Quantitative faecal haemoglobin test (FIT) in children with gastroenterological diagnosis

Kocna Petr, Szitányi Peter, Kelífová Silvie, Vaníčková Zdislava, Zima Tomáš
Department of Paediatrics and Adolescent Medicine and Institute of Medical Biochemistry and Laboratory Diagnostics, 1st Faculty of Medicine Charles University and General University Hospital, Prague, Czech Republic

SUMMARY
Quantitative analysis of haemoglobin (Hb) level in stool samples (FIT) using automated analysers is routinely used in colorectal screening in adults, but has never been described in children diagnostics. The aim of this study is analysis of faecal Hb level in children samples in years 2010 - 2016 tested by FIT Faecal Blood detection OC-Sensor (Elwah).

Methods
From the total number of FIT samples analysed (n=2488) in children (age 0.1-14 years, mean 7.65, median 1232/2488 samples with Hb level > 100 µg/g of stool (n=226, 9.2%) were selected. By retrospective analysis of the possible causes of FIT positivity we identified (n=170, 72%) cases with confirmed gastroenterological diagnoses. Six groups according to the most widely represented gastroenterological diagnoses were created - IDO (n=68, 28.5%) of the total number of gastroenterological diagnoses, Crohn disease (n=17, 10%), gastrooestroduodenal reflux (n=17, 10%), cow’s milk protein intolerance (CMPi) (n=27, 10.3%) and small faeces (n=17, 10%). The sixth group consisted of those gastroenterological diagnoses not overlap classified, marked as others (n=28, 12%).

Results
The median values of faecal Hb levels were in the group of IDO 102.3 µg/g (range 20.4 - 658.5 µg/g), in acute gastroenteritis 220.8 µg/g (range 23.6 - 598.8 µg/g), in CMPi 110.4 µg/g (range 22.3 - 571.6 µg/g) in gastrooestroduodenal reflux 43.2 µg/g (range 15.6 - 98.5 µg/g) and in faeces 37.6 µg/g (range 25.8 - 1776 µg/g). In the group of others 48.5 µg/g (range 25.4 - 396.4 µg/g).

Conclusions
Quantitative analysis of haemoglobin (Hb) level in stool samples we described in children most widely gastroenterological diagnoses. Evaluation of cut-off criteria values for future diagnostic procedures will follow in future investigations. The study is supported by the Research Project RVO VFN 84168.

GASTROINTESTINAL BLEEDING IN CHILDREN
Gastrointestinal bleeding in general is in children relatively rare compared to adults, but could be potentially life-threatening condition in childhood. In pediatric age bleeding differs according to the age and different conditions/diagnoses. Once the bleeding site could be upper and lower (gastrointestinal tract) acute and chronic, its detection in gastrointestinal tract may be present from non-GI organs (mucosalis, colorogetic, stomach, renal pathology, others). Clinically bleeding presents as teneatiae, anemia, metrorrhagia or occult faecal bleeding.
Most frequent reasons in different age groups are mentioned below:
Upper gastrointestinal tract bleeding in neonates: stress gastritis, hemorrhagic diseases of the newborn
Lower gastrointestinal tract bleeding in neonates: necrotizing enterocolitis, intramural bleeding of GER, gastritis.
Lower gastrointestinal tract bleeding in children aged 1 month to 1 year: neoplastic causes caused by GER, gastritis.
Lower gastrointestinal tract bleeding in children aged 1 month to 1 year: anal fissures, intussusceptions, cows milk protein allergy, colitis
Upper gastrointestinal tract bleeding in older children: peptic ulcers, esophageal varices result from portal hyper trophy, tkg varicose, drugs and alcohol abuse.
Lower gastrointestinal tract bleeding in older children: polyph, juvenile type, Meckel diverticulum, vascular lesions, IBD, Inflammatory diarrhea.
In clinical practice, detailed clinical assessment of a child is required for therapeutic intervention, but age-specific treatment and management strategies are not the topic of this presentation. Relevant interpretation of faecal occult bleeding could help the clinician in stratification of diagnostic approach and lead to prediction of our patients group in decision process.

FIT - IMMUNOCHINEMICAL METHODS FOR Fecal OCCULT BLOOD TEST

The principle of chemical tests to detect occult blood is based on the test that haemoglobin and its deoxynucleotide is in a similar way to peroxidase, by catalyzing the transfer of an oxygen atom from the peroxidase to a chromogen such as neoviline, o-coumaric or amperometric. Oxidation of the chromogen is indicated by the production of a blue, red or pink color.

FIT - IMMUNOCHINEMICAL METHODS FOR Fecal OCCULT BLOOD TEST

Modern fecal occult blood testing is now being an immunochromatographic test that is specific for human hemoglobin. This test utilizes a qualitative, rapid, sensitive and cost-effective method for the detection of blood. This method has high degree of analytical sensitivity.

QUANTITATIVE FIT METHOD

Haemoglobin in stool was measured using quantitative immunochromatographic method.Semiconductor was carried out by following the instructions of producer.

Stool was sucked up by the brush with the sampling brush on the stool surface and immersed into the solution in test tube. The tube kept in the temperature between 4 - 8°C 18 htermeration.
Samples were analysed continuously during a one week on the OC Sensitised test analyzer (Ikossi Chemical Co., Tokyo, Japan).

Quantitative assessment of human haemoglobin in stool (gHb/GST) outstanding the level of human haemoglobin using polyclonal antibody against human haemoglobin.
The analysis is performed using turbidimetric measurement by 680 nm in ethanol of measurement 5 - 2000 ng/ml Hb. This reaction monoclonal human haemoglobin antibodies, which had been fixed to line, meet with haemoglobin in the sample resulting in a new application reaction.
The change in optical density of the reaction solution is analyzed. Optical density expresses in proportion to higher concentrations of Hb/GST in the sample.

FIT RETROSCOPIC ANALYSIS OF FIT TEST

Relative representation of individual diagnostic groups: Fissures ani (FIT), IBD - inflammatory bowel disease (IBD), Hemorrhoid-Schleman purpura with abdominal pain (HSP), Acute gastroenteritis (ACE), Cow's milk protein intolerance (CMPi), Gastrooestroduodenal reflux (GERD), other gastroenterological diagnosis (OTHER) detected by FIT with three cut-off limits: Hb values > 20 µg Hb/g stool [n = 217], 10 - 20 µg Hb/g stool [n = 51], 4 - 10 µg Hb/g stool [n = 113].

CONCLUSIONS

- Previously used Haemocult test changed in 2010 to OC-Sensor FIT test
- FIT positivity with the cut-off of 15 µg Hb/g faeces is defined for CRC screening in adults
- Indications in pediatric medicine are quite different that CRC screening in adults
- FIT cut-off should be different for upper and lower part of digestive system due to marked degradation of glubin
- FIT test analytical sensitivity are 100-1000 times higher than that for previously used Haemocult
- FIT cut-off should be evaluated for every considered diagnosis