

**DNA PROFILING OF ASPERMIC SEMEN SAMPLES FROM VASECTOMIZED  
MALES BY USING Y-PLEX™ AMPLIFICATION KIT**

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Amplification of Y-STR loci provides critical information during analysis of male-female mixture samples such as rape case. Analysis of mixture sample from rape cases, typically, involves differential extraction of sperm cells followed by evaluation of autosomal STR loci. However, this conventional approach has limitations when the male donor in the rape case is either vasectomized or azoospermic and sperm cells are not present. The use of Y-PLEX™6, which enables amplification of six STR loci on Y-chromosome namely DYS 19, DYS 385, DYS 389II, DYS 390, DYS 391, and DYS 393 in a single PCR amplification provides valuable results in such cases. Since the Y-PLEX™6 amplifies only Y-chromosomal loci, it is possible to obtain the male profile in a mixture sample of male and female donors.

Post-vasectomized semen sample (N=6) after at least four months of surgery were collected by masturbation in a clean sterile container. After liquefaction, the pre- and post-centrifugation samples were observed under phase contrast microscope to confirm the absence of any mature spermatozoa. DNA was extracted from aliquots (200 µl) of semen samples by phenolchloroform method, amplified by using Y-PLEX™6 and analyzed on 310 Genetic Analyzer. We have observed a wide variation in the yield of extracted DNA from 0.06 ng/µl to 5.0 ng/µl, which is possibly due to the varied number of epithelial and/or white blood cells that are occasionally present in these samples. The analytical calculation reveals that, the profile for all six loci can be obtained from a sample equivalent to about 40 µl semen volume of low DNA quant sample. Thus, a small stain of semen from vasectomized male is sufficient to obtain the DNA profile.

In conclusion, this technique is sensitive enough that a DNA profile of vasectomized males, either individually or in a mixture, can be easily obtained by analysis of a small stain of semen.