

## SS18 (SYT) gene break apart probe reagent Instructions Manual

**[Product Name]** SS18 (SYT) gene break apart probe reagent

**[Package specification]** 5Tests /box, 10Tests /box

### **[Product introduction]**

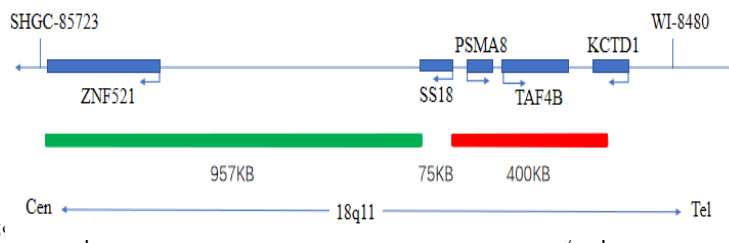
This kit uses orange fluorescein and green fluorescein labeled SS18 probe, and the probe could be bound to the target site by in situ hybridization.

### **[Product Main Components]**

The kit consists of SS18 dual color probe, as shown in Table 1. T

**Table 1 Kit composition**

| Component name        | Specifications | Quantity | Main components                     |
|-----------------------|----------------|----------|-------------------------------------|
| SS18 dual color probe | 50μl/Tube      | 1        | SS18 orange probe, SS18 green probe |
| SS18 dual color probe | 100μl/Tube     | 1        | SS18 orange probe, SS18 green probe |



### **[Storage conditions & Validity]**

Keep sealed away from light at  $-20^{\circ}\text{C}\pm 5^{\circ}$  and thawing that should not exceed 10 times. After opening, within 24 hours for short-term preservation, keep sealed at  $2\sim 8^{\circ}\text{C}$  in dark. For long-term preservation after opening, keep the lid sealed at  $-20^{\circ}\text{C}\pm 5^{\circ}\text{C}$  away from light. The kit was transported below  $0^{\circ}\text{C}$ .

### **[Applicable Instruments]**

Fluorescence microscope imaging system, including fluorescence microscope and filter set suitable for DAPI (367/452), green (495/517) and orange (547/565).

### **[Sample Requirements]**

1. Applicable specimen type: paraffin embedded specimen of surgical resection or biopsy tissue.
2. The tissue in vitro should be fixed with 4% neutral formaldehyde fixative within 1 hour. After the tissue is fixed, it is often dehydrated and embedded in paraffin.

### **[Instruction]**

#### **1. Hybridization pretreatment**

Recommended to use the "FISH Pretreatment Reagent" of Wuhan HealthCare Biotechnology Co., Ltd. for pretreatment.

#### **2. Denaturing hybridization**

The following operations should be carried out in the dark room.

- ① Take out the probe, let it stand at room temperature for 5min, turn it upside down with force, mix the probe well, centrifuge it briefly (do not vibrate with vortex apparatus), drop 10μl into the hybridization area of the cell drop, cover the 22mm×22mm cover glass immediately, the probe should be evenly spread under the cover glass without bubbles, and seal the edge with rubber (the edge sealing must be thorough to prevent the dry slide from affecting the test results in the hybridization process).
- ② Place the glass slide in the hybridization instrument, denature at  $85^{\circ}\text{C}$  for 5 min (the hybridizer should be preheated to  $85^{\circ}\text{C}$ ) and hybridized at  $42^{\circ}\text{C}$  for 2-16h.

#### **3. Washing**

The following operations should be carried out in a dark room.

- ① Carefully tear off the adhesive around the cover glass with tweezers to avoid sticking off or moving the cover glass. Immerse the cell drop into 2xSSC for about 5s, and take it out. Gently push one corner of the cover glass to the edge of the slide with tweezers, and gently remove the cover glass with tweezers;
- ② The cells were placed at 2xSSC room temperature for 1min;
- ③ 3% NP-40/0.4 xSSC solution preheated at  $68^{\circ}\text{C}$  for 2min;

④ The sides were immersed in deionized water preheated at 37°C for 1min, and then dried naturally in the dark.

#### 4. Counterstaining





The following operations should be performed in a darkroom

10μl DAPI compound dye is dropped in the hybridization area of the glass slide and immediately covered. The suitable filter is selected for glass slide observation under the fluorescence microscope.

#### 5. FISH results observation

Place the stained sections under a fluorescence microscope and the cells area is first confirmed under a low magnification objective (10×); under magnification objective (40×) a uniform cells distribution is observed; then the nucleus size uniformity, nuclear boundary integrity, DAPI staining uniformity, no nuclei overlapping, cells clear signal are observed in the high magnification objective (100x).

#### [Interpretation of common signal types]

|  |                                     |
|--|-------------------------------------|
|  SS18 gene 5' site signal<br> SS18 gene 3' site signal |                                     |
|   | Negative: 2 fusion                  |
|   | Positive: 1 orange 1 green 1 fusion |

#### [Precautions]

- ① This product cannot be used for clinical diagnosis, but only for scientific research.
- ② The results of this kit will be affected by various factors of the sample itself, but also limited by hybridization temperature and time, operating environment and the limitations of current molecular biology technology, which may lead to wrong results.
- ③ Users must understand the potential errors and accuracy limitations that may exist in the detection process.
- ④ All chemicals are potentially dangerous. Avoid direct contact. Used kits are clinical waste and should be properly disposed of.

#### [Basic information]

Name of registrant / manufacturer: Wuhan HealthCare Biotechnology Co., Ltd.

Address: Floor 1-4, Building #8, Optics Valley Precision Medicine Industry Base Phase I, #9 Gaokeyuan 3rd Road, East Lake High-Tech Zone, Wuhan City, Hubei Province, People's Republic of China.

Contact: 027-87570662

Name of after-sales service unit: Wuhan HealthCare Biotechnology Co., Ltd.

Contact: 18140559890

Production address: Floor 1-4, Building #8, Optics Valley Precision Medicine Industry Base Phase I, #9 Gaokeyuan 3rd Road, East Lake High-Tech Zone, Wuhan City, Hubei Province, People's Republic of China.

#### [approval date and modification date of the specification]

V1.0 approval date: April 1, 2019

V1.2 approval date: December 7, 2021