Rheonix CARD™ Procedure

The Rheonix HPV CARD™ test (not yet cleared by FDA for human IVD applications) is capable of performing a fully automated analysis of human cervical samples for the presence of 20 clinically relevant HPV types in a multiplex PCR assay. Briefly described, after the operator introduces the vaginal swab, previously collected and stored in a transfer buffer, into the device (the only operator step), the remaining operations are all performed by the Rheonix CARD™.

1. Cells are lysed and DNA is purified.
2. DNA is subjected to multiplex PCR in the presence of biotinylated primers resulting in amplification of any one of the 20 target HPV types and the human β-globin gene.
3. HPV and globin amplicons are denatured and delivered onto the DNA microarray reactor followed by hybridization to the specific capture probes.
4. Hybridized strands are detected via incubation with streptavidin conjugated HRP and substrate (TMB).
5. Image analysis identifies the specific HPV type(s).

Representative Results

The above filter image resulted following processing of clinical sample #9030 (see Table on the right) on the Rheonix HPV CARD™. The three corner spots labeled with “SP” are spotting controls; the two spots in the second row show the positive HPV 16 result; and the third row shows detection of globin used as an internal control indicating successful nucleic acid purification. All probes are spotted at the same concentration except HPV 16, 18, and globin, which are spotted at 3 different concentrations.

Comparison against an FDA-approved product

Evaluation of samples that were negative on an FDA-approved product

Conclusions

The Rheonix HPV CARD™ test provides a fully automated system for the rapid and reliable molecular detection of 20 clinically relevant HPV types. Furthermore, due to the low cost and minimum training required in using the Rheonix CARD™ product, this test will have widespread application potentials in both industrialized and developing nations.