Increased Efficiency with New Assay for Zygosity Analysis

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We developed a new and improved assay for zygosity determination in twins using the MassARRAY® system (Agena Biosciences). The goal for the new design was to facilitate a faster, simpler, and more cost-effective zygosity analysis, by designing a single multiplexed reaction containing targets with the highest minor allele frequencies based on data from testing 25,000 samples.

METHOD

A custom assay containing 47 SNPs was developed in 2007 using iPLEX Gold chemistry to determine the likelihood of zygosity of twins – whether they are monozygotic (identical) or dizygotic (fraternal). Since then, 12,800 twin pairs were tested at MAF. In collaboration with the Swedish Twin Registry, a new and cost effective assay containing 37 autosomal SNPs and 2 sex markers on the zinc finger of the X and Y chromosomes was developed using iPLEX Pro chemistry. The new design was verified against 45 reference samples with known genotypes and further tested against DNA from saliva of 310 twin pairs. The zygosity score was calculated using odds ratios of monozygosity (MZ), taking into account an error rate of 0.1%.

RESULTS

The call rates for the new and old assay designs were 99.7% and 94.9%, respectively and the genotypes were 100% concordant between the two designs. The results for reference samples were also concordant with the HapMap database, with a call rate of 99.8%. The sex markers scored 100% correctly in all runs. Odds ratios for MZ score using the new and old assay design was greater than $6x10^6$ and $6x10^7$, respectively. Odds ratios for dizygotic score (DZ) was 0 for both designs.

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