

**NB100-386 Protocol****Western Blot protocol for Rad21 Antibody (NB100-386)**

Rad21 Antibody (CM110-2C10): [https://www.novusbio.com/products/rad21-antibody-cm110-2c10\\_nb100-386](https://www.novusbio.com/products/rad21-antibody-cm110-2c10_nb100-386)

1. Load protein on gel (ie: ~8 ug of non-transfected, control HeLa extract) and run.
2. Transfer protein to nitrocellulose (Schleicher&Schuell, cat# BA83).
3. Block the membrane with 5% milk for 30 minutes at room temperature.
4. Incubate the membrane with anti-cohesin [cat# NB 100-386] (1:500), overnight at 4C or 2 h at room temperature. Milk or PBS-T are good for this dilution.
5. Wash the membrane 3 times, 10 minutes per wash in PBS with 0.1% Tween-20 (PBS-T).
6. Incubate with secondary antibody for 30 minutes at room temperature. Use milk in PBS-T as a diluent.
7. Rinse the membrane 2 times with deionized water and place in an ECL working solution.
8. Expose to the film.