### ENCODE DCC Antibody Validation Document

**Date of Submission:** 9/13/12

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**Lab:** Snyder

**Antibody Name:** Anti-Rad21 antibody - ChIP Grade  
**Target:** Rad21

**Company/Source:** Abcam

**Catalog Number, database ID, laboratory:** ab992  
**Lot Number:** 734371

**Antibody Description:** Synthetic peptide (Human) conjugated to KLH - which represented a portion of human Rad21 encoded within exon 14 (LocusLink ID 5885).

**Target Description:** The protein encoded by this gene is highly similar to the gene product of Schizosaccharomyces pombe rad21, a gene involved in the repair of DNA double-strand breaks, as well as in chromatid cohesion during cell cycle M phase. The highly regulated association of this protein with mitotic chromatin specifically at the centromere region suggests its role in sister chromatid cohesion in mitotic cells. (provided by RefSeq)

**Species Target:** Mouse  
**Species Host:** Rabbit

**Validation Method #1:** Immunoprecipitation  
**Validation Method #2:** siRNA

**Purification Method:** Affinity  
**Polyclonal/Monoclonal:** Polyclonal

**Vendor URL:** [http://www.abcam.com/Rad21-antibody-ChIP-Grade-ab992.html](http://www.abcam.com/Rad21-antibody-ChIP-Grade-ab992.html)

**Reference (PI/Publication Information):**

Please complete the following for antibodies to histone modifications:

*if your specifications are not listed in the drop-down box, please write-in the appropriate information*

<table>
<thead>
<tr>
<th>Histone Name</th>
<th>AA modified</th>
<th>AA Position</th>
<th>Modification</th>
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*The protein encoded by this gene is highly similar to the gene product of Schizosaccharomyces pombe rad21, a gene involved in the repair of DNA double-strand breaks, as well as in chromatid cohesion during cell cycle M phase. The highly regulated association of this protein with mitotic chromatin specifically at the centromere region suggests its role in sister chromatid cohesion in mitotic cells. (provided by RefSeq)*
A band of ~130kD was immunoprecipitated from CH12 and MEL nuclear extracts using ab992. This antibody has been validated for human cell lines by immunoprecipitation and siRNA knockdown.
Validation 1: Immunoprecipitation (IP) in both CH12 and MEL cell lines

Arrow indicates immunoprecipitated band of Rad21 (~130 KDa).

Validation 2: Immunoprecipitation (IP) in both CH12 and MEL cell lines

Human Rad21 encoded within exon 14

Epitope: Rad21 Antibody is Rabbit polyclonal, epitope represents a portion of human Rad21 encoded within exon 14

Antibody: Rad21 Source: Abcam ab992

1gG: IP with control IgG
IP: IP with tested antibody
S: Supernatant after IP
NE: Nuclear extract
This antibody has been validated by siRNA knockdown for human cell. See documents submitted for human cell lines for details.