Influence of gestational age and day of life on IRT and PAP in cystic fibrosis screening

L. Mense1, M. Stöpsack2, J. Hammermann1, 1University Hospital Carl Gustav Carus, Dresden, Department of Pediatrics, Dresden, Germany; 2Dresden University of Technology, Institute of Clinical Chemistry and Laboratory Medicine, Dresden, Germany

Objectives: The determination of immunoreactive trypsin (IRT) and pancreatitis-associated protein (PAP) in dried blood spots is one possibility of conducting a neonatal cystic fibrosis screening. In the present study we examined the influence of gestational age and day of life on IRT and PAP.

Methods: Data from the newborn screening center of Dresden, Germany in the years from 2008 to 2010 were studied. IRT measurement was done with AutoDELFIA Kit (PerkinElmer Turku). PAP measurement with MucoPAP Kit (DYNABIO Marseille). All specimens were tested on IRT and for samples exceeding 50 ng/mL PAP measurement was added. For this study we analyzed all data of newborn screening cards which were taken in the age of 36–72 h after birth and contained information of gestational age.

Results: 42,607 newborn screening results met the inclusion criteria. In 1889 cases determination of PAP was executed. While IRT showed a minor correlation to gestational age (r = 0.540, p < 0.001), PAP and IRT correlated strongly as well (r = 0.484, p < 0.001). Day of life showed only a minor correlation to IRT (r = −0.035, p < 0.001) and PAP as well (r = −0.050, p = 0.029).

Conclusion: A severe influence of gestational age on IRT could not be seen while PAP was strongly affected. Therefore, borderline PAP results in preterm babies control samples and sweat testing should be initiated liberally even after completed 32 weeks of gestation (final time for other screenings).

Improved cut off combination for IRT and PAP in newborn screening for cystic fibrosis

M. Stöpsack1, J. Hammermann2, 1Medical Faculty of Dresden University of Technology, Clinical Chemistry and Laboratory Medicine, Dresden, Germany; 2Dresden University of Technology, University Children’s Hospital, Pulmology, CF-Center, Dresden, Germany

Background: SARLES introduces the use of Pancreatitis Associated Protein (PAP) as second step after IRT elevation in newborn screening for Cystic Fibrosis (CF) to minimize the problems with low IRT specificity, parental stress and delayed diagnosis after 2. IRT and unwanted carrier detection.

Methods: IRT measurement was done with AutoDELFIA Kit (PerkinElmer Turku), PAP measurement with MucoPAP Kit (DYNABIO Marseille). Blood samples taken 36 to 72 h after birth, gestational age >32 weeks, were tested for IRT. For samples exceeding 50 ng/mL PAP measurement was added. All newborns with result combinations IRT > 50 ng/mL and PAP > 1.8 ng/mL or IRT > 100 ng/mL and PAP > 1.0 ng/mL or those with IRT > 150 ng/mL alone were referred to pilocarpin iontophoresis. To improve sensitivity and specificity of marker combination, indication of further diagnosis was set on IRT cut off value >60 ng/mL and for IRT*PAP product >100.

Results: We tested 50,693 newborn samples. 1135 (2.2%) of them had IRT > 50 ng/mL and got PAP measurement, whereof 110 (0.21%) were further elevated in IRT*PAP combination according to SARLES. The use of the higher IRT cut off together with IRT*PAP product would have reduced the recall rate to 95 (0.18%) and detected all known cases of CF from screened population (by now seven patients) and from selective analyses (18 patients, 3 of them don’t fulfill SARLES criteria).

Conclusion: Biochemical CF screening by IRT*PAP product leads to improved recall rate and appreciable increased sensitivity. Compared to mutation analysis it avoids undesirable carrier identification and high analysis costs. Compared to IRT/IRT strategy it avoids second blood sample and prolonged parental stress.

The performance of a new, high throughput neonatal IRT assay for cystic fibrosis screening on the PerkinElmer automated GSP® instrument

P. Kerokoski1, H. Lindroos1, M.-L. Mäkinen1, R. Kinos1, P. Nyström1, R. Gustafsson1, A. Hiekkanen1, J. Seppälä1, 1PerkinElmer, Inc., Specialty Diagnostics, Wallac Oy, Turku, Finland

The objective was to evaluate the performance of a neonatal immunoreactive trypsin(ogen) (IRT) kit used with the new Genetic Screening Processor (GSP®). The new GSP® IRT kit is based on the current AutoDELFIA® IRT kit chemistry (a time-resolved immunofluorometric assay), which was modified to be compatible with the GSP® instrument.

Precision, interference, correlation, and screening performance of the new GSP® IRT kit (catalog number 3306–0010) was studied using dried blood spots as samples. Precision of the GSP® IRT kit was below 8.5% throughout the measuring range (9–500 ng/mL blood) even when a stored calibration curve (run 20–24h earlier) was used. Icteric, lipemic, and hemolytic specimens did not interfere with the assay; neither did EDTA, Na-citrate, or Li-heparin. The correlation between the GSP® IRT and the B005–212 AutoDELFIA® IRT kits was found to be y = 0.94 × 0.35 (weighted Deming regression). The newborn population medians and upper percentiles were similar with the methods compared. The IRT values of all 12 confirmed CF-positive cases were clearly separated from unaffected cases with both the GSP® IRT and the AutoDELFIA® IRT kits, i.e. the CF cases were above the 99th percentile of the newborn population (n = 2106). The stability studies showed that the reagents (assay buffer, tracer) were stable for 14 days in the cooled reagent carousel of the instrument.

The new GSP® IRT kit offers the benefits of the new GSP® instrument (easy to use, high throughput, calibration curve valid for 24 hours, barcoded reagents with long on-board time) while preserving the excellent performance of the current AutoDELFIA® IRT kit.