Psychosocial stress and cortisol stress reactivity predict breast milk composition

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Maternal and infant anthropometrics

Maternal height was measured using a stadiometer. Weight and body fat were measured with Tanita SC-240 MA Body Scale (Tanita Corp., Japan). Infant weight was measured to the nearest 1 g by using a hospital analog infant scale. The supine length was measured to the nearest 0.1 cm using an infant measuring board (Seca GmbH, Germany).

Breast milk sample analysis

Content of macronutrients

Milk samples of 10 ml were slowly warmed to 40 °C before testing and homogenized with MIRIS Sonicator (an ultrasonic homogenizer) (1.5 s/1 ml). Each sample was tested three times, using 3 ml of milk per one replication, and the average from 3 measurements was reported.

Content of fatty acids

Separation was carried out under the following conditions: column HP-88 (100m x 0,250 mm x 0.20  $\mu$ m), column temperature from 70°C (5min) to 240°C (4°C/min), the carrier gas - helium (20 cm/s), injection spray 1 $\mu$ l, 260°C, split 100:1. DHA and AA were identified based on retention times obtained for standards and by comparing their mass spectra with the standards.

Salivary sample analysis

Enzyme-linked immunosorbent assays is a solid phase based on the principle of competitive binding. The optical density was determined with a microplate reader (LEDETECT96, Labexim Products, Austria) at 450 nm. The sensitivity provided by the manufacturer is 0,09 ng/ml, with a detection range of 0,09 - 30 ng/ml. All the analyzed samples felt into this range.