Heritability of expression profiles in twin families

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Introduction

Monozygotic (MZ) twins have identical genotypes, but hardly ever identical phenotypes. At which level (methylation status, RNA or protein expression) do differences arise? We looked at expression profiles in MZ twin pairs and their same-sex sibs.

Subjects & procedure

There were 22 participants from 8 families. In 6 families a MZ twin pair and a same-sex sib (12 males and 6 females) and in 2 families a MZF pair took part. Average age was 18.0 yrs. All Ss lived at home with the parents. Blood withdrawal took place in the lab after overnight fasting around 10.30 am. About 20 minutes after collection, 2.5 ml blood from one heparin tube was added to a PAX tube with code A. After addition of lipopoly-saccharide (LPS; 10 ng/ml blood) to the same heparin tube, the tube was inverted gently and stored at 37°C during 5 hours. Then, 2.5 ml of the challenged blood was added to a PAX tube with code B. The PAX-A tube was, after 5 hours at room temperature, inverted gently and stored at -20°C. After leaving the PAX-B tube 1.5 hour at room temperature, the tube was inverted and stored at -20°C.

Expression profiling

Agilent 44k human whole genome microarrays were used, hybridising baseline whole blood RNA (Cy3) and LPS challenged (Cy5) for each participant on one array. Background correction (Edwards, offset 30), within-array normalization (Loess) and between normalization (Aquantile) was performed.

Analyses & Results 1

LPS is a potent stimulus for blood cells: around 5% of the detectable genes were regulated > 4-fold by LPS in all samples.

Probes with expression values between 6.5 and 15.3 were included for further analyses (12250 genes). Data were available for 19 out of 22 participants.

We first selected genes that on average showed a mild regulation of 1.75-fold by LPS-induction (3384 genes). Using these genes the hierarchical clustering of the participants for basal and LPS-induced samples showed a good segregation of the two blood samples (basal expression, Green; LPS-induced expression, Red). In addition, a clear segregation was observed for twin samples (xxx_1 and xxx_2), indicating that these genes show familial clustering.

For each gene we estimated the heritability, using moment estimators (for computational efficiency). The histogram plots heritabilities. Around 40% of heritabilities is zero, or close to zero. To test (H0 : h² = 0) whether they are significantly different from zero we used the marginal permutation test. This test obtains the null-distribution by permutation of the twin-sibling labels. The p-value for each gene is then calculated as the proportion of the null-distribution that exceeds the observed heritability for that gene. 446 genes have a marginal p-value < 0.01, and 1148 a marginal p-value < 0.05.