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**ON THE COVER** Treating Cancer. Imagine you are an unfortunate person in the general population who has been diagnosed with cancer, as was the woman to the right, the man at the bottom, or the twins at the left. Would you like to be treated with a standard protocol, or would you rather receive targeted therapy as part of an individualized treatment plan, even one that is different than your sibling? Targeted therapy provides the foundation of precision medicine, where understanding the phenotype and genotype of a person’s cancer can identify which treatments are likely to be most beneficial. Although precision medicine continues to improve, there are challenges to the implementation of precision cancer therapy. This issue of Clinical Chemistry contains a Q&A in which five experts discuss the applications of precision medicine and how targeted therapy contributes to the overall goal of precision medicine in the management of patients with cancer.

**Impact of Mean Cell Hemoglobin on Hb A1c–Defined Glycemia Status**
By Santiago Rodriguez-Segade, et al.

This research was focused on the effects of several hematological alterations, on glycated hemoglobin levels especially those influencing hemoglobin A1c as a criterion for diagnosis of diabetes. Although several contradictory effects have been published, their consequences for the diagnosis and management of dysglycemia are not certain nor is it clear which hematological indices have the most influence. The authors reviewed 21,844 outpatients in whom they found that the influence of mean cell hemoglobin and mean corpuscular volume on hemoglobin A1C largely explained the discrepancies in criteria for diagnosis and, additionally, discrepancies regarding which patients achieved glycemia management goals.

**Free and Glucuronide Whole Blood Cannabinoids’ Pharmacokinetics after Controlled Smoked, Vaporized, and Oral Cannabis Administration in Frequent and Occasional Cannabis Users: Identification of Recent Cannabis Intake**
By Matthew N Newmeyer, et al.

Extended detection of delta-9-tetrahydrocannabinol in blood is possible following chronic, frequent cannabis intake. Some minor cannabinoids may be markers of recent cannabis use; however, their blood pharmacokinetics have not been directly compared following smoked, vaporized, and oral cannabis administrations. Therefore, in this study the authors characterized the blood pharmacokinetics of 10 cannabinoids in frequent and occasional cannabis smokers following cannabis administrations via three routes. Smoked and vaporized cannabis demonstrated comparable cannabinoid delivery. Cannabigerol and cannabiol were detected following inhaled cannabis with short detection windows. The authors conclude that cannabinoid concentration ratios can limit last use estimations to within 8 hours, and that these blood pharmacokinetic data should improve interpretation of cannabinoid tests.
Stable-Isotope Dilution HPLC–Electrospray Ionization Tandem Mass Spectrometry Method for Quantifying Hydroxyurea in Dried Blood Sam
By Anu Marahatta, et al.

This article describes a novel approach for measuring hydroxyurea concentrations in whole blood from patients with sickle cell anemia being treated with the drug. Hydroxyurea was measured by tandem mass spectrometry in whole dried blood collected using either paper cards or volumetric absorptive microsampling devices. The performance characteristics of the method enabled measurements of hydroxyurea in 10 microliter samples of blood, and results were comparable for both blood-sampling methods. This method for measuring hydroxyurea from dried blood samples will make therapeutic drug monitoring and individualized pharmacokinetics possible using heel or finger-prick samples from pediatric patients with sickle cell anemia treated with hydroxyurea.

Diurnal Rhythm of Cardiac Troponin: Consequences for the Diagnosis of Acute Myocardial Infarction
By Lieke J.J. Klinkenberg, et al.

This study was aimed to challenge the critical assumption of random fluctuation of cardiac troponin in the diagnostic workup for acute myocardial infarction. By hourly blood sampling day-and-night in 24 individuals without a recent history of acute myocardial infarction, the authors demonstrated that troponin T, but not troponin I exhibited a diurnal rhythm. In a multicenter diagnostic study of 2782 patients with acute chest pain, they showed that the diagnostic accuracy of troponin T was very high and comparable among patients presenting early-morning as compared to evening. Nevertheless, the diurnal rhythm should be considered when using troponin T for screening purposes.

GDF-15 Is Associated with Cancer Incidence in Patients with Type 2 Diabetes
By Noemi Pavo, et al.

The aim of this study was to investigate whether circulating GDF-15 levels can predict the incidence of cancer in a diabetic patient cohort. From 919 patients with type 2 diabetes and no history of malignant disease, 66 patients were diagnosed with cancer during a median follow-up of 60 months. Increased concentrations of baseline GDF-15 were found to be significantly associated with cancer incidence even after multivariate adjustment. GDF-15 may serve as a biomarker to help identify individuals with type 2 diabetes at high risk for future cancer incidence, thereby needing intensified treatment for the reduction of risk factors.
Using targeted sequencing of paralogous sequences for noninvasive detection of selected fetal aneuploidies
By Christopher K Ellison, et al.

In this study the authors developed a noninvasive prenatal testing assay that leveraged paralog sequences for detection of selected fetal aneuploidies using cell-free DNA from maternal plasma. For this assay, 1060 primer pairs were used to enrich selected regions from cell-free DNA. In a blinded set of 480 samples and using the MaterniT21 PLUS assay as a reference, all 385 euploid samples, all 31 trisomy 21 samples, and 14 of 16 trisomy 18 samples, were detected with no false positive results. This study introduces a novel noninvasive prenatal testing approach using targeted sequencing of paralog motifs and establishes proof-of-concept for a low-cost, scalable method for the identification of fetal aneuploidies.

Generation of Full-Length Class I Human Leukocyte Antigen Gene Consensus Sequences for Novel Allele Characterization
By Peter M Clark, et al.

This study set out to first, establish a computational framework to identify and characterize novel HLA alleles and second, to identify trends in novel allele discovery rates and variability of genomic features (UTRs, exons and introns) within class I HLA loci. The Omixon Holotype HLA assay was utilized alongside the Omixon Twin software to generate fully phased HLA alleles from a set of 50 samples at three loci (HLA-A, HLA-B and HLA-C), generating fully phased consensus sequences for 300 alleles. The study results indicate a higher degree of intronic polymorphism than previously thought, with 8% of genotyped alleles harboring novel intronic polymorphisms and an additional 5% of alleles with an incomplete sequence annotation. These results provide a framework for full-length sequence characterization and facilitate the discovery of previously unannotated full-length HLA alleles, with potential to reveal many alleles with non-coding polymorphisms of yet unknown biological function.

Biological Variation of Hemostasis Variables in Thrombosis and Bleeding: Consequences for Performance Specifications
By Moniek PM De Maat, et al.

This study set out to determine the biological variation of coagulation variables involved in thrombosis and bleeding, to provide a recommendation for performance specifications and to assess whether hemostasis assays fulfil that recommendation. A longitudinal study was carried out by repeated blood sampling (in total 13 times over a 1 year period) in 40 healthy individuals’ hemostasis variables. The effect of biological variation on parameters of analytical variation, and proposed required performance specifications was evaluated. Biological variation was highly different for various hemostasis variables but most of the hemostasis tests variables fulfilled the current quality criteria for diagnosis and monitoring of routine hemostasis assays.
Short-term Variability of Vitamin D–Related Biomarkers
By Pamela L Lutsey, et al.

Quantifying the variability of biomarkers is important, as high within-person variability can lead to misclassification of individuals. In this manuscript the authors demonstrated that six-week short-term variability, as assessed by the within-person coefficient of variation, was quite low for vitamin D binding protein, 25-hydroxy vitamin D, calcium, albumin and phosphorus; intermediate for free and bioavailable 25-hydroxy vitamin D; but fairly high for parathyroid hormone and fibroblast growth factor 23. As such, multiple measurements of fibroblast growth factor 23 and parathyroid hormone may be needed to minimize misclassification. These results provide insight into the extent of potential misclassification of vitamin D markers in research and clinical settings.

Improved Diagnostic Performance of High-Sensitivity Cardiac Troponin Assays Is an Artifact of Censored Data
By Carel J Pretorius, et al.

Highly sensitive troponin assays are perceived to increase diagnostic sensitivity, albeit at the cost of reduced specificity. Diagnostic sensitivity however is known to be unrelated to analytical imprecision, which largely determines the detection capability or analytical sensitivity of an assay. This study modeled the effect of decreasing detection capability and data censoring on the ROC area under the curve. These results demonstrate that the improved detection capability of highly sensitive troponin assays does not impact on the diagnostic performance. Data censoring below the detection limit, however, leads to an artifactual decrease in area under the curve that may falsely create this impression.

Dried Blood Spot Reference Intervals for Steroids and Amino Acids in a Neonatal Cohort of the National Children's Study
By Dennis J Dietzen, et al.

The generation of healthy reference intervals for newborns is difficult because of the large numbers of patients needed and the limited amount of blood available from neonates. The authors of this study gained access to a large number of dried blood spots from newborns enrolled in the National Children's Study. Using LC-tandem mass spectrometry, they generated reference intervals for 25 amino acids and 4 steroid hormones applicable to children in the first 4 days of life. This approach yielded dependable reference intervals for neonates and provides a viable mechanism for further development of accurate interpretive criteria for laboratory tests performed in newborns.