22Rv1 (ATCC, Cat#CRL-2505)
Farnham Lab (150923 version)

Complete growth medium: RPMI1640+10%FBS with optional addition of penicillin (100units/mL), streptomycin (100µg/mL).

Subculturing:
For cells in 15cm dish:

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
3. Add 6-8 mL of 0.25% (w/v) Trypsin-0.53mM EDTA solution to rinse cell.
4. Discard Trypsin-EDTA solution and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes). **Note**: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
5. Add 6-8mL of complete growth medium and aspirate cells by gently pipetting.
6. Add appropriate aliquots of the cell suspension to new culture vessels, a 1:3 to 1:6 ratio is recommended.
7. Incubate cultures at 37°C and change medium every 2-3 days.

Freeze medium: Complete growth medium supplemented with 5% (v/v) DMSO