
2. Call your country customer care center listed at rochefoundationmedicine.com "Contact us" to request a pick-up if needed.



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