**Product Information Sheet for MRA-1184**

**PARASITE**

**MR4 Number:** MRA-1184  
**Organism:** Plasmodium falciparum  
**Strain:** SenTh028.04  
**Alt. Strain designation:** T28.04  
**Geographic Origin:** Human patient isolate, Thiès, Senegal  
**Date of isolation:** 2004  
**Genotype:** TATCGGATTGTGCCTACGCTG  
(Broad 24 SNP Bar Code, Daniels et al. Malaria Journal 7:223, 2008)  
**Unit size:** 0.5ml  
**Depositor:** Dyann Wirth, Sarah Volkman, Harvard School of Public Health; Souleymane Mboup, Daouda Ndiaye  


**MR4 Growth and Preservation Protocols**

**Recommended Blood Type for in vitro culture:** Type O blood (washed); pooled human serum Type A or Type O recommended.

**Growth Temperature:** 37°C

**Media Preparation:** To make 1.0 L of incomplete medium, start with either 928 ml liquid RPMI-1640 (without NaHCO₃, without L-glutamine), or, to 900 ml Tissue Culture grade water, add 10.43 g of powdered RPMI-1640 (without NaHCO₃, without L-glutamine). To the RPMI-1640 media, add 25 ml of 1 M HEPES (final = 25 mM), 27 ml 7.5% sodium bicarbonate solution (final =0.2% NaHCO₃), 10 ml of 200 mM L-glutamine (final =2 mM), 10 ml 20% Glucose (final = 20 mM, optional), 27.2 mg hypoxanthine (10 ml of 20 mM hypoxanthine stock in 1M NaOH), 0.25 ml 10mg/ml Gentamicin (final =2.5 ug/ml, optional). Add TC grade water to 1.0 L. Mix thoroughly and filter with 0.22 cm sterile filter unit. Store at 2–8°C.

**Cryopreservation:** Only immature parasite stage (rings) are viable by this method. We recommend a parasitemia of 3% or higher of ring stage parasites for cryopreservation.

1. Centrifuge the culture at 1000 x g for 5 min.
2. Wash the pellet once with 10 or more volumes of incomplete RPMI-1640 media. Centrifuge at 1800 for 5 min and leave sufficient supernatant to resuspend the pellet.
3. To the volume of packed RBCs, add slowly dropwise one volume of cold (4°C) Glycerolyte 57. Let stand for 5 min at room temperature.
4. Add an additional 3 volumes of cold Glycerolyte 57 to the pellet dropwise. Mix well and aliquot 0.5 ml into 1.5 ml sterile cryopreservation vials.
5. Place the samples into freezing containers (e.g., Nalgene Cryo 1°C / min Freezing Container) and store at −80°C for 24-48 hr.
6. Transfer to liquid nitrogen for long term storage.

**Important notes:** This reagent was authenticated by the contributor. Please contact malaria@atcc.org for any comment.
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Biosafety Level: 2

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In Materials and Methods “P. falciparum line Dd2 (MRA-156, MR4, ATCC® Manassas Virginia),...”. In the acknowledgment portion: “We thank MR4 for providing us with malaria parasites contributed by (name of depositor).”

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