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Protocol status: Working
 We use this protocol in the Harry de Koning group and it is working

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Bloodstream-form culture

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ABSTRACT

Protocol obtained from the Alsford Lab at the London School of Hygiene & Tropical Medicine

(<https://blogs.lshrm.ac.uk/alsfordlab/protocols/bloodstream-form-culture/>)

MATERIALS

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Fetal Bovine Serum, qualified **Life Technologies Catalog #10437-028**

Sodium bicarbonate **Sigma Aldrich Catalog #S6014**

β-mercaptoethanol **Sigma Aldrich Catalog #M3148**

- 1 Make up to 4.5 litres with distilled water, filter sterilise, store at 4°C in 450 ml volumes.

- 2** Prior to use, add 50 ml fetal bovine serum (FBS; Sigma), giving a final concentration of 10% FBS and store at 4°C for up to two months.
- 3** NB: Some early publications indicate that FBS should be heat inactivated at 56°C for 30 minutes prior to use in bloodstream-form *T. brucei* culture – this is not necessary; in fact our unpublished data suggests that using non-inactivated FBS leads to faster, more robust cell growth.
- 4** Cultures should be grown in 5% CO₂ incubator at 37°C, and should not exceed a density of 2.5×10⁶ cells per ml.