

## FeraTrack™ MRI contrast agent kit for *in vitro* labeling

5 tracking experiments 130-096-498  
25 tracking experiments 130-096-499

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### 1. Description

<b>Components</b>	<p>1×200 µL FeraTrack™ Contrast Particles, MRI cell tracking agent (superparamagnetic iron oxide [SPIO] nanoparticles) 100 µL FeraTrack™ Loading Reagent</p> <p>or</p> <p>5×200 µL FeraTrack™ Contrast Particles, MRI cell tracking agent (superparamagnetic iron oxide [SPIO] nanoparticles) 500 µL FeraTrack™ Loading Reagent</p>
<b>Capacity</b>	<p>For 5×10<sup>6</sup> total cells, up to 5 tracking experiments</p> <p>or</p> <p>for 25×10<sup>6</sup> total cells, up to 25 tracking experiments.</p>
<b>Product format</b>	<p>FeraTrack™ Contrast Particles are supplied in a sterile PBS/EDTA buffer solution with an iron concentration of 45 mM, manufactured and controlled under an ISO 9001 certified quality system.</p> <p>FeraTrack Loading Reagent is supplied as a sterile lipid-containing solution.</p>
<b>Appearance</b>	<p>FeraTrack Contrast Particles are a black to reddish-brown aqueous solution of dextran-coated SPIO particles.</p> <p>FeraTrack Loading Reagent is a clear colorless to amber lipid solution.</p>
<b>Storage</b>	<p>Store protected from light at -20 °C. The expiration date is indicated on the vial label.</p>

**For laboratory and animal research use only. Warning: Not for human or animal therapeutic or diagnostic use. Make sure to comply with all laws and regulations governing research on animals.**

#### 1.1 Background information

Magnetic resonance imaging (MRI) is the most frequently used technique for serial *in vivo* cell tracking applications due to high resolution of soft tissues, which makes it especially useful for imaging of the brain, muscles, or the heart.<sup>1</sup> In order to improve detectability of transplanted cells and produce a strong contrast against surrounding tissue, intracellular labeling of cells with iron oxide particles before transplantation has been described.<sup>2</sup> Our newly developed FeraTrack Contrast Particles are superparamagnetic iron oxide (SPIO) particles specifically formulated for *in vitro* labeling of various cell types prior to transplantation and pre-clinical MRI. Once within cells, SPIOs can induce decreased signal intensity on T1, T2, and T2\*-weighted images.<sup>3</sup>

#### 1.2 Applications

The FeraTrack labeling kit was optimized for proper *in vitro* labeling of cell lines (NIH-3T3, Jurkat), primary cells (granulocytes, neural progenitors), and stem cells (hematopoietic, mesenchymal, and embryonic stem cells) from different species (mouse, human, rat). Similar results were obtained with differentiation assays comparing labeled and unlabeled stem cells indicating biocompatibility of intracellular labeling. Feasibility of the newly developed contrast particles for MRI-based *in vivo* cell tracking has been evaluated exemplary by transplantation of mouse neural progenitors and rat granulocytes. FeraTrack is not applicable for intracellular magnetic labeling of natural killer (NK) cells.

#### 1.3 Physico-chemical properties of FeraTrack Contrast Particles

Compound: dextran-coated ferumoxide; chemically, ferumoxide is a non-stoichiometric magnetite of the average formula Fe<sub>3</sub>O<sub>4</sub>. Mean particle size: 100 nm (hydrodynamic diameter, intensity weighted). Particle size range: 60–140 nm. Dosage: 100 µg Fe / 1×10<sup>6</sup> cells.

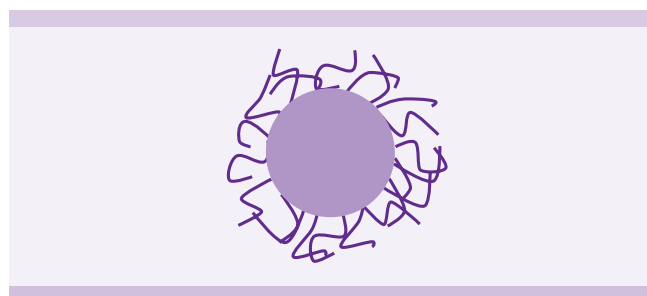


Figure 1: Schematic diagram of a FeraTrack nanoparticle.

## 1.4 Quality control

FeraTrack Contrast Particles and FeraTrack Labeling Reagent are tested for absence of microbial contamination by Thioglycate and Caso-Bouillon sterility testing.

## 1.5 Requirements

- Polystyrene cell culture dish, e.g., 6-well plate
- Standard CO<sub>2</sub> incubator
- Cell culture medium (w/o serum and antibiotics)
- Phosphate-buffered saline (PBS)

## 2. Protocol

### 2.1 General advices

- ▲ Read the entire protocol before starting.
- ▲ Ensure sterile handling of FeraTrack Contrast Particles, FeraTrack Labeling Reagent, and cells.
- ▲ Removal of serum and antibiotics from medium will improve intracellular magnetic labeling performance.
- ▲ Standard animal handling procedures and local regulations must be followed in case of *in vivo* application of labeled cells.

### 2.2 Preparation of cells

1. Adherent cells: plate 0.5–1×10<sup>6</sup> cells per 9 cm<sup>2</sup> cell culture dish (6-well plate) in culture medium one day before labeling to ensure 90% confluency at the time of labeling.
2. Suspension cells: plate 0.5–1×10<sup>6</sup> suspension cells per 9 cm<sup>2</sup> cell culture dish (6-well plate) in 1 mL culture medium without serum and antibiotics at least 10 minutes prior to labeling.

### 2.3 Preparation of the labeling mix

1. Prepare the labeling mix by addition of 20 µL of FeraTrack Loading Reagent and 40 µL of FeraTrack Contrast Particles to 200 µL cultivation medium (w/o serum and antibiotics) and mix gently.
2. Incubate at room temperature for 10 minutes.
3. Add 740 µL of cultivation medium (w/o serum and antibiotics).

## 2.4 Intracellular magnetic labeling

### Adherent cells:

1. Wash cells with PBS to remove serum from the culture dish.
2. Add 1 mL of cultivation medium (w/o serum and antibiotics). Proceed with 3.

### Suspension cells:

3. Add labeling mix dropwise to the cells.
4. Incubate cells at 37 °C for 4–6 h in a CO<sub>2</sub> incubator.
5. After 4–6 h remove labeling solution from the cell culture dish.
6. Wash cells at least twice with PBS.
7. Add cultivation medium (if required with serum and antibiotics) to labeled cells for subsequent cultivation steps or harvest cells for alternative downstream applications.

## 3. References

1. Rogers, W.J. *et al.* (2006) Technology inside: *in vivo* cell tracking by use of MRI. *Nat. Clin. Pract. Cardiovasc. Med.* 10: 554–562.
2. Hoehn, M. *et al.* (2007) Cell tracking using magnetic resonance imaging. *J. Physiol.* 584: 25–30.
3. Bulte, J.W. and Kraitchman, D.L. (2004) Iron oxide MR contrast agents for molecular and cellular imaging. *NMR Biomed.* 17: 484–499.

## 4. Related products

A comprehensive product portfolio for the imaging modalities MRI, CT, US, OI, SPECT, and PET is available at [www.miltenyibiotec.com/viscover](http://www.miltenyibiotec.com/viscover).

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