**Custom Column 3’**

<table>
<thead>
<tr>
<th>Category</th>
<th>DNA Oligo Synthesis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Modification Code</td>
<td>CC</td>
</tr>
<tr>
<td>Reference Catalog Number</td>
<td>26-6201</td>
</tr>
<tr>
<td>5 Prime</td>
<td>Y</td>
</tr>
<tr>
<td>3 Prime</td>
<td>Y</td>
</tr>
<tr>
<td>Internal</td>
<td>Y</td>
</tr>
<tr>
<td>Molecular Weight(mw)</td>
<td>308</td>
</tr>
</tbody>
</table>

There is no additional charge if the degenerate mixed base is at an internal position or at the 5’ end. Degenerate mixed base charges are only for 3’ end positions. A custom column charge is applied.

Custom column (3’) is a set of special controlled pore glass (CPG) synthesis solid supports that are used for synthesizing oligonucleotides with degenerate (mixed-base) nucleotides at the 3’-end.

Degenerate base means more than one base possibility at a particular position, this is usually the case when a DNA sequence is derived from amino acid sequence with codon based sequence. An oligo sequence can be synthesized with multiple bases at the same position, this is termed as degenerate base also sometime referred as ‘wobble’ position or ‘mixed base’.

IUB (International Union of Biochemistry) has established single letter codes for all possible degenerate possibilities. An example is “R” that is A+G at the same position with 50% of the oligo sequence will have an A at that position, and the other 50% have G. A degenerate base position may have any combination of two, three, or four bases.

For degenerate (mixed bases) positions use the following IUB codes.

- R = A+G
- Y = C+T
- M = A+C
- K = G+T
- S = G+C
- W = A+T
- H = A+T+C
- B = G+T+C
- D = G+A+T
- V = G+A+C
- N = A+C+G+T

**Alternate Oligo Design Strategies** The use of degenerate bases leads to complexity of oligo sequence and thus reduction in the percentage of the unique sequence. Consider the use of modifications such as Inosine that hybridizes to all four bases. See the listing of modifications as substitutes to reduce complexity if degenerate bases are used. Degenerate Base Modifications

**Custom Spiking & Custom Column(3’ base spiking/mix)**

Custom spiking is the addition of differing molar concentration of bases at a single position, this is different from degeneracy at a position based on codons. Codon based degeneracy is usually equimolar concentration of each base at the same position (done at no extra charge for all internal and 5’ position, see order form for single letter IUB codes). Custom spiking (example, 10% A, 75% G, 5% C & 10% T) has to be specified as required on the order form. See the following link...
Spiking Custom
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Custom column has to be prepared when the degeneracy and custom spiking is at the 3’ position. Customers who wish custom spiking at certain positions of their oligo must include the relevant specifics (position and spiking composition) in the comments section of the on-line order form for that particular oligo.