ID-products in use
25(OH) vitamin D from dried blood spots as well as calprotectin and myostatin (pages 3-5)

One step ahead
In the standardization of 25(OH) vitamin D status determination (page 4)

Break-through: oxPTH analysis
Determination of OxPTH enables for the first time adequate PTH monitoring in dialysis patients (page 6)

New avenues in LC-MS/MS laboratory diagnostics
Cooperation with instrument manufacturers (page 6)

PhiCal® Calprotectin ELISA in Abbott’s FIRE-study (page 6)

New: Med-Science team in Berlin
Immundiagnostik establishes office in Berlin (page 7)

Lab-Tip: Multiple dilutions of stool samples (page 7)

NEW ON THE MARKET

MRSA-PCR Now with corresponding lysis buffer – convenient and quick analysis (page 2)

PSEUDOMONAS AERUGINOSA-PCR for the determination of intestinal bacteria (page 2)

HISTAMINE INTOLERANCE New ELISAs analyze the concentration of the neurotransmitter (page 2)

INSULIN RESISTANCE We offer a comprehensive biomarker-package for routine & research (page 3)
PhiCal® Calprotectin ELISA – no test is better!

Test result „Bravo“ in German interlaboratory trial

Immundiagnostik’s PhiCal® Calprotectin assay (K 6927) is the predominant assay in the determination of faecal calprotectin for diagnostics and therapy monitoring of inflammatory bowel diseases. The excellent reliability of the test was rated „BRAVO“ in a recent German interlaboratory trial of Instand e.V.

Precise analysis in international comparison

While other commercial assays produce incorrect data due to standardization problems, the PhiCal® Calprotectin ELISA delivers precise, clinically correct values in an English interlaboratory trial (UK NEQAS) (Whitehead et al. 2012).

Superior in the discrimination of IBD and IBS

New data from the Focus 2012 meeting confirm the predominance of our PhiCal® Calprotectin test in comparison to competing assays in the discrimination of inflammatory bowel diseases (IBD) and irritable bowel syndrome (IBS) in clinical routine (Tomkins et al., 2012). A British study assessed the significance of calprotectin in the primary diagnosis of newly admitted patients in a clinic setting and in therapy monitoring of patients with chronic inflammatory bowel diseases. The analytical precision and clinical relevance of four commercial calprotectin ELISAs were compared. The PhiCal® Calprotectin ELISA achieved an excellent correlation with intestinal inflammation and came out on top as automated test (Srinivas et al., 2012).

Stabilization buffer enables faultless calprotectin determination

A crucial factor in the measurement of faecal calprotectin is the stability of the parameter. The calprotectin concentration falls significantly in faecal samples after only one day, independent of storage conditions (e.g. room temperature, 4°C, or -20°C). An accurate determination of calprotectin is especially difficult in samples with high concentrations. The PhiCal® Calprotectin assay employs a specific buffer which keeps calprotectin stable for several weeks at room temperature, thereby enabling a precise determination and an accurate clinical diagnosis.

ELISA better than automated rapid test

The clinical accuracy of the ELISA is superior to that of an automated rapid test, which produces more false data in direct comparison (>10%, in a measurement range of 30-300 µg calprotectin/g stool), especially more false positive data.


(Overall, PhiCal performed better than EK-CA in distinguishing IBS from active IBD using 50 µg/g cut-off“.

(We will adopt the monoclonal assay from Immunodiagnostik as it is automated.
(Srinivas et al., 2012, DDW abstract)
Methicillin-resistant Staphylococcus aureus (MRSA) is responsible for many diseases that are difficult to treat. The bacterium is resistant against beta-lactam antibiotics (e.g. Penicillin) as well as against cephalosporins and is therefore a serious threat to hospitals and nursery homes.

Our MutaPLEX® & MutaREX® MRSA real time PCR kits have specifically been designed for the direct MRSA-identification.

The MRSA analysis with the PCR kits MutaREX® MRSA & MutaPLEX® MRSA is now facilitated by the corresponding lysis buffer MutaCLEAN® PLUS. The buffer enables a quick lysis of gram positive bacteria and is therefore ideal for MRSA sample preparation. An elaborate DNA extraction is not necessary anymore: The sample simply incubates in the lysis buffer for 15 min. before PCR analysis.

The PCR test enables the quantitative determination of the eubacteria group Pseudomonas aeruginosa in faecal samples for the discrimination of irritable bowel syndrome (IBS) and inflammatory bowel disease. Pseudomonas aeruginosa is indicative for IBS since it proliferates in a peak-like manner in IBS-patients in contrast to healthy subjects. Furthermore, the role of Pseudomonas aeruginosa in the development of the leaky-gut-syndrome is a matter of current research.

Many people suffer from histamine induced food intolerance, with corresponding symptoms such as migraine headache, digestive problems, irritation of the nasal mucosa or other allergy-like symptoms. The enzyme diamine oxidase (DAO) degrades histamine and is often impaired in these patients. Our new histamine ELISAs and our DAO-tests now enable a comprehensive analysis of histamine metabolism in different matrices. This tool box has been designed for routine diagnostics of histamine intolerance as well as for research on the function of histamine as a neurotransmitter.

Osteoprotegerin (OPG) is a bone protein, that is produced by a number of different tissues and cell types including osteoblasts. The glycoprotein represents a negative regulator of bone resorption by acting as a decoy receptor for RANKL, thereby neutralising its function in osteoclastogenesis.

In addition to its specific protective role in bone formation, OPG appears to be involved in the development of vascular calcification during the inflammatory events of atherosclerosis. The determination of OPG in serum is therefore useful in the research of various illnesses, such as osteoporosis, diseases with locally induced bone resorption activity, arthritis or cardiovascular pathologies. Furthermore, OPG has been identified together with other bone metabolism parameters as predictive marker for a progressive course of Morbus Bechterew. In this context, OPG is a potential candidate for therapy monitoring and optimization as well as for pharmaceutical development.

Our new, CE-certified OPG-ELISA features a high sensitivity, is suited for small sample sizes (20 µl) and can conveniently be performed in just one day.

Find more products on our website

INSULIN RESISTANCE

We offer a unique biomarker panel for diabetes-II prevention

RBP4 (K 6110)
RBP4 (1-point calibration) (K 6120)
Proinsulin intact (K 7821)
Adiponectin total (K 6250)
Leptin (KD2395)
Resistin (K 8029)
Visfatin (KA44VISFTH05)
Myostatin (K 1012)

The metabolic syndrome is characterized by adipositas, hypertension, disorders of the lipid metabolism and a resulting elevated risk for type-II diabetes, often already recognizable in a reduced glucose tolerance (pre-diabetes).

Immundiagnostik offers a comprehensive ELISA routine diagnostic panel for diabetes-II prevention: Risk patients can be tested for proinsulin as a reliable indicator of an imminent insulin resistance or for RBP-4 as a marker for undetected insulin resistance, when other clinical symptoms of diabetes-II (fasting glycaemia & HbA1C normal) are still missing.

Furthermore, we offer a broad test panel including unique tests for research on the causal events leading from cardiovascular diseases to diabetes-II.

ID-PRODUCTS IN USE

DRIED BLOOD SPOT VITAMIN D ANALYSIS

25(OH) Vitamin D analysis from dried blood spots now clinically validated

To facilitate collection and shipping of blood samples for vitamin D status analysis, Immundiagnostik has developed the D-Vital ID® test set for dried blood spots from capillary blood. This material can be mailed to a laboratory for analysis via conventional couriers without cooling. Vitamin D determination of the D-Vital ID® samples is exclusively coupled to Immundiagnostik’s 25-OH vitamin D ELISA. This market-proven assay delivers reliable vitamin D data which are comparable in their diagnostic accuracy to reference methods such as HPLC or LC-MS/MS.

The D-Vital ID® test set supplements Immundiagnostik’s large product range in vitamin D analysis with a product, that significantly simplifies the preparation and shipping of blood samples, thereby enabling vitamin D status monitoring on a larger scale with little effort.

Immundiagnostik’s dried blood spot vitamin D analysis from capillary blood using the D-Vital ID® test set has recently been tested at the University of Potsdam and the University of Mainz in a clinical setting:

Samples from capillary and venous blood were acquired in parallel from 96 subjects. The concentration of 25(OH) vitamin D was subsequently determined with Immundiagnostik’s ELISA using the dried blood spot and venous blood samples. The data comparison showed a very good agreement of both methods ($r^2 = 0.63$). The results of this study confirm that the vitamin D analysis from capillary blood with the D-Vital ID® test set is comparable to the classical determination from venous blood. The novel test set is therefore an ideal alternative to conventional vitamin D screening.


Test set D-Vital ID® for dried blood spots


D-Vital ID® (DZ9002)
CONGESTIVE HEART FAILURE

Myostatin ELISA validated for clinical research on congestive heart failure

Myostatin is a negative regulator of muscle growth and presumably plays a regulatory role in the cardiovascular system. The first ELISA for the determination of human myostatin, developed by Immundiagnostik, has now been used by the University of Cologne together with the University hospital of Mainz in the clinical monitoring of patients with decompensated congestive heart failure (CHF). The parameter was first significantly elevated in the patient group and dropped in the course of an ongoing therapy with standard CHF-medication. The authors observed a myostatin-lowering effect during CHF-therapy in parallel to the expected drop in NT-proBNP concentration (s. Fig. 3A).

The data confirm that Immundiagnostik’s Myostatin ELISA reliably reflects the clinical status and therapeutical effect in CHF-patients. The validated ELISA is therefore ready for a broad range of applications in cardiovascular research, e.g. for research on cardial cachexia.

LEADING THE WAY

in the standardization of 25(OH) vitamin D status analysis

The determination of 25(OH) vitamin D is a routine analysis in laboratory diagnostics. However, there is an urgent need to standardize the various methods that are currently used in order to establish a universal reference method for correct and clinically valid data interpretation.

For example, the processing of higher sample numbers by fully automated immunoassays is error-prone due to the incomplete separation of the vitamin D binding protein (VDBP) (Heijboer et al. 2012); In samples with high VDBP concentrations (e.g. from pregnant women), this incomplete purification of vitamin D leads to false-low results and in samples with low VDBP concentrations (e.g. from intensive care patients) to false-elevated results.

In direct comparison to the 25(OH) vitamin D Immundiagnostik-ELISA the other commercially available automated immunoassays deliver variable data, the results are to a large extent dependent on the used method (Farrell et al.; Carter; Moon et al.; all 2012).

To be on the safe side rather use the gold standard: Immundiagnostik’s 25(OH) vitamin D ELISA features an excellent correlation to chromatographic methods (r= 0.92 with LC-MS/MS) and meanwhile has been established as benchmark reference assay in the standardization of 25(OH) vitamin D analysis (Scharla & Lempert, 2012).

The test is automatable to process larger sample volumes and the Xpress-version requires only 4 hours.

25(OH) Vitamin D Xpress (K 2107)

Simply different versions
Simply the best!
Simply the best!
Simply monoclonal
Simply automatble
Simply different versions for your needs
*S. Scharla & U. Lempert; P60, Osteologie 2012
ACUTE KIDNEY INJURY

**PhiCal® Calprotectin ELISA identifies intrinsic acute kidney damage**

The prognosis of patients suffering from acute kidney injury depends to a large extent on an early diagnosis and the immediate start of an appropriate therapy. The quicker the underlying cause of kidney malfunction has been identified the better are the chances for a successful treatment.

Especially the differentiation of prerenal and intrinsic acute kidney injury is significant since both diseases require fundamentally different therapies: Prerenal damage represents a problem that is located at the front-end of the kidney, e.g. massive fluid loss and/or hypotension. Since the nephrons itself are intact, a simple volume adjustment can already improve the patient’s condition. An intrinsic kidney injury involves damage of the organ itself, caused for example by acute tubular necrosis, interstitial nephritis, glomerulonephritis or vasculitis – a complex, costly and intensive therapy is required.

In their search for a reliable routine parameter for the discrimination of prerenal and intrinsic kidney injury, clinical researchers of the Charité in Berlin have determined the inflammation marker calprotectin in the urine of patients with acute kidney injury:

Subjects with intrinsic kidney injury had a significantly higher calprotectin concentration than patients with prerenal damage (s. Fig. 1A). The authors conclude from these data that calprotectin in urine is an adequate diagnostic marker for the differentiation of prerenal (calprotectin low) and intrinsic (calprotectin high) acute kidney injury.

In addition, with a sensitivity of 92.3% and a specificity of 97.1% the **PhiCal® Calprotectin ELISA** has been validated as a reliable test for clinical routine.

**PROJEKT & COOPERATIONS**

**OXIDATIVE STRESS MARKER IN DIABETES PATIENTS**

Joint project with Boehringer Ingelheim and Bayer Healthcare

Together with Boehringer Ingelheim Pharma GmbH & Co. KG and Bayer Healthcare AG Immundiagnostik investigates the role of oxidative stress in the development and progression of chronic kidney diseases (e.g. diabetic nephropathy). The project focuses on the effect of novel pharmaceuticals, such as DPP-4 inhibitors as well as on activators of the guanylate cyclase. The intention is to follow up on initial data from animal experiments in phase II/III clinical studies.

Berthold Hocher and colleagues presented first results of this cooperation on this year’s ADA (American Diabetes Association) congress in Philadelphia/USA.

**RELAXIN-2 AS VASCULAR MODULATOR**

Cooperation with Charité Berlin and the University of Potsdam

Relaxin-2 influences the cardiovascular system via G-protein-coupled receptors and exhibits a compensatory function in patients with heart failure. Thomas Dschietzig and colleagues have employed Immundiagnostik’s relaxin assay in animal experiments and demonstrated a protective role of relaxin in TNF-α-induced endothelial dysfunction. (Dschietzig et al., Cardiovascular research 04/2012; DOI: 10.1093/cvr/cvs149)

**LABORATORY DIAGNOSTICS OF OXIDATIVE STRESS**

Advancement of Immundiagnostik’s test panel

Oxidative stress is involved in the pathogenesis and progression of a variety of diseases, e.g. atherosclerosis, diabetes, neurodegenerative disorders, rheumatoid arthritis and even cancer. The University of Graz identifies and validates in cooperation with Immundiagnostik analytic methods and diagnostic parameters of oxidative stress, such as carboxylated proteins, oxidized LDL or nitrotyrosin. The goal of these research projects is to enable and optimize therapeutic strategies to fight oxidative stress. Our Med-Science team will feature a seminar „Diagnostics of oxidative stress“ in 2013.

More information:
marion.kronabel@immundiagnostik.com

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Break-Through for Kidney Therapy Control

Determination of OxPTH enables for the first time adequate PTH monitoring

Secondary hyperparathyroidism is a frequent result of progressive kidney malfunction in patients suffering from chronic renal failure. The level of parathyroid hormone (PTH) rises in the course of the disease and can lead to bone loss, fractures, and vascular calcification with an increased risk of cardiovascular mortality. Monitoring of the PTH level to identify and treat secondary hyperparathyroidism in time is therefore an essential part of therapy control in patients suffering from chronic renal failure.

However, a precise PTH analysis in clinical routine is difficult: Some assays determine the total PTH concentration, including degradation products, which leads to false-elevated data. Other tests avoid this problem but still lack clinical accuracy in the diagnosis of hyperparathyroidism. Immundiagnostik and the Institute for Nutritional Sciences of the University Potsdam have jointly developed a completely novel PTH test, that can exactly determine the concentration of the biologically active hormone:

Given that an elevated oxidative stress burden in patients with chronic renal failure leads to PTH oxidation and a resulting loss of PTH receptor binding, the immunoassay is based on the discrimination of oxidized PTH (oxPTH, biologically inactive) and non-oxidized PTH (biologically active). An initial sample preparation step employs an affinity chromatography to remove all oxPTH from the patient plasma. Subsequently, non-oxidized, biologically active PTH is detected in this pre-treated sample with a conventional Sandwich-ELISA.

Using this novel assay, researchers could demonstrate that in dialysis patients a considerable, yet individually variable part of the PTH is oxidized and therefore biologically inactive. The test will now be used in clinical studies on the correlation of oxPTH and bone disorders as well as cardiovascular diseases in dialysis patients. First results of the cooperation have been published (s. below) and will be presented at the ISN Nexus 2012 conference in Copenhagen (20.-23.09.).

(Calprotectin ELISA

In Service: PhiCal® Calprotectin ELISA in Abbott’s FIRE-Study

The determination of calprotectin in stool is an established diagnostic marker for chronic intestinal bowel diseases. The significance of the calprotectin concentration in predicting relapse in Morbus Crohn patients is currently under investigation in the multicentric FIRE-study by Abbott (FIRE = Faecal Marker of Intestinal Inflammation for RElapse prediction in routine monitoring of Morbus Crohn patients). The goal of the FIRE-study is, among other aspects, to identify appropriate basal levels of calprotectin during remission as well as to determine threshold concentrations for the prediction of a relapse risk. The time span between a relevant rise in calprotectin level and the onset of clinical relapse is of particular significance for therapeutic intervention.

Using the PhiCal® Calprotectin ELISA the faecal calprotectin concentration will be assessed in approximately 500 Morbus Crohn patients in 50 participating German clinical institutions over a period of two years. The collected data will provide information on the predictive potential of calprotectin with respect to the affection pattern, the duration and the course of the Crohn’s disease, thereby establishing the value of the parameter for a personalized therapy.

New Avenues in LC-MS/MS-Diagnostics

Cooperation with instrument manufacturers to advance product portfolio

For the further development of the chromatography product pipeline Immundiagnostik counts on a close cooperation with leading manufacturers of diagnostic instruments. The new biomarker LC-MS/MS product line will focus on indications in the areas of cardiovascular diseases and bone metabolism.

One successful example is the development of the innovative and highly precise 1,25(OH)₂ Vitamin D ImmunoTube® LC-MS/MS kit. Immundiagnostik validated the test protocol and proprietary ImmunoTube® affinity purification sample preparation step in close collaboration with ThermoScientific, AB Sciex and Waters, utilizing their powerful instruments.

This fruitful industrial cooperation will be continued in a joint development of mass spectrometry tests for the qualitative and quantitative analysis of diagnostic marker proteins for use in routine and research.

Immundiagnostik organizes already today individual demos together with leading manufacturers of LC-MS/MS instruments, e.g. for the determination of 1,25(OH)₂ vitamin D.
COMPANY NEWS

NEW: „MED-SCIENCE“ TEAM IN BERLIN

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As of now, Immundiagnostik operates a Med-Science office in Berlin that is in charge of customer liaison and support as well as of training and continued education in the areas of routine diagnostics and research. The bureau will advance the publication of novel research with established markers and the introduction of innovative parameters in clinical routine. Other tasks of the team include scientific marketing, the identification of new business areas, networking, and the coordination of research cooperations.

The team:
Marion Kronabel has previously been director of European affairs at BIO Deutschland, director of the European working group for pharma biotechnology (EAPB) and more than 10 years deputy director of Heidelberg’s technology park. Thomas Dschietzig, Professor at the Charité Berlin, is a specialist for cardiovascular diseases, esp. heart failure. Berthold Hocher is Professor at the University of Potsdam. His research focuses on nephrological diseases and their impact on the cardiovascular system (www.uni-potsdam.de/eem/index/prof-hocher.html).

EVENTS

- Biomedical Laboratory Science
  18. – 22. August, Berlin

- INDC (12th International Nutrition & Diagnostics Conference)
  27. – 30. August, Prague, Czech Republic

- ADMA

- ASBMR (American Society of Bone & Mineral Research)
  12. – 15. October, Minneapolis, USA
  Booth #425

- UEGW (United European Gastroenterology Week)
  20. – 24. October, Amsterdam,
  The Netherlands
  Booth #35

LAB-TIP

Multiple determinations of serial stool sample dilutions deliver correct data

For a precise determination of calprotectin concentrations it is helpful to dilute stool samples several times, e.g. 1:2500, 1:5000 and 1:10000. These multiple determinations of different dilutions validate the single data points, which is especially useful in samples with high calprotectin concentrations. This procedure is recommended in particular for those labs that participate in interlaboratory trials. Sample dilution with Immundiagnostik’s stabilization buffer included in the PhiCal® Calprotectin ELISA kit guarantees for genuine data and correct clinical results.

CONTACT

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