



Bone Marrow

Targeted Cell Isolation & Enrichment

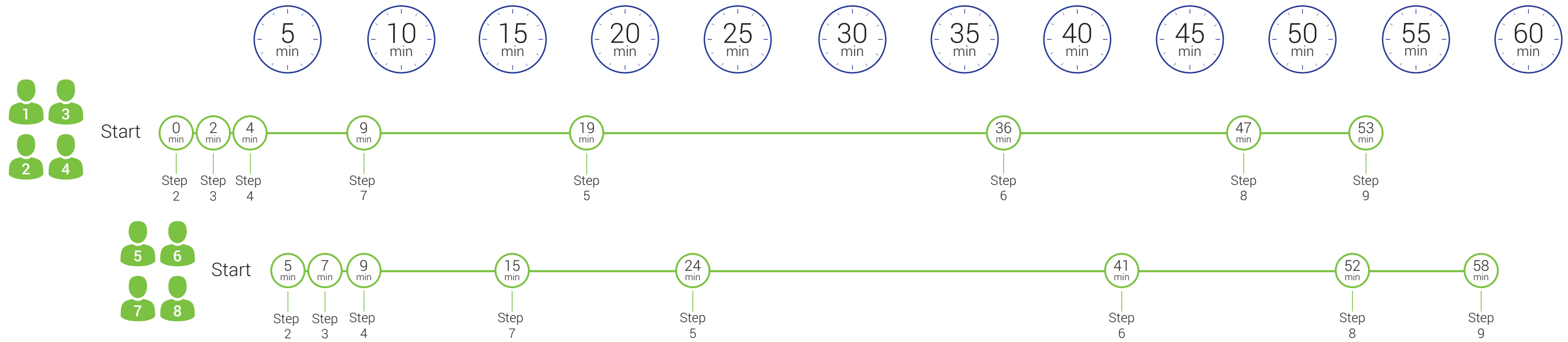
8 Specimens. 80 Slides. 60 Minutes.

WAVESENSE
Enriching Patient Lives



Single-Tube Cell Isolation & Enrichment

8 Specimens. 80 Slides. 60 Minutes.



Step 1

Prior to target cell enrichment, prepare a slide smear from the bone marrow specimen for morphological review. The slide smear will serve as a reference that sufficient viable cells are present.

To identify the presence of target cells on your smear, use WaveSense PlasmaCellSM or other appropriate cellular stain.

Step 2

Using a 15 mL conical tube, create a homogeneous mixture by combining RPMI 1640 media with the bone marrow specimen. To determine the amount of bone marrow specimen to allocate for each EpiSep[®] HS use the following criteria:

- If a primary bone marrow is received use ≤500 µL of specimen.
- For a specimen from a secondary, tertiary pull or hemodilute bone marrow, use a greater volume. It is recommended to use 500 µL to 1 mL.

Specimen/media volume to EpiSep HS conversion chart:

| | |
|--------------------|----------------------|
| 1 mL = 1 EpiSep HS | 6 mL = 6 EpiSep HS |
| 2 mL = 2 EpiSep HS | 7 mL = 7 EpiSep HS |
| 3 mL = 3 EpiSep HS | 8 mL = 8 EpiSep HS |
| 4 mL = 4 EpiSep HS | 9 mL = 9 EpiSep HS |
| 5 mL = 5 EpiSep HS | 10 mL = 10 EpiSep HS |

Step 3

Vortex to resuspend the paramagnetic antibodies.

As specified in the chart below, add 10 µL of the appropriate paramagnetic antibody to the specimen/media mixture in the 15 mL tube for each EpiSep HS.

Paramagnetic antibody volume to EpiSep HS conversion chart:

| | |
|---------------------|-----------------------|
| 10 µL = 1 EpiSep HS | 60 µL = 6 EpiSep HS |
| 20 µL = 2 EpiSep HS | 70 µL = 7 EpiSep HS |
| 30 µL = 3 EpiSep HS | 80 µL = 8 EpiSep HS |
| 40 µL = 4 EpiSep HS | 90 µL = 9 EpiSep HS |
| 50 µL = 5 EpiSep HS | 100 µL = 10 EpiSep HS |

Step 4

Cap and gently invert the 15 mL conical tube several times (3-4). Attach the 15 mL CellCycler Tube Adapter to the 15 mL conical tube then insert it into the CellCycler. Set voltage to 7.5V DC (14.0 - 16.0 RPM) for 15-30 minutes. The CellCycler is used to keep cells and reagents in uniform suspension during the incubation of cells with paramagnetic antibodies.

Step 5

At the end of the incubation period, loosen the cap of the 15 mL conical tube and place it firmly into a MTD-15 (#A4102-1) magnetic tube dock for 5 minutes. **NOTE: Do not incubate more than 5 minutes in the MTD-15 as it may induce aggregation.**

At the end of the 5-minute incubation, remove supernatant from the side opposite the MTD-15 magnet. Be careful not to scrape cells off the side of the conical tube.

After the supernatant is removed with an aspirator or transfer pipette, place the supernatant into a waste container labeled biohazard. **NOTE: Supernatant contains non-target cells that may be useful for further analysis.**

Remove the 15 mL conical tube from the MTD-15. Slowly add the first 1.0 mL of hypotonic solution (0.75M KCL) while gently mixing to resuspend the target cells.

As specified below, add the additional hypotonic solution required and continue to gently mix. Cap the 15 mL conical tube, invert the tube to mix cells and place in a 37°C environment for 10 minutes.

Specimen volumes < 500 µL = 5 mL of 0.075M KCL.
Specimen volumes ≥ 500 µL = 10 mL of 0.075M KCL.

Step 6

At the end of the 10-minute incubation period, remove the 15 mL conical tube from the incubator. Slowly add 1 mL of Carnoy's fixative. Invert gently to mix.

Loosen the cap of the 15 mL conical tube and place it firmly into a MTD-15 (#A4102-1) magnetic tube dock for 5 minutes. **NOTE: Do not incubate more than 5 minutes in the MTD-15 as it may induce aggregation.**

At the end of the 5-minute incubation, remove supernatant from the side opposite the MTD-15 magnet. Be careful not to scrape cells off the side of the conical tube.

Remove the 15 mL conical tube from the MTD-15. Resuspend the cells with Carnoy's fixative using the same volume conversion in Step #2 and gently mix. **NOTE: For storage of the tube for reflex testing, please refer to www.wavesense.com/faq**

Step 7

Label each EpiSep HS with a pencil and insert into the MSD5 (#A1104-1) five position magnetic slide dock. Make sure it clicks to indicate full insertion.

Step 8

Add 1 mL of suspension from the 15 mL conical tube to the inner wall of each EpiSep HS. Allow 1 minute for the suspension to drain or until all fluid has drained from the well. **NOTE: Do not let the slides sit and dry with the EpiSep HS cap still attached.**

Remove EpiSep HS cap by gently pulling up while slide is still wet and in the MSD5. Place slides into a humidity-controlled environment for 5 minutes. (45-50% humidity)

Step 9

Slides are now ready for your laboratory's standard FISH protocol or special stain process. Inspect monolayered cell target area by viewing slides under a phase contrast microscope.





WaveSense S1110 Start-Up Promotion

The WaveSense S1110 Start-Up Promotion includes all the tools you need to bring the most specific and sensitive targeted cell isolation and enrichment solution to your FISH slide preparations. Just select your antibody and you are ready to go.

Due to the unparalleled sensitivity of its products, WaveSense provides the most effective targeted cell isolation and enrichment solution, requiring only a small specimen to deliver the maximum number of pure target cells.

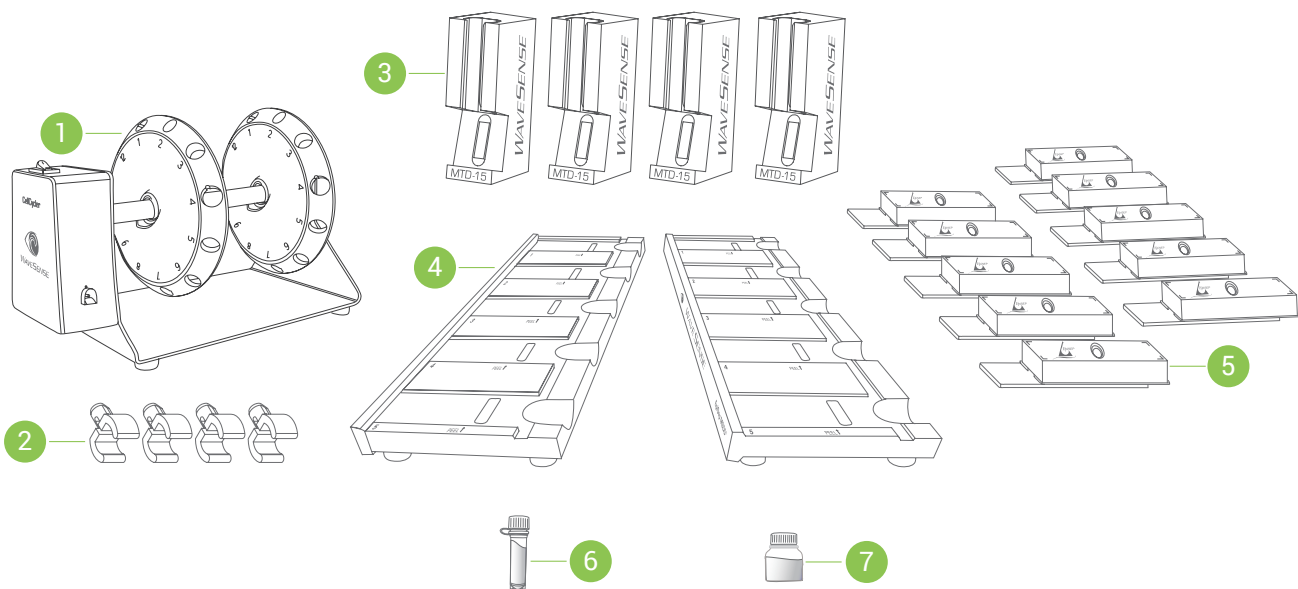
Importantly, the WaveSense direct-to-slide process eliminates steps required by competitive processes that are both time-consuming and compromise specimen integrity... making the WaveSense cell enrichment platform the industry's most effective and efficient workflow solution.

Put the WaveSense direct-to-slide cell enrichment technology to work for you today at this special introductory offer.

All products are intended for In Vitro Diagnostic Use.

Products

| | Product Name | Model # | Quantity | Price | Discount | Promotion Price |
|--------------|--|----------|----------|-------------------|------------------------------|-------------------|
| 1 | CellCycler™ | A2102-1 | 1 | \$795.00 | \$795.00 | \$0 |
| 2 | 15 mL Tube Adapter (Pack of 4) | A2108-1 | 1 | N/C | N/C | N/C |
| 3 | 15 mL Magnetic Tube Dock (MTD-15) (Box of 4) | A4102-4 | 1 | \$540.00 | \$540.00 | \$0 |
| 4 | Five Position Magnetic Slide Dock (MSD5) | A1104-1 | 2 | \$1,200.00 | \$700.00 | \$500.00 |
| 5 | EpiSep® HS (Box of 10) | A3104-10 | 5 | \$1,750.00 | \$0 | \$1,750.00 |
| 6 | Choice of Paramagnetic Antibody (500 µl) | TBD | 1 | \$350.00 | \$350.00 | \$0 |
| 7 | PlasmaCell ^{QC} Stain (3 mL) | R2144-1 | 1 | \$100.00 | \$100.00 | \$0 |
| Total | | | | \$4,735.00 | \$2,485.00 | \$2,250.00 |



Note: Promotion S1110 is an introductory offer available exclusively to new WaveSense customers. EpiSep is a registered trademark of WaveSense Incorporated.
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