CHEMICON® INTERNATIONAL
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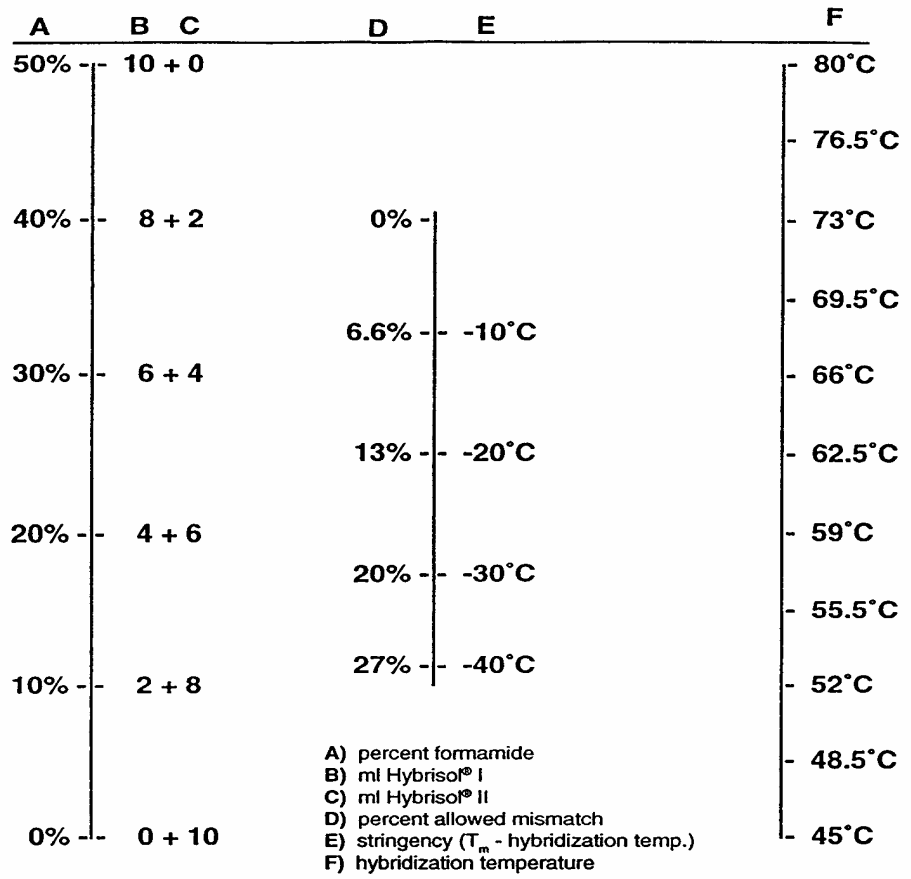
Adjustment of Hybridization Conditions
Using Hybrisol® Solutions (Bolton & McCarthy, 1962)

INSTRUCTIONS FOR USE: To determine hybridization conditions, align a straight edge with either the percent allowable mismatch (D) or the stringency (E). The percent formamide and incubation temperature are read at the intersection of the appropriate scale and the straight edge. Example: for stringent hybridizations (Tm - 20°C) Chemicon recommends 50% formamide and 45°C.



NOTE: This nomograph is based on 6X SSC (1 M Na+), and 45% GC content.
This nomograph was constructed using the following equation (Bolton & McCarthy, 1962):
Stringency = 16.6 log[Na+] + 81 + 0.41[%GC] - 0.72 [% formamide] - hyb temperature
To determine T_m for oligonucleotide probes use the following equation:

Stringency = 81˚C + 0.41 (%GC) - 0.72 (% Formamide) - 500/n - 20 n = bp of oligo

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