Urine based newborn screening study applying high-resolution NMR spectroscopy in Turkey

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Background: Approximately 1,000 neonates are affected by congenital metabolic diseases in central Europe and 1,500 in Turkey. Undetected and untreated these diseases can lead to irreversible organ failures, invalidity or death. Fully automated NMR spectroscopy of body fluids is used as an analytical approach for diagnosis of known, but also as yet unknown inborn errors of metabolism.

Body fluid \(^{1}\)H-NMR spectroscopy: NMR spectroscopy of body fluids can be a complementary technique to find the diagnosis of metabolic diseases. \(^{1}\)H-NMR spectroscopy of body fluids shows the majority of proton-contacting compounds and therefore provides an overall view of metabolism. NMR spectroscopy of body fluids may be considered as an alternative analytical approach for diagnosing known, but also as yet unknown, inborn errors of metabolism.

Results: After identification of metabolites in NMR spectra, simple integration of selected metabolites of interest yields fully quantitative information on metabolite concentration. Figure 3 gives an example. The concentrations of D-galactose (246 mmol/mol), 4-hydroxyphenylacetic acid (1274 mmol/mol), double of what has been described as pathological (631 mmol/mol) \cite{4}.

Follow-up investigation: It is necessary to monitor children which were identified by statistical analysis. We plan to collect urine samples from these subjects at a later point in time. This will allow to distinguish cases were metabolic levels have returned to normal from pathological cases, thus significantly reducing the number of false positive screening results.

Statistical analysis: The statistical analysis and quantification of metabolite concentrations will be based on a combination of 1D-spectra and fast 2D-J-resolved spectra. Here, the 2D-spectra support the spectral identification and deconvolution based quantification of metabolites identifying line position and multiplet structure as obtained from a 2D-spectrum. Further statistical modules developed at Bruker BioSpin GmbH use a normal model for untargeted screening and allow the detection of unknown diseases for Turkish babies. In addition, targeted screening is used to detect known diseases.

Study design: Open, one-arm, non-interventional study. Data from 1000 healthy Turkish neonates will be included to explore the range of variation (concentration and chemical shift) of specific metabolites in this population and to identify the pathological thresholds of these metabolites in urine.

Study Objectives: Primary objective of the study was to explore the range of variation (concentration and chemical shifts) of specific metabolites without clinically relevant findings. Secondary objective was the integration of the results from a healthy population of neonates into an NMR-knowledge base to perform routine and completely automatic screening for congenital metabolic diseases using targeted and untargeted approaches out of one measurement per sample.

Patients and methods: Urine samples of 690 neonates from 8 centers in Turkey were investigated by using fully automated NMR spectrometers in two different laboratories (INFAl and Bruker).

Results: It is known that healthy neonates can have different pathological metabolites in high concentration after birth. It is important to compare concentration ranges of these pathological metabolites with clinical data to reach a reliable conclusion for the future development of the child.

Conclusion:
- Establish a statistical normal model of 'healthy' urine spectra
- Establish automatic quantification and 'normal ranges' for common metabolites
- Identify pathological metabolites that can be used as disease markers
- Integrate into a comprehensive screening test for newborn health
- The statistical analysis and quantification of metabolites allow to develop a normal model in a specific population and also a general assessment of neonatal health state

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References