Seegene

DPO™ Technology, Super Multiplex PCR
Novel Oligo Platform for PCR: DPO™

DPO™ technology is a fundamental tool for blocking extension of non-specifically primed templates generating consistently high specificity. The strength and utility of this DPO™ technology can be used successfully incorporated into molecular diagnostics systems such as multiplex diagnosis and SNP genotyping systems.

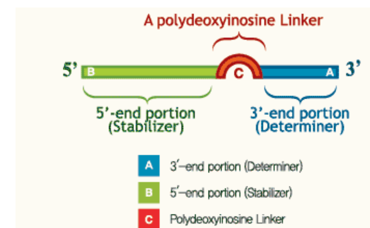
Successful PCR starts with proper priming between an oligonucleotide primer and the template DNA. However the inevitable risk of mismatched priming cannot be avoided in the currently used primer system, even through considerable time and effort are devoted to primer design and optimization of reaction conditions.
A novel DPO™ system that is structurally and functionally different from the primer system currently in wide-spread use blocked extension of non-specially primed templates, and thereby generates consistently high PCR specificity even under less than optimal PCR conditions.

Features of DPO™

- Freedom in primer design & PCR optimization
- Unparalleled high specificity
- Guaranteed reproducibility
- No primer competition and dimerization in Multiplex
- Single base discrimination
- Broad applications

Structure of DPO™

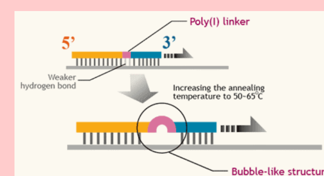
DPO™ comprises of two separate priming regions (a first priming region and a second priming region) joined by a polydeoxyinosine linker. The linker forms like a "bubble-like structure" which itself is not involved in priming, but rather delineates the boundary between two parts of primer.

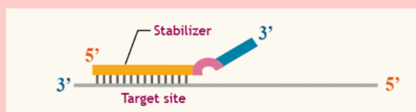


Principle of DPO™

DPO™ has two functional priming regions (one is longer than the other) separated by the poly (I) linker. These two unequally distributed priming regions generate dual priming reactions based on the following scheme, resulting in only target-specific products.

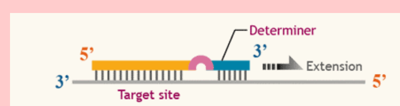
Step 1: Poly(I) linker activation
Deoxyinosine has a relatively low melting temperature compared to the natural bases, due to weaker hydrogen bonding so that the poly (I) linker will form a bubble-like structure at a certain annealing temperature and separates a single primer into a two functional regions.





Step 2: First priming reaction
The longer 5'-segment preferentially binds to the template DNA and initiates "stable annealing".
It acts as a Stabilizer".

Step 3: Second priming reaction
The short 3'-segment selectively binds to a target site and determines "target-specific extension".
It acts as a "Determiner".



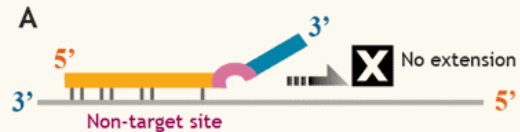
Seegene

DPO™ Features

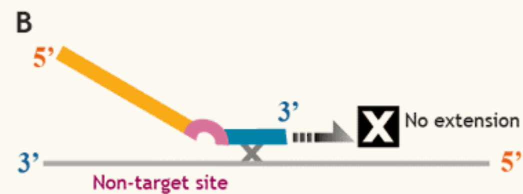
DPO™ provides numbers of advantages in PCR. A major advantage is that DPO™ generates unparalleled high specificity by blocking extension of non-specifically primed templates. Although "Stabilizer" alone binds to a non-target site, "Determiner" will not proceed with extension if there is any mismatch on a binding site (Fig. A). In addition, "Determiner" alone fails to make a priming at an annealing temperature (Fig. B). Therefore, a target-specific product is generated only when both of Stabilizer and Determiner of DPO™ make a perfect match with a target site.

DPO™'s main features:

1. Superior PCR specificity & sensitivity
2. Guaranteed reproducibility
3. No primer competition and dimerization in Multiplex
4. Single base discrimination
5. Broad applications

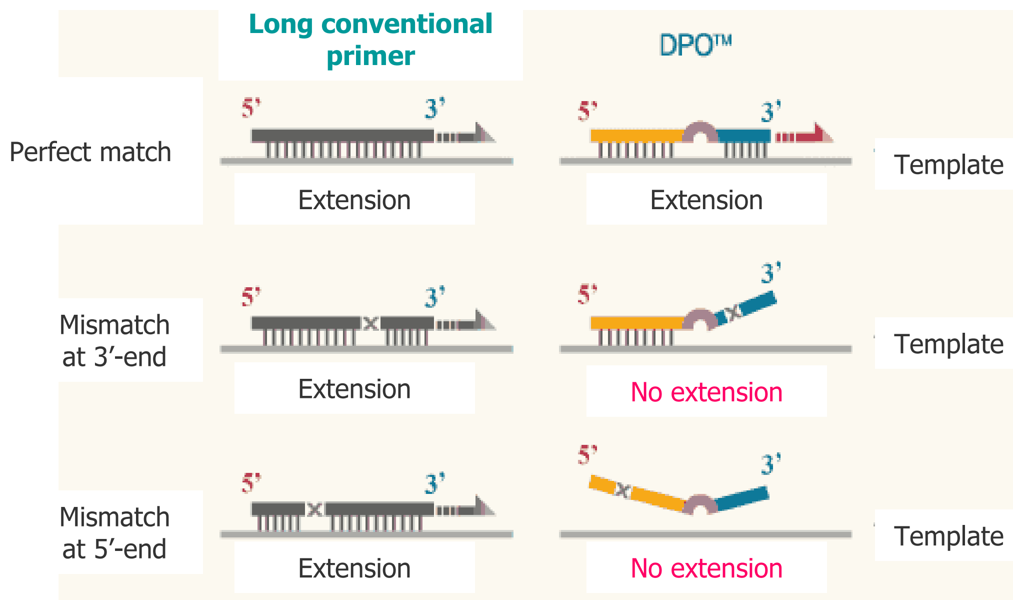


Although the longer 5′-segment binds a non-target site, the short segment resist non-specific extension



The short 3′-portion alone fails to make a priming at annealing temperature

Comparison between conventional primers vs. DPO



Another major advantage is that DPO™ makes primer design extremely simple and easy

Since two separate priming reactions provide a primer with a comfort zone (high tolerance) in annealing. Following the first stable priming reaction by Stabilizer, the second critical priming reaction by Determiner gives one additional chance to correct the specificity. For this reason, DPO™ does not require a rigid optimization of PCR conditions and primer search parameters including primer length, GC content, annealing temperature, and secondary structure (hairpin, self or cross dimer).