la (24,3% prema 13%; P < 0,001) u usporedbi s ispitanicima čije su koncentracije mokraćne kiseline u serumu bile unutar referentnog raspona.
Zaključak: Hiperurikemija je povezana s koncentracijom glukoze natašte i dislipidemijom u općoj populaciji.

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HDL-cholesterol (24.3% versus 13%; P < 0.001) compared to subjects with serum uric acid levels within the reference range.
Conclusion: Hyperuricemia is associated with fasting glucose concentrations and dyslipidemia in general population.

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P03 – Hematologija i koagulacija

P03-1 (Usmeno priopćenje)

Prikaz slučaja: određivanje krvne slike kod izrazite lipemije

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Uvod: Spektrofotometrijsko određivanje koncentracije hemoglobina podložno je utjecaju hipertrigliceridemije u uzorku. Izmjerena vrijednost hemoglobina je netočno previsoka, a također i računski parametri MCH i MCHC.
Cilj: Kako točno odrediti koncentraciju hemoglobina i računske parametre u izrazito lipemičnom uzorku.
Materijali i metode: Uzorak krvne slike izmjeren je na hematološkom brojaču ADVIA 2120 koji određuje hemoglobin dvjema metodama: spektrofotometrijski i mjerenjem CHCM. Zbog velike razlike dobivenih vrijednosti određen je i hemoglobin u plazmi i trigliceridi u serumu.
Rezultati: Spektrofotometrijski hemoglobin iznosio je 158,0 g/L, a računski preko CHCM-a 93,0 g/L. Izmjereni trigliceridi bili su 120,66 mmol/L. Izmjereni hemoglobin u plazmi bio je 89 g/L što je korišteno za računsku korekciju spektrofotometrijskog hemoglobina čija je vrijednost nakon preporučenog načina korekcije iznosila 92,3 g/L.
Zaključak: Metoda računskog određivanja hemoglobina putem CHCM-a potpuno je neovisna o lipemiji.

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P03 – Haematology and coagulation

P03-1 (Oral presentation)

Case report: complete blood count in severe lipemic sample

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Background: Severe lipemia is a known interference that can falsely elevate a hemoglobin result obtained from colorimetric hemoglobin method. When hemoglobin is falsely increased, the MCH and MCHC are also falsely increased.
Aim: Determination and correction of hemoglobin concentration and calculated parameters in severe lipemic sample.
Material and methods: Complete blood count was determined on ADVIA 2120 which determinates hemoglobin by two methods: colorimetric and directly measuring CHCM to calculate cellular hemoglobin. Since the results between these two methods were significant, we determined plasma hemoglobin and triglyceride in serum.
Results: Colorimetric hemoglobin concentration was 158.0 g/L. Cellular hemoglobin was 93.0 g/L. Triglycerides in serum were 120.66 mmol/L. Hemoglobin in plasma was 89.0 g/L which we used in correcting hemoglobin in whole blood. After correction hemoglobin concentration was 92.3 g/L.
Conclusion: Using CHCM to calculate hemoglobin is unaffected by lipemia.

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P03-2 (Usmeno priopćenje)

Postotak hipokromnih eritrocita kao pokazatelj funkcionalnog manjka željeza kod bolesnika na hemodijalizi

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Uvod: Hipokromni eritrociti osjetljiv su pokazatelj sideropenične eritropoeze i funkcionalne anemije kod bolesnika na hemodijalizi liječenih eritropoetinom. Cilj rada bio je prikazati rezultate hematoloških i biokemijskih parametara dobivenih sistematskim pregledom hemodijaliziranih bolesnika i pronaći njihovu međusobnu povezanost.
Materijali i metode: U istraživanje su uključena 44 bolesnika. Laboratorijska dijagnostika uključivala je određivanje hemoglobina (Hb), postotka hipokromnih eritrocita (%HYPO), retikulocita, željeza, TIBC, UIBC i feritina. Hematološki parametri određeni su na hematološkom analizatoru ADVIA 120 (Bayer, SAD), željezo, TIBC i UIBC na analizatoru Olympus AU400 (Olympus, Japan) i feritin na analizatoru ARCHITECT i2000 SR (Abbott, SAD).
Rezultati: U skladu sa smjernicama za liječenje anemija u bolesnika s kroničnim zatajenjem bubrega u ispitivanoj skupini Hb < 110 g/L nađen je kod 29 (66%) bolesnika, %HYPO > 2,5 kod 13 (29%), %HYPO > 10 kod 2 (4,5%), feritin < 100 ng/mL kod 14 (31,8%) bolesnika. Za procjenu dijagnostičkog značaja %HYPO u otkrivanju funkcionalnog manjka željeza upotrebljena je ROC analiza uz koncentraciju feritina < 100 ng/mL kao kriterij. Izračunata površina ispod krivulje (AUC = 0,72; 95%CI = 0,565-0,845) i pripadajuća optimalna granična vrijednost (%HYPO > 2%) ukazuju na mogući nedostatak željeza uz dijagnostičku osjetljivost i specifičnost koje iznose 64,2% i 76,7%.
Prema Spearmanovoj korelaciji nađena je značajna povezanost između %HYPO i Hb (P = 0,046) i %HYPO i UIBC (P < 0,001).
Zaključak: Dobivene vrijednosti mjerenih parametara pokazuju povezanost %HYPO s Hb i %HYPO s UIBC. Međutim, prema prihvaćenim smjernicama za liječenje anemije nisu dostignute preporučene ciljne vrijednosti za hemoglobin i feritin.

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P03-2 (Oral presentation)

The percentage of hypocromic red blood cells as a marker of functional iron deficiency in hemodialysis patients

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Introduction: Hypocromic red blood cells are sensitive marker of sideropenic erythropoiesis and functional anemia in hemodialysis patients treated with erythropoietin. The aim of this study was to show the relationship between the results of some hematological and biochemical parameters obtained from hemodialysis patients on their period checkup.
Materials and methods: The study included 44 patients. Laboratory diagnostics included determination of hemoglobin (Hb), percentage of hypocromic red blood cells (%HYPO), reticulocytes, iron, TIBC, UIBC and ferritin. Hematological parameters were determined on cell counter ADVIA 120 (Bayer, USA), iron, TIBC and UIBC on Olympus AU400 analyzer (Olympus, Japan) and ferritin on ARCHITECT i2000 SR analyzer (Abbott, USA).
Results: In accordance with the guidelines for treating anemia in patients with chronic kidney failure in examinated group, Hb < 110 g/L was found in 29 (66%) patients, %HYPO > 2.5 in 13 (29%), %HYPO > 10 in 2 (4.5%), ferritin < 100 ng/mL in 14 (31.8%) patients. For the assessment of diagnostic significance of %HYPO in discovering the functional iron deficiency, ROC analysis is used with cut off ferritin concentration < 100 ng/mL as a criteria. Calculated area under the curve is 0.72 (95% CI = 0.565-0.845) and the corresponding optimal cut off value of %HYPO > 2% indicate a possible iron deficiency with a diagnostic sensitivity and specificity 64.2%, and 76.7%.
According to Spearman correlation a significant relationship between %HYPO and Hb (P = 0.046) and %HYPO and UIBC (P < 0.001) was found.
Conclusion: Measured parameters show a significant connection between %HYPO and Hb and %HYPO and UIBC. However, according to accepted guidelines for treatment of anemia the recommended target values for hemoglobin and ferritin have not been reached.

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P03-3 (Usmeno priopćenje)

Tromboelastometrija u trudnica s preeklampsijom

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Uvod: U toku normalne trudnoće dolazi do fizioloških promjena u mehanizmu koagulacije koje često rezultiraju hiperkoagulabilnošću. Smatra se da su mnoge komplikacije u trudnoći povezane s poremećajima zgrušavanja, kao npr. preeklampsija i eklampsija. Prevaga tromboksana (TXA2) nad prostaciklinom (PGI2) uzrokuje hipertenziju, oštećenje bubrega, aktivaciju trombocita i mikrotrombozu. Tromboelastometrija je metoda koja pruža informacije o najvažnijim poremećajima u zgrušavanju vrlo brzo od uzorkovanja u punoj krvi i predstavlja važan doprinos u razumijevanju i praćenju hemostatskih promjena.
Cilj: Svrha ovog rada je dokazati eventualne promjene u tromboelastogramu u trudnica s preeeklampsijom u odnosu na zdrave trudne žene.
Ispitanice i metode: U 28 zdrave trudnice (35 do 41 tj. trudnoće) i 34 trudnice s preeklampsijom (35 do 39 tj.trudnoće) učinjen je tromboelastogram (Rotem delta, Pentapharm GmbH).
Određivani su parametri CT - reakcijsko vrijeme, CFT - vrijeme stvaranja stabilnog ugruška, MCF - maksimalna čvrstoća ugruška, alfa kut i postotak redukcije MCF nakon 60 minuta.
Rezultati: Svi parametri dobiveni tromboelastometrijom i u zdravih trudnica i u trudnica sa znakovima preeklampsijea ukazuju na hiperkoagulabilnost, ali razlike između te dvije skupine nisu bile statistički značajne.
Zaključak: Smanjena produkcija PGI2 u majčinoj krvi i placentnoj cirkulaciji smatra se etiološkim faktorom patofizioloških promjena kod preeklampsije što dovodi do vazokonstrikcije i tromboze uteroplacentarnih arterija. TEG je metoda kojom se ne mogu mjeriti te lokalne promjene, ali može biti vrijedan doprinos u praćenju hemostatskih poremećaja.

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P03-3 (Oral presentation)

Tromboelastometry in pregnant women with preeclampsia

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Introduction: Pregnancy is considered a hypercoagulable state. There is much interest in the relationship between coagulation status and complications of pregnancy. Increased incidence of thromboembolic phenomena have been reported in pregnant women. Pre-eclampsia and eclampsia may complicate some pregnancies and is often associated with abnormalities of hemostasis. The tromboelastograph (TEG) has been proposed as a useful, inexpensive tool to screen for patients with hypercoagulable state. A TEG measures whole blood coagulation and fibrinolysis.
Aim: The purpose of this study was to document tromboelastografic (TEG) changes in preeclampsia or eclampsia and to compare these resuts with the same results in health, normal, term pregnant women.
Patients and methods: A TEG was performed in 28 normal, term pregnant women (35-41 weeks of pregnancy) and in 34 preeclamptic women admitted to the delivery (35-39 weeks of pregnancy) using native whool blood (Rotem delta, Pentapharm GmbH). The TEG variables included CT - clotting time, CFT - Clot formation time, MCF - Maximum Clot Firmness, alpha angle and percentage of reduction in MCF at 60 minutes.
Results: TEG parameters for both groups showed that term pregnant women as well as preeclamptic women were in hypercoagulable state, but no significant differences between these two groups were observed.
Conclusion: Although standard laboratory tests are still necessary to detect coagulation abnormalities the TEG is useful as it is compact, easily located within the delivery suite, relatively easy to use and produces initial results very quickly. Preeclampsia is associated with endothelial damage, impaired production of prostacycline and increased deposition of fibrin within the vascular bed. The TEG does not measure these local changes, but can be used to determine the effects on overall clotting.

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P03-4

Osjetljivost analizatora funkcije trombocita PFA-100 na von Willebrandovu bolest

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Uvod: PFA-100 je specifični analizator funkcije trombocita koji mjeri vrijeme nastanka trombocitnog ugruška (CT) u prisutnosti kolagena i adrenalina (CEPI), te kolagena i ADP-a (CADP). Cilj istraživanja bio je utvrditi osjetljivost PFA-100 na von Willebrandovu bolest (VWB) s obzirom na različite aktivnosti (VWF:RCo) i vrijednosti antigena (VWF:Ag) von Willebrandovog faktora.
Materijali i metode: Ispitanici su svrstani u skupine prema vrijednostima VWF:RCo/VWF:Ag: skupina N (> 50%), skupina 1 (40-50%), skupina 2 (30-39,9%), skupina 3 (20-29,9%) i skupina 4 (< 20%), te su im izmjereni CEPI-CT i CADP-CT.
Rezultati: U skupini s normalnim VWF:RCo (N = 145), dobiveni su rezultati unutar referentnog intervala: CEPI-CT u 82,1%, CADP-CT u 86,2% te za oba testa u 74,5% ispitanika. U skupini s normalnim VWF:Ag (N = 141), dobiveni su rezultati unutar referentnog intervala: CEPI-CT u 74,5%, CADP-CT u 78,0%, te oba testa u 66,7% ispitanika. U skupinama sa sniženim VWF:RCo, dobivene su sljedeće osjetljivosti za CEPI-CT, CADP-CT i kombinaciju oba testa: skupina 1 (N = 15): 60%, 46,7%, 40%; skupina 2 (N = 10): 50%, 60%, 50%; skupina 3 (N = 13): 76,9%, 92,3%, 76,9%. U skupini 4 (N = 18) je dobivena 100% osjetljivost za CEPI-CT, CADP-CT i kombinaciju oba testa. U skupinama sa sniženim VWF:Ag, dobivene su sljedeće osjetljivosti za CEPI-CT, CADP-CT i kombinaciju oba testa: skupina 1 (N = 8): 62,5%, 87,5%, 62,5%; skupina 2 (N = 7): 85,7%, 100%, 85,7%. U skupini 3 (N = 4) i skupini 4 (N = 8) je dobivena 100% osjetljivost za CEPI-CT, CADP-CT i kombinaciju oba testa.
Zaključak: PFA-100 pokazuje visoku osjetljivost na VWB, posebice kod nižih VWF:RCo/VWF:Ag, uz 100% osjetljivost kod ispitanika s vrijednostima < 20%.

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P03-4

Sensitivity of the Platelet Function Analyzer (PFA-100) as a screening test for laboratory diagnosis of von Willebrand disease

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Introduction: Many studies have shown high sensitivity of Platelet Function Analyzer 100 (PFA-100) to von Willebrand disease (VWD).
Methods: We evaluated sensitivity of PFA-100 to different degrees of VWF:RCo/VWF:Ag deficiency, by measuring closure times (CTs) with collagen/epinephrine (CEPI) and collagen/ADP (CADP) in groups with different VWF:RCo/VWF:Ag values: group with normal values (> 50%), group 1 (40-50%), group 2 (30-39.9%), group 3 (20-29.9%) and group 4 (< 20%).
Results: In the group with normal VWF:RCo values (N = 145), normal CTs were obtained in 82.1% cases with CEPI, 86.2% with CADP, and 74.5% cases with both tests. In the group with normal VWF:Ag values (N = 141), normal CTs were obtained in 74.5% cases with CEPI, 78.0% with CADP and 66.7% cases with both tests. In the groups with decreased VWF:RCo values, the calculated sensitivities for CEPI-CT, CADP-CT and both tests, were as follows: group 1 (N = 15): 60%, 46.7% and 40%, respectively; group 2 (N = 10): 50%, 60% and 50%, respectively; group 3 (N = 13): 76.9%, 92.3% and 76.9%, respectively; group 4 (N = 18): 100% for CEPI-CT, CADP-CT and both. In groups with decreased VWF:Ag values, the calculated sensitivities for CEPI-CT, CADP-CT and both tests, were as follows: group 1 (N = 8): 62.5%, 87.5% and 62.5%, respectively; group 2 (N = 7): 85.7%, 100% and 85.7%, respectively; group 3 (N = 4) and 4 (N = 8): 100 for CEPI-CT, CADP-CT and both.
Conclusion: PFA-100 as a screening test for VWD shows higher sensitivity towards lower VWF:RCo/VWF:Ag values, with 100% sensitivity for patients with values below 20%.

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P03-5

Usporedba sniženog broja trombocita na različitim hematološkim analizatorima

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Uvod: Iako hematološki analizatori precizno i točno određuju broj trombocita, ne mogu u potpunosti razlikovati trombocite od fragmenata blasta, eritrocita i precipitata krioglobulina. Cilj je bio ispitati odnos broja trombocita i utemeljenost vlastitih iskustvenih spoznaja o usporedivosti rezultata s različitih analizatora.
Materijali i metode: Broj trombocita usporedno je određen optičkom metodom na analizatorima Advia 120 (Bayer, USA), Sysmex XE-2100 (Toa, Kobe, Japan), Cell-Dyn Sapphire (Abbott Diagnostics, USA) i impedancijom na analizatoru Sysmex XE-2100. Ispitano je 30 uzoraka s normalnim brojem trombocita, 50 uzoraka s trombocitima 21-100 x 10^9/L i 20 uzoraka ispod 20 x 10^9/L trombocita prema vrijednostima s analizatora Advia 120.
Rezultati: U zdravih ispitanika i bolesnika s trombocitima 21-100 x 10^9/L korelacijski koeficijenti (r) i koeficijenti determinacije (R^2) između analizatora su bili r = 0,931; R^2 = 0,866. U bolesnika s trombocitima ispod 20 x 10^9/L koeficijenti su bili: Advia-Sysmeximp r = 0,781; R^2 = 0,611; Advia-Sysmexopt r = 0,616; R^2 = 0,380; Advia-Sapphireopt. R = 0,756; R^2 = 0,572. Uvjeti Passing-Bablok regresije zadovoljeni su kod zdravih ispitanika, a nisu kod bolesnika s trombocitima 21-100 x 10^9/L između: Advia-Sysmexopt (95% CI, b = 0,850-0,986) i Advia-Sapphireopt (95% CI, a = -10,833-(-2,134)) te kod bolesnika s trombocitima ispod 20 x 10^9/L između Advia-Sapphireopt (95% CI, a = -8,167-(-0,875)). 95 % granice podudaranja srednjih razlika iz sažetog Bland-Altmanovog dijagrama bile su za trombocite ispod 20 x 10^9/L između: Advia-Sysmeximp -8,829 do 4,528; Advia-Sysmexopt -8,341 do 9,441 i Advia-Sapphireopt -10,998 do 1,598.
Rezultati: Ispitivanje je pokazalo klinički značajno neslaganje niskih rezultata broja trombocita između analizatora. U svakodnevnom radu preporuča se ove bolesnike pratiti na istom analizatoru ili verificirati rezultate mikroskopski ili protočnom citometrijom.

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P03-5

Comparison of decreased platelet counts on different haematology analyzers

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Introduction: Although haematology analyzers provide the high precision and accuracy in determining the number of platelets, they cannot completely distinguish the platelets from other cell fragments. The aim of the study was to examine the correlation of platelet counts and own empiric cognitions about the comparability of results from different analyzers.
Materials and methods: The platelet counts were determined parallel by optical method on analyzers Advia 120 (Bayer, USA), Sysmex XE-2100 (Toa, Kobe, Japan), Cell-Dyn Sapphire (Abbott Diagnostics, USA) and by impedance method on analyzer Sysmex XE-2100. In the study were tested 30 normal subjects, 50 patients with the platelets 21-100 x 10^9/L and 20 patients with the platelets below 20 x 10^9/L toward values obtained on Advia 120. In normal subjects and patients with platelets 21-100 x 10^9/L the correlation coefficient (r) and coefficient of determination (R^2) between analyzers were r = 0.931; R^2 = 0.866. In patients with platelets below 20 x 10^9/L coefficients between analyzers were: Advia-Sysmeximp r = 0.781; R^2 = 0.611; Advia-Sysmexopt r = 0.616; R^2 = 0.380; Advia-Sapphireopt r = 0.756; R^2 = 0.572. Passing-Bablok regression were satisfied for the normal subjects but it were not satisfied for the patients with platelets 21-100 x 10^9/L between: Advia-Sysmexopt (95% CI, b = 0.850 to 0.986); Advia-Sapphireopt (95% CI, a = -10.833 to -2.134) and in patients with platelets below 20 x 10^9/L between Advia-Sapphireopt (95% CI, a = -8.167 to -0.875). 95 % limits of agreements of the mean differences obtained from the summarised Bland-Altman diagram were for platelets below 20 x 10^9/L between: Advia-Sysmeximp -8.829 to 4.528; Advia Sysmexopt -8.341 to 9.441; Advia- Sapphireopt -10.998 to 1.598.
Results: The study showed the clinical significant disagreement of results between analyzers in patients with decreased platelet counts. In the daily work, it is recommend observe these patients on the same analyzer, or making the additional verification of result by microscope or flow cytometry.

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P03-6

Analitička evaluacija testa Innovance D-DIMER

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Uvod: Sve veći broj zahtjeva za kvantitativnim određivanjem D-dimera u posljednjih je nekoliko godina rezultirao razvojem velikog broja novih, potpuno automatiziranih testova za kvantitativno određivanje D-dimera.
Materijali i metode: Izvršena je analitička evaluacija imunoturbidimetrijskog testa s česticama lateksa za kvantitativno određivanje D-dimera Innovance D-DIMER na koagulometru Behring Coagulation System (Siemens Medical Solutions Diagnostics).
Rezultati: Određivanjem nepreciznosti u seriji u uzorcima plazmi s niskim, graničnim, lagano i izrazito povišenim vrijednostima D-dimera dobiveni su koeficijenti varijacije (CV) od 2,1% do 5,5%, dok su vrijednosti CV-a za kontrolne uzorke s niskom i izrazito povišenom vrijednosti D-dimera iznosili od 5,5% do 8,4%. Utvrđena je dobra linearnost metode u mjernom području od 0,17 do 5,45 mg/L FEU, te donja granica analitičke osjetljivosti od 0,099 mg/L FEU. Za određivanje referentnog intervala uporabljeno je 40 uzoraka plazmi zdravih osoba, a dobivena gornja granica iznosila je 0,495 mg/L FEU. Usporedba dobivenih rezultata u 419 svježih uzoraka plazmi izvršena je s rezultatima dobivenim uporabom testa Vidas D-DIMER Exclusion na instrumentu mini Vidas (bioMérieux). lako je linearnom regresijskom analizom prema Passingu i Babloku dobivena visoka korelacija (y = 1,370x–0,108, r = 0,952, P < 0,001), analiza prema Blandu i Altmanu pokazala je lagano više vrijednosti s testom Innovance D-DIMER koje su bile izraženije s porastom vrijednosti. U klasifikaciji bolesnika u odnosu na graničnu vrijednost dobiveno je vrlo dobro slaganje rezultata između testova Innovance D-DIMER i Vidas D-Dimer Exclusion (kappa-koeficijent = 0,860; 95% interval pouzdanosti, 0,811-0,908).
Zaključak: Rezultati ovog istraživanja ukazuju da test Innovance D-DIMER ispunjava sve analitičke zahtjeve za rutinsku primjenu.

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P03-6

Evaluation of the Innovance D-DIMER analytical performance

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Introduction: A widespread use of D-dimer in recent years has led to the development of a number of new fully automated quantitative D-dimer assays.
Methods: We evaluated the analytical performance of the recently developed particle-enhanced immunoturbidimetric assay Innovance D-DIMER on the Behring Coagulation System analyzer (Siemens Medical Solutions Diagnostics).
Results: The within-run coefficients of variation (CVs) for samples with low, borderline, slightly elevated and extremely elevated D-dimer ranged from 2.1% to 5.5%, whereas the between-run CVs for control samples with low and extremely elevated D-dimer ranged from 5.5% to 8.4%. The assay method exhibited very good linearity in the working range between 0.17 and 5.45 mg/L FEU with the analytical detection limit of 0.099 mg/L FEU. The upper reference value determined in 40 plasma samples from apparently healthy volunteers was 0.495 mg/L FEU. For the method comparison study, the results obtained in 419 fresh plasma samples were compared with the results obtained with the Vidas D-DIMER Exclusion on the mini Vidas (bioMérieux). Linear regression analysis according to Passing and Bablok demonstrated a highly significant correlation (y = 1.370x–0.108, r = 0.952, P < 0.001). Bland and Altman difference plots demonstrated slightly higher results obtained with Innovance D-DIMER that were more pronounced with increasing values. Very good agreement between Innovance D-DIMER and Vidas D-Dimer Exclusion was observed in classifying patient results as above or below the cut-off value (kappa coefficient = 0.860; 95% CI, 0.811-0.908).
Conclusions: This study demonstrates that Innovance D-DIMER fulfills all analytical requirements for implementation in daily routine.

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P03-7

Procjena dijagnostičkog značaja hematoloških parametara u otkrivanju nedostatka vitamina B_{12} ili folata

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Uvod: Megaloblastična anemija je makrocitna anemija (MCV > 97 fL) koju karakterizira poremećaj sazrijevanja jezgre u odnosu na citoplazmu, a najčešće je razlog nedostatak vitamina B_{12} ili folata.
Cilj: Cilj ovog rada je ispitati povezanost hematoloških parametara s nedostatkom vitamina B_{12} ili folata u skupini bolesnika s MCV > 97 fL.
Materijali i metode: U istraživanje je uključeno 69 bolesnika (28 žena i 41 muškarac) s MCV > 97 fL, srednje dobi izražene medijanom (73 godine). Uz MCV određeni su slijedeći parametri: koncentracija hemoglobina, postotak makrocita, indeks segmenata, postotak hipersegmentiranih granulocita (%HS) te koncentracija vitamina B_{12} i folata. Hematološki parametri određeni su na hematološkom analizatoru ADVIA 120 (Bayer, SAD), %HS i indeks segmenata određen je pregledom razmaza periferne krvi svjetlosnim mikroskopom BH 2 (Olympus, Japan), a koncentracija vitamina B_{12} i folata određena je na analizatoru ARCHITECT i2000 SR (Abbott, SAD), CMIA imunokemijskom metodom.
Rezultati: Temeljni kriterij za provedenu ROC analizu bila je dijagnostička primjena hematoloških parametara u otkrivanju nedostatka vitamina B_{12} ili folata. Pri tome je analiza pokazala povezanost između nedostatka vitamina B12 i %HS. Za %HS izračunata površina ispod krivulje (AUC) je 0,656, a interval pouzdanosti (95% CI) iznosi 0,529 do 0,769, optimalna granična vrijednost (cut off) iznosi 7% te vrijednost za dijagnostičku osjetljivost i specifičnost 77,2% i 56,8%. Pri otkrivanju nedostatka folata niti jedan od ispitivanih parametara nije pokazao dijagnostički značaj.
Zaključak: Prema rezultatima ovog rada zaključujemo da je postotak hipersegmentiranih granulocita osjetljiviji pokazatelj deficita B_{12} u odnosu na druge hematološke parametre.

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P03-7

Assessment of diagnostic significance of hematological parametars in discovering the deficiency of vitamin B_{12} or folate

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Introduction: Megaloblastic anemia is makrocytic anemia (MCV > 97 fL) characterized by nuclear maturation defect and. The deficiency of vitamin B_{12} or folate is often the reason for that. The aim of this study is to show the relationship between the hematological parametars and the deficiency of vitamin B_{12} or folate in the group of patients with MCV > 97 fL.
Materials and methods: The study included 69 patients (28 women and 41 men) with MCV > 97 fL and medium age 73 years. Following parametars were measured: hemoglobin concentration, percentage of makrocytes, segment index, percentage of hypersegmented neutrophils (%HS) and the concentration of vitamin B_{12} and folate. Hematological parameters were determined on cell counter ADVIA 120 (Bayer, USA), %HS and segment index were determined on peripheral blood smear in bright field microscopy (BH-2, Olympus, Japan), and the concentration of vitamin B_{12} and folate were determined on the analyzer ARCHITECT i2000 SR (Abbott, USA), with CMIA immunochemical method.
Results: The basic criteria for ROC analysis was a diagnostic application of hematological parameters in discovering a deficiency of vitamin B_{12} or folate. The analysis showed a connection between the deficiency of vitamin B_{12} and %HS. Calculated area under the curve (AUC) for %HS was 0.656, the confidence interval (95% CI) was 0.529 to 0.769, optimal cut off value was 7% and the value of diagnostic sensitivity and specificity 77.2% and 56.8%. None of the tested parameters showed diagnostic significance in discovering the deficiency of folate.
Conclusion: With the results of this study we can conclude that the percentage of hypersegmented neutrophils is a sensitive indicator of B_{12} deficiency in relation to other hematologic parameters.

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P03-8

Monoklonski IgM s aktivnošću lupus antikoagulanta – utjecaj na koagulacijske testove

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Uvod: Prisutnost monoklonskog imunoglobulina može prouzročiti poremećaj u primarnoj ili sekundarnoj hemostazi. U ovom su radu prikazana 3 bolesnika s monoklonskim proteinom klase IgM, s patološkim vrijednostima PV-a i APTV-a, bez kliničkih znakova krvarenja.
Materijali i metode: Za sve bolesnike izrađeni su rutinski koagulacijski testovi: PV s rekombinantnim tromboplastinom (Innovin) i ljudskim placentarnim tromboplastinom (Thromborel S); APTV (Actin FS); aktivnost faktora zgrušavanja s istim reagensima za PV i APTV (Siemens Medical Solutions Diagnostics). Prisutnost lupus antikoagulanta (LA) je ispitivana prema preporukama Pododbora za lupus antikoagulant i antitijela ovisna o fosfolipidima Međunarodnog društva za trombozu i hemostazu (SSC ISTH). Antitijela prema protrombinu i beta-2-glikoproteinu I određena su enzim-imunotestom (AESCU. Diagnostics). Imunofiksacija seruma napravljena je na Sebia Hydrasys.
Rezultati: Kod sva tri bolesnika dobivene su izrazito patološke vrijednosti PV-a i APTV-a koje se nisu korigirale nakon miješanja s normalnom plazmom. Izmjerene aktivnosti faktora ukazivale su na inhibiciju gotovo svih faktora zgrušavanja s porastom aktivnosti nakon razrjeđivanja; u uzorcima sva 3 bolesnika dokazana je prisutnost LA. Ponovljenim mjerenjem PV-a s Thromborelom S dobiveni su rezultati unutar referentnog intervala. Enzim-imunotestom izmjeren je visoki titar antitijela prema protrombinu i beta-2-glikoproteinu I. Nakon slučajnog nalaza monoklonskog IgM kod bolesnika 1 (43,0 g/L), imunofiksacijom seruma kod druga dva bolesnika također je potvrđen monoklonski IgM (28,8 g/L i 6,97 g/L).
Zaključak: Kombinacija dobivenih rezultata upućuje na prisutnost monoklonskog IgM koji ima snažnu aktivnost LA i specifičnost prema protrombinu i beta-2-glikoproteinu I.

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P03-8

Monoclonal IgM paraprotein with lupus anticoagulant activity – influence on coagulation testing

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Introduction: The presence of monoclonal immunoglobulin can induce disturbances of either primary or secondary hemostasis. Hereby we report 3 cases with monoclonal IgM paraprotein and prolonged screening coagulation tests despite absence of bleeding manifestations.
Methods: Routine coagulation tests were performed for all patients: PT with recombinant thromboplastin Innovin and human placental thromboplastin Thromborel S; APTT with Actin FS; one-stage factor assays with the same PT- and APTT-reagents (Siemens Medical Solutions Diagnostics). The presence of lupus anticoagulant (LA) was tested according to the guidelines proposed by the SSC Subcommittee on Lupus Anticoagulant and Phospholipid-Dependent Antibodies of the ISTH. In addition, antibodies against prothrombin and beta-2-glycoprotein I were determined with enzyme immunoassay (AESCU. Diagnostics GmBh). Immunofixation of patients' sera was performed on Sebia Hydrasys.
Results: Prolonged PT with Innovin and APTT were obtained, without corrections after mixing studies. In factor assays, inhibition of almost all factor activities was detected, with rise in factor activities after multiple dilutions of plasma samples. Sequentially, the presence of LA was confirmed. Suprisingly, PT testing with Thromborel S resulted in normal PT-results, suggesting the possible presence of antibodies against prothrombin. This was confirmed by positive finding of antibodies against both prothrombin and beta-2-glycoprotein I. Based on accidental detection of monoclonal IgM in patient 1 (43.0 g/L), monoclonal IgM was also confirmed in other 2 patients (28.8 g/L and 6.97 g/L, respectively).
Conclusions: This combination of findings could be assigned to the presence of monoclonal IgM paraprotein with both antiprothrombin and anti-beta2-glycoprotein I specificity and strong LA activity.

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P03-9

Umbilikalna krv – važnost hematološke analize u procjeni prikladnosti uzorka za pohranu

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Uvod: Transplantacija krvotvornih matičnih stanica spada u standardne postupke liječenja malignih hematoloških i nekih drugih bolesti. U posljednjem desetljeću se u tu svrhu sve više koriste i matične stanice iz umbilikalne krvi srodnog ili nesrodnog davatelja. Cilj rada bio je ispitati točnost mjerenja broja eritroblasta u umbilikalnoj krvi na hematološkom analizatoru, te usporediti raspoložive metode za određivanje koncentracije krvotvornih matičnih stanica.
Matrijali i metode: Analizirano je 223 uzorka umbilikale krvi uzetih na standardni način pri porodu. Napravljena je usporedba rezultata broja eritroblasta s dvaju hematološka analizatora (Sysmex XE- 2100 i Abbott CELL DYN Sapphire), s obzirom da ih ubilikalna krv redovito sadrži u manjem ili većem broju. Rezultati s analizatora uspoređeni su s referentnom manuelnom mikroskopskom metodom. Također je provjerena mogućnost mjerenja krvotvornih matičnih stanica na Sysmex XE-2100 hematološkom analizatoru usporedbom sa brojem CD 34 pozitivnih stanica izmjerenih protočnom citometrijom.
Rezultati i zaključci: Točnost brojanja leukocita od presudne je važnosti za uzorke umbilikalne krvi koji se pohranjuju u banku, s obzirom da se iz ukupnog broja leukocita dobiva apsolutni broj CD34 pozitivnih stanica po kojem se procjenjuje kvaliteta uzorka za moguću transplataciju. Usporedbom s ručnim brojenjem eritroblasta, ustanovili smo veću točnost Sysmex XE 2100 analizatora (r = 0,98) naspram Cell Dyn Sapphira (r = 0,77), čije korištenje ne preporučujemo za uzorke umbilikalne krvi. Hematološki analizator Sysmex XE2100 još uvijek nema dovoljnu osjetljivost i reproducibilnost za točnu procjenu broja krvotvornih matičnih stanica u umbilikalnoj krvi. Korelacija između dvije metode (hematološki analizator i referentna protočna citometrija) je vrlo slaba (r = 0,31), te se broj matičnih stanica i dalje pouzdano može odrediti jedino protočnom citometrijom.

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P03-9

Umbilical cord blood – the importance of hematology analyzer data in estimating the appropriateness of the unit for long term storage

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Uvod: Allogenic or autologous hematopoetic stem cells transplantation is a standard therapy procedure for hematologic malignancies and some other disorders. Recently, umbilical cord blood (UCB) has been increasingly used as an alternative stem cells source. The aim of this study was to assess the accuracy of hematologic analyzers for UCB analysis, and to compare the available methods for hematopoietic progenitor cells number determination.
Materials and methods: 223 samples of UCB were analyzed by flow cytometry and two hematologic analyzers (Sysmex XE-2100 and Abbott Cell Dyne Sapphire). In this study we investigated the possibility of stem cells detection and numbering by hematology analyzer instead of flow cytometry. We also compared hematological analyzers in their ability to detect eritroblasts, which are always present in cord blood and must be detected accurately to avoid falsely elevated number of leukocytes. This is crucial for the accurate feasibility assessment of each unit stored for possible use in stem cell transplantation.
Results and conclusions: The results of our study show that for hematopoetic stem cells detection, the correlation of two methods (hematologic analyzer and flow cytometry) is very low (r = 0.31) and not significant, probably due to the low number of stem cells in UCB. Flow cytometry remains the only recommended method for hematopoetic stem cells detection in cord blood.
Leukocyte count accuracy in UCB – one of the hematologic analyzers (Abbot Cell Dyn Sapphire) performed considerably worse than the other (Sysmex XE 2100) with respect to the ability to detect erytroblasts, when compared to the reference manual method (respective correlation coefficients 0.77 vs. 0.98).

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