Sudan University of Science and Technology

College of Graduate Studies

"Diagnostic Value of Serum Procalcitonin and Highly Sensitive C- reactive protein in early detection of bacterial blood Infection"

القيمه التشخيصيه لفحص البروكالسيتونين وبروتين سي عالي التفاعل كمؤشر في الكشف الاولي لألتهاب الدم البكتيري

A Dissertation Submitted in Partial fulfillment of Requirements of the Degree of M.Sc. .in Medical Laboratory Science (Clinical Chemistry)

Prepared by:

Nashwa Mirghani Ahmed Almaki

-B.Sc. in Medical Laboratory Sciences-Clinical Chemistry -Sudan University of Science and Technology) 2005)

-Higher diploma in Medical Laboratory Science-Alzaiem Alazhari University (2007)

Supervised by:

Dr. Seif Elden Ahmed Mohamed

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الصفحه المعدله من الجامعه

April 2021

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بِسْمِ ٱللَّهِ ٱلرَّحْمَٰنِ ٱلرَّحِيمِ

(وَيَزِيدُ ٱللَّهُ ٱلَّذِينَ ٱهْتَدَوْا۟ هُدًى ۗ وَٱلْبَٰقِيَٰتُ ٱلصَّٰلِحَٰتُ خَيْرٌ عِندَ رَبِّكَ ثَوَابًا وَخَيْرٌ مَّرَدًّا)

(سورة مريم الاية 76)

Dedication

I dedicate this work to my loving supportive family

May I always have you safe and close

Acknowledgments

I want to express gratitude to my supervisor for his guidance and continuous .support through this work

Thank you

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LIST OF ABBREVIATIONS:

1. (ΔPCT): Serum PCT kinetics
2. (AKI): Acute kidney injury
3. (AUC): The area under the curve.
4. (BCs): Blood Cultures
5. (CKD): Chronic kidney disease
6. (CRP): C-reactive protein
7. (FUO): Fever of unknown origin
8. (ICU): Intensive care unit
9. (IL-8): Post-operative Interleukin
10. (PCT): Procalcitonin
11. (PE): Preeclampsia
12. (POD): Post-operative day
13. (SIRS): Systemic inflammatory response syndrome
14. (VAP): Ventilator-associated pneumonia
15. (UTI): Urine Tract infection
16. (WBCs): White blood cells
17. (IMC): International Medical Center

ABSTRACT

Background and aim: Several markers of inflammation and sepsis such procalcitonin (PCT)and hs-C.reactive protein (hs-CRP) are being examined to study their accuracy for the diagnosis of bacterial infections in order to treat properly and effectively.

This study aims to explain the diagnostic value of serum Procalcitonin test in comparison with hs-C.reactive protein as diagnostic tools for bacterial infections in blood .

Method: Analytical cross-sectional hospital-based study include123participants from International Medical Center hospital (IMC) A large hospital in Jeddah, Saudi Arabia hospital from Jan ,1st 2018 to Dec 31st 2018.whom they entered the hospital with fever, high rate of breathing. hs-CRP and PCT in addition to blood culture had been requested. hs-CRP and PCT were measured immediately by automated Cobas machine, the results obtained was analyzed using SPSS version 23.

Results: The results showed that (78.9%) of patients were above the age of 60, males are slightly higher than females. PCT and hs-CRP results have been compared with the Blood Culture's results, sensitivity of PCT & hs-CRP was 100% while specificity was 98 % & 88% respectively.

Receiver Operating Curve (ROC) for PCT; AUC= 0.984 with CI (0.954-1.015), P.value 0.00, while hs-CRP; AUC=0.646, with CI (0.547-745), P- value 0.006, which consider more significant on PCT than CRP

Conclusion: Procalcitonin is the best marker for diagnosis of blood bacterial

infection than hs-CRP.

المستخلص

الخلفية و الهدف: يتم فحص العديد من علامات الالتهاب والإنتان مثل علامات البروكالسيتونين (PCT) والبروتين المتفاعل (CRP) لدراسة دقتها التشخيصية لالتهابات البكتيرية من أجل علاجها بشكل صحيح وفعال.

هذه الدراسة تهدف لتوضيح القيمة التشخيصية للبروكالسيتونين بالمقارنة مع بروتين سي التفاعلي كأدوات تشخيصية محددة بين المرضى الذين تم تشخيصهم بالالتهاب البكتيري.

الطريقة: دراسة وصفية مقطعية أجريت على 123 مشترك في المركز الطبي الدولي ، مستشفى كبير في جدة

المملكة العربية السعودية من 1 يناير 2018 الى 31 ديسمبر 2018 ، اللذين تم ادخالهم للمستشفى بحمى ، تنفس عالي بروتين سي التفاعلي عالي الحساسية و البروكالسيتونين بالأضافة لمزارع الدم.

تم فحص بروتين سي التفاعلي و البروكالسيتونين باستخدام جهاز Cobas الاتوماتيكي و تحليل النتيجة بأستخدام البرنامج الاحصائي SPSS النسخة رقم 23.

النتائج: أظهرت النتائج أن معظم المرضى فوق سن الستين عاما (78.9%) من إجمالي المرضى الذين شاركوا في هذه الدراسة ، الوسط الحسابي للعمر60 عاما . نسبة الذكور والاناث متماثلة تقريبا مع غلبة طفيفة للذكور.

تمت مقارنة نتائج البروكالسيتونين و ال hs-CRP مع نتائج المزارع البكتيرية لتاتي نتائج الحساسية لهما معا 100% بينما الدقة للبروكالسيتونين 98% و للبروتين التحسسي عالي الحساسية 88.%

و حيث تاتي قيمة P value بقيمة 0.00 للبروكالسيتونين و 0.006للبروتين عالي الحساسية بيحث يتضح ان PCT افضل من CRP عالي الحساسية في التشخيص.

الخلاصة: البروكالسيتونين من افضل البروتينات المستخدمة كعلامات تشخيصية لالتهاب الدم بالبكتيريا

Chapter One

1.1. Introduction:

Inflammation is a basic way in which the body reacts to infection, irritation or other injury, the key feature being redness, warmth, swelling and pain (Stankov.,2012).

Bacterial infection is a major cause of morbidity and mortality (Martin *et al*. 2003 & Angus *et al*. 2001). Diagnosis of bacterial infections can be difficult because of the variety of its clinical presentations; for example, it may be problematic to differentiate viral from bacterial infections.(Tenover el at.,1997)

According to the WHO`s report on infectious disease: overcoming antimicrobial resistance; Treating viral infections or non-infective causes of inflammation with antibiotics is not only ineffective, but also contributes to the development of resistance, increased cost of treatment and adds the risks of toxicity and allergic reactions, thus negatively affecting both patient, health care system and community(WHO.,2000).

Sepsis is a systemic, toxic host response to infection leading to severe sepsis (acute organ dysfunction secondary to suspected infection) and septic shock.(Angus el at.,2013).

Severe sepsis and septic shock are major healthcare problems, affecting millions of people around the world each year, killing one in four (and often more), and increasing in incidence (Angus *et al.*2001, Dellinger 2003, Martin *et* al. 2000 & Dombrovskiy *et al*.,2007) its mortality rate ranges from 21% to 48%. The most significant feature in the prognosis of patients is bloodstream infection, so rapid and early diagnosis of bloodstream infection is important for proper treatment (Koch *et al*.,2014).

C-Reactive protein **(**CRP**)**is synthesized by the liver mainly in response to IL-6 and also in many types of inflammation. CRP level measurements were used to aid in the diagnosis of bacterial infections. (Ridker .,2003**)**

C-reactive protein (CRP) is an ancient highly conserved molecule and a member of the pentraxin family of proteins. CRP is secreted by the liver in response to a variety of inflammatory cytokines. Levels of CRP increase very rapidly in response to trauma, inflammation, and infection and decrease just as rapidly with the resolution of the condition. Thus, the measurement of CRP is widely used to monitor various inflammatory states. CRP binds to damaged tissue, to nuclear antigens and to certain pathogenic organisms in a calcium-dependent manner. The function of CRP is felt to be related to its role in the innate immune system. Similar to immunoglobulin (Ig) G, it activates complement, binds to Fc receptors and acts as an opsonin for various pathogens. Interaction of CRP with Fc receptors leads to the generation of pro-inflammatory cytokines that enhance the inflammatory response. Unlike IgG, which specifically recognizes distinct antigenic epitopes, CRP recognizes altered self and foreign molecules based on pattern recognition. Thus, CRP is thought to act as a surveillance molecule for altered self and certain pathogens. This recognition provides early defense and leads to a pro-inflammatory signal and activation of the humoral, adaptive immune system (Du clos TW.,2000).

Procalcitonin (PCT) is produced by thyroid C cells and converted to calcitonin before being released into the bloodstream.

Circulating levels of PCT which are produced by liver Monocytes, macrophages, and lung and intestinal lymphocytes are generally very low in healthy individuals, but can increase by 100 to 1000 fold in response to systemic bacterial infections (Whicher, Bienvenu&Monneret.,2001).

Procalcitonin (PCT), a protein of 116 amino –acid with molecular weight of 13

KDa, was discovered 25 years ago as prohormone of calcitonin produced by C-cell of the thyroid gland and intra-cellularly cleaved by proteolytic enzyme into the active hormone. Circulating levels of PCT in healthy subject are below Detection limit. PCT become important in the detection and differential diagnostics of inflammatory states. The production of PCT during inflammation is linked with bacterial endotoxin and with inflammatory cytokines (TNF, IL-6). PCT detectable in the plasma during inflammation is not produced in C-cells of the thyroid. The probable sites are neuroendocrine cells in the lungs or intestine (Marune P et al.,2000).

PCT is used as a biomarker or initiating or terminating antibiotic therapy in various clinical settings, including the emergency department, intensive care unit (ICU), and primary care (Schuetz et al.,2010)

The mechanism for PCT production after inflammation and its role are not known completely. It is believed that PCT is produced by the liver (Bouadma *et al.,*2010) and peripheral blood mononuclear cells (Briel *et al*.,2008), modulated by lipopolysaccharides and sepsis-related cytokines.

Procalcitonin (PCT) and C-reactive protein (CRP) markers are being examined in this research to study their accuracy for the diagnosis of bacterial infections.

1.2. RATIONALE:

Blood cultures are considered to be the gold standard test for diagnosing blood born infections, despite the possible time delay (hours to days) until positive results can be confirmed. Therefore, great interest in biomarkers that could be used in either the emergency department or critical care to help effectively diagnosis of sepsis. Liu and co-workers [2016], research in biomarkers for diagnosis of sepsis concluded that Procalcitonin (PCT) and interleukin-6 (IL-6) had some degree of diagnostic accuracy.

Procalcitonin (PCT) is one of the recent inflammatory markers that been used in laboratory medicine as aid for diagnosis sepsis ,this study aims to determine the better use of PCT and CRP which is higher fast, and more specific and sensitive. To our knowledge there were few studies has been conducted with regard of PCT correlation with sepsis in the Middle East countries such as Sudan and Saudi Arabia.

1.3. OBJECTIVES:

1.3.1. General objective:

To Diagnose the value of serum Procalcitonin & high sensitive C- reactive protein for detecting bacterial blood Infection.

1.3. 2. Specific objectives:

- To compare Serum PCT and hs-CRP level in patients with bacterial infection and those without bacterial infection.(Study group)
- To calculate the sensitivity and specificity of the Serum PCT and hs-CRP in relation to bacterial blood culture.
- To evaluate the association of Serum PCT and hs-CRP with type of the Bacteria.
- To determine the usefulness of serum PCT on speed of diagnosis of the Bacterial infection.

Chapter Two

2. LITERATURE REVIEW

2.1. Sepsis

Sepsis is a systemic immune response to infection by microbial organisms. Sepsis is defined as the presence (probable or documented) of infection together with systemic manifestations of infection. The most common primary source of infection resulting in sepsis is the lungs, accounting for about half of all cases, followed by the abdomen, and the urinary tract. No definitive source is found in one third of cases [Munford Robert S, *et al* 2014]. Sepsis encompasses a spectrum of illness that ranges from minor signs and symptoms through to organ dysfunction (severe sepsis) and shock. Increasing severity correlates with increasing mortality, which rises from 25-30% for severe sepsis up to 40-70% for septic shock.(Lever A, *et al*.,2007).

Sepsis and its complications have a significant and increasing impact on health sector, and are one of the leading causes of mortality. The incidence of sepsis is increasing in all areas of the world. (Martin *et al*., 2012)

Timely diagnosis and treatment are highly important in reducing the morbidity and mortality associated with sepsis. At-times the diagnostic uncertainty still remains high despite the available clinical information. Thus, a laboratory test with more specificity is essential. Serum biomarkers like procalcitonin may aid in the early diagnosis of sepsis and therapeutic intervention. (Shiferaw, et al.,2016)

2.1.1 Risk factors for sepsis:

Everyone is susceptible to sepsis, but the risk is higher in weakened immune systems, Infants and children, the elderly, people with chronic illnesses like diabetes, AIDS, cancer, kidney , liver disease also People suffering from a severe burn or physical trauma.

Common symptoms of sepsis are fever, chills, rapid breathing and heart rate,

rash, confusion, and disorientation. The systemic inflammatory response (SIRS) to infection, manifested by at least two of ;Temperature of >38 °C or <36°C ,Heart rate of >90 beats per minute , Respiratory rate of >20 breaths per minute or partial pressure of CO_2 of <32 mmHg or White blood cell count of >12,000 per ml or <4,000 per ml, or >10% immature (band) forms (Muckart & Bhagwanjee.,1997)

2.2. Laboratory test helpful in diagnosis of sepsis:

The following were the proposed laboratory tests for the diagnosis of sepsis.

2.2.1. White blood cells (WBCs): to screen for or diagnose a variety of conditions that can affect an infection, inflammation or a disease that affects the production or survival of WBCs; to monitor treatment of a blood disorder or to monitor therapy that is known to affect WBCs. (Magrini.,2014)

2.2.2. The blood differential test*:* is used to diagnose a variety of medical conditions. These may include infections, autoimmune diseases, anemia, inflammatory diseases, and leukemia and other types of cancer. It is a common test that is frequently used as part of a general physical exam.

A blood differential test measures the amount of each type of white blood cell (WBC) that you have in your body. White blood cells (leukocytes) are part of your immune system, a network of cells, tissues, and organs that work together to protect you from infection. There are five different types of white blood cells:

Neutrophils are the most common type of white blood cell .These cells travel to the site of an infection and release substances called enzymes to fight off invading viruses or bacteria.(Ballard & Harold S.,1997).

Lymphocytes There are two main types of lymphocytes; B cells and T cells. B cells fight off *invading* viruses, bacteria, or toxins. T cells target and destroy the body's *own* cells that have been infected by viruses or cancer cells. (Spiering &Martin J., 2015).

Monocytes remove foreign material, remove dead cells, and boost the body's immune response.(Kovacs *et al*.,2002)

Eosinophils fight infection, inflammation, and allergic reactions. They also defend the body against parasites and bacteria.(Acharya et al., 2014).

Basophils release enzymes to help control allergic reactions and asthma attacks. However, your test results may have more than five numbers. For example, the lab may list the results as counts as well as percentages (Peters *et al*.,2004).

2.2. 3. Blood Cultures (BCs): are considered the gold standard for detecting pathogens in patients with sepsis; however, given the time required, it cannot be applied to make early therapeutic decisions (Bone *et al*.1992).

2.2.4. C-Reactive protein*:* Is a non-glycosylated circulating pentraxin composed of 5 identical subunits arranged with pentameric symmetry.

Also it is an acute-phase reactant, and CRP level measurements are frequently used to aid in the diagnosis of bacterial infections. It is synthesized by the liver, mainly in response to IL-6, which is produced not only during infection but also in many types of inflammation (Ridker.,2003). It binds to polysaccharides in pathogens, activating the classical complement pathway.

2.2.5. Procalcitonin: Procalcitonin (PCT) is a 116-amino acid protein with a molecular mass of 13 kDa that is produced by thyroid C cells and converted to calcitonin before being released into the bloodstream.

Circulating levels of PCT—which are produced by liver Monocytes, macrophages, and lung and intestinal lymphocytes—are generally very low in healthy individuals, but can increase by 100 to 1000-fold in response to systemic bacterial infections (Whicher, Bienvenu&Monneret.,2001).

PCT is used as a biomarker for initiating or terminating antibiotic therapy in various clinical settings, including the emergency department, intensive care

unit (ICU), and primary care (Schuetz

As mentioned above among several markers of inflammation and sepsis, procalcitonin (PCT) and C-reactive protein (CRP) markers are being examined to study their accuracy for the diagnosis of bacterial infections.

PCT is the prehormone of calcitonin, which is normally secreted by the C cells of the thyroid in response to hypercalcemia; under these normal conditions, negligible serum PCT concentrations are detected (Schuetz *et al*.,2009).

The mechanism for PCT production after inflammation and its role are not known completely. It is believed that PCT is produced by the liver (Bouadma*et al.,*2010) and peripheral blood mononuclear cells, modulated by lipopolysaccharides and sepsis-related cytokines. (Briel*et al*., 2008).

2.3 Previous studies:

The literature regarding studies if efficiency and comparison between CRP and PCT are numerous, this review includes recent, relevant and variant studies, covering many methodologies, study participants with different risk factors and demographics.

A prospective cross-sectional study, among neonates in Neonate Intensive Care Unit (NICU) at King Abdul-Aziz Medical City, Riyadh (Saudi Arabia). Number of participants included healthy control neonates (n=80), clinical sepsis group (n=80) with clinical signs of sepsis but their blood culture was negative and sepsis group with clinical signs of sepsis and their blood culture was positive. CRP sensitivity was of 78 % and specificity of 70 %. On the other hand, CRP specificity was (70%). Results showed that similar finding was found using CRP and PCT combination (Fattah MA.,2017).

A cross sectional study conducted in Sudan in the efficacy of procalcitonin in early diagnosis of neonatal sepsis. The results showed that CRP have 93.5%

sensitivity and50% specificity ,on the other hand PCT showed better sensitivity and specificity 96.8%,85.7 respectfully (El-Amin et al.,2017).

A meta-analysis was performed to evaluate the accuracy of determination of procalcitonin (PCT) and C-reactive protein (CRP) levels for the diagnosis of bacterial infection. PCT level was more sensitive (88% vs. 75%) and more specific (81% vs. 67%). The sensitivity for differentiating bacterial from viral infections was also higher for PCT markers (92%vs.86%); the specificities were comparable (73% vs. 70%) (Simon *et al*.,2004).

A systematic review of 17 articles stated that the concentrations of both PCT and CRP were higher in the serious bacterial infections (SBI) group than in the non- bacterial infection group. Sensitivity for differentiating bacterial infections from nonbacterial infections was higher for PCT compared with CRP (Hu *et al*.,2017). Also a comprehensive systemic search including 18 studies, comprising 3470 patients aimed to assess the accuracy of presepsin for the diagnosis of sepsis in adult patients and compared the performance between presepsin, C-reactive protein (CRP), and procalcitonin (PCT). Found no significant difference between presepsin and PCT .However, for studies conducted in ICU, the pooled sensitivity of presepsin was found to be higher than PCT while the pooled specificity of presepsin was lower than PCT((Wu *et al*.,2017).

Serum samples were collected from febrile patients and processed for blood cultures. PCT and CRP. The patients were divided into three groups according to the final diagnosis: bacteraemia group (group1), bacterial infection with negative blood culture (group 2) and non-bacterial infection group (group 3).There were significant ($P<0.05$) difference in the levels of PCT and CRP among the three groups. The PCT levels of patients with Gram-positive bacterial infections were lower than Gram-negative bacterial

infections (0.53 vs. 2.13, P < 0.01). The best cut-off value to detect bacterial infections was 0.26 ng/ml for PCT (Qu *et al*.,2015).

A retrospective study with 59 patients was divided into sepsis and septic shock groups, as well as survivor and non-survivor groups, according to the severity of the disease and patient survival. On the 2nd, 3rd, and 5th days, the CRP level was higher in the non-survivor group than in the survivor group, and the serum CRP level was higher in patients in the septic shock group than in patients in the sepsis group. PCT levels were significantly different between non-survivor and survivor groups, whereas they did not differ between patients in the sepsis and septic shock groups. Serum PCT kinetics (ΔPCT) was similar between groups (Cui *et al*.,2019).

Many studies in children were also carried out, for instance a study where plasma was obtained from pediatric oncology patients presenting with febrile neutropenia; median PCT levels were significantly increased in patients with a bacterial infection, in contrast to CRP. After 24–48 hr, only PCT was significantly elevated during bacterial infection (Miedema *et al*. 2011). Another one with children undergoing intensive chemotherapy, found a highly significant difference in PCT levels between bacterial and nonbacterial episodes. Sensitivity and specificity of PCT were 94 and 96.5%, respectively (Hatzistilianou *et al*.,2010).

A prospective study with 49 patients, who had 60 febrile episodes and were hospitalized. All patients had been diagnosed with neutropenic fever after intensive chemotherapy. The PCT and CRP levels were significantly higher in neutropenic- fever patients (group I and group II separately) than in control patients (P<0.001) throughout the study period, the median of PCT concentrations shows a tendency to fall after the 8th hour of onset of fever, whereas in patients with documented infections PCT concentrations fell after

the 48th hour. the PCT was more helpful when measured periodically in diagnosing inflammation in pediatric neutropenic- fever patients than CRP(Secmeer *et al*.,2007).Also it observed that serum levels of PCT and CRP were higher in the sepsis group than non-sepsis group ($p<0.001$). The sensitivities of both PCT and CRP at the recommended cut-off level for all infants were 68.2% and 38.6% respectively, the sensitivities of both biomarkers for the diagnosis of neonatal sepsis were increased to 95.5%. The birth weight and gestational age had no effects on the diagnostic value of these serum biomarkers (Yang *et al*.,2016). Paper observed that at baseline and during the febrile episodes, the highest levels of all parameters were observed in cases of gram-negative bacteraemia. However, in fever of unknown origin (FUO) and localized infections, low or only slightly elevated median levels of all parameters were documented. In comparison with the other inflammatory parameters, PCT (optimum cut-offlevel0·5 μg/l) was a more sensitive and more specific parameter in the diagnosis of high-risk (gram-negative bacteraemia) and low-risk (FUO) episodes (Fleischhack *et al*.,2000).

Some studies among pregnant ladies were also carried out. A research with 50 normal pregnant women and 59 pregnant women with preeclampsia (PE), Mean of CRP and PCT was higher in pregnant women with PE than normal pregnant women. The optimal cut-off point for CRP was 5.24 with a sensitivity of 62.7% and a specificity of 56%. The optimal cut-off point for PCT was 0.042 with a sensitivity of 71% and a specificity of 54% (Jannesari& Kazemi.,2017).

A retrospective analysis to evaluate the diagnostic value of procalcitonin (PCT) and C-reactive protein (CRP) for the prediction of subclinical intrauterine infection in patients with premature rupture of membranes

(PROM). The study included 276 cases. The area under the ROC curve of CRP is 0.632, and it is significantly better than PCT. The sensitivity, specificity, positive predictive value, and negative predictive value of CRP are 0.686, 0.958, 0.501, and 0.765, which are all better than those of PCT. The group analysis according to different gestational weeks demonstrated that for the group of 28–33+6 weeks pregnant women, the area under the ROC curve of CRP and PCT is 0.869, and 0.787. The both test have satisfactory accuracy. But the sensitivity and specificity of PCT is 0.830, and 0.950, respectively. That is significantly better than CRP (Li *et al.,*2016).

A retrospective study of 293 patients who presented to the ED in which a urinalysis test and a PCT level was obtained within the first 24 h of presentation .A PCT threshold of 0.25 ng/ml corresponded to the best combination of sensitivity (67%) and specificity (63%), with a positive predictive value and negative predictive value of 26% and 91%, respectively (Levine *et al*.,2018).

A study with161 participant aged 75 years or older in which 95 with probable bacterial infection and 66 without infection. Observed that Patients with probable bacterial infection criteria, 72% of them had PCT >0.5 ng/mL. Patients without infection, 8% of the patients had PCT >0.5 ng/mL. Sensitivity and specificity of PCT to bacterial infection with the cutoff value of 0.5 ng/mL was 72% and 92%, respectively (Gómez-Cerquera *et al*.,2015). Another study with 382 subjects (mean age, 78.9 years) were consecutively enrolled and stratified in two groups at the time of the admission based on the absence or presence of chronic kidney disease CKD, these two groups were further divided according to the presence or absence of sepsis and the serum PCT was analyised. It was detected that PCT was highly sensitive and specific in patients presenting with sepsis and no CKD. (Lo *et al*.,2016).

A study with a total of 308 elderly female patients with systemic inflammatory response syndrome (SIRS) was enrolled for this prospective study. The levels of all the inflammatory variables were significantly higher in the sepsis group than in non-sepsis group (all $P < 0.05$). Additionally, PCT and CRP were independent factors for diagnosis of blood stream infection. AUC of the combination of two biomarkers of PCT and CRP was 0.694 for diagnosis of sepsis which was higher than the either biomarkers alone with AUC of 0.628 for PCT and 0.627 for CRP .The combination group of PCT and CRP showed better values of sensitivity, specificity, positive predictive, and negative predictive (86.2%,59.1%,65.1 %, 81.3%) (Sun, Liang & Shao.,2017).

Surgical patients observed the same conclusion, a research of 200 patients undergoing four routine elective spinal procedures were included for analysis it observed that procalcitonin was the most sensitive and specific marker for the detection of surgical site infection in the immediate post-operative period with sensitivity and specificity of 100% and 95.2% respectively. Although Procalcitonin is an inflammatory marker, extent of surgical physiological insult did not alter its biokinetics as opposed to the other inflammatory markers making it a valuable marker of infection (Aljabi *et al*.,2019).

A prospective study where 44 patients presenting with febrile neutropenia after autologous stem cell transplantation were recruited. Mean levels of PCT and CRP were significantly increased in infections of critically ill patients who by dysfunction or failure of one or more organs/system depend on survival from advanced instruments of monitoring and therapy. The combination of the biomarkers PCT and Interleukin (IL-8) achieved a high sensitivity of 90% and specificity of 74% for the identification of serious

complications in febrile neutropenia, whereas the combination of CRP and PCT or IL-8 achieved a high sensitivity of 100%, but with the addition of a low specificity of 47or 41% (Michel *et al*.,2017).

A research with 60 burned people with and without infection assessed the value of the information for diagnosis of sepsis. A significantly higher PCT level was observed in the septic group compared to those without sepsis (8.45 ± 7.8 vs. 0.5 ± 1.0, respectively, $p < 0.001$); no significant differences were found in CRP levels. (Barati *et al*.,2008).

A study aimed to evaluate the diagnostic efficacy of serum procalcitonin (PCT), c- reactive protein (CRP) concentration and clinical pulmonary infection ventilator- associated pneumonia Of the included 49 patients, PCT serum concentration was significantly different ($P<0.05$) , the serum CRP was not statistically different between the two groups ($P>0.05$). A significant correlation was found between serum PCT and CRP concentrations ($r=0.55$, $P<0.01$)(Chen *et al*.,2018).

In summary of the literature, Serum PCT and CRP have good clinical diagnostic and prognostic value for patients with sepsis and septic shock.

Chapter Three

3. Materials and Methods

3.1. Materials:

3.1. 1. Study design:

A descriptive cross-sectional hospital based study.

3.1.2. Study area:

The study was carried out in International Medical Center hospital (IMC) A large hospital in Jeddah, Saudi Arabia.

3.1.3. Study period:

This study has been carried out between January 2018 and December 2018

3.1.4. Study population:

All patients come to (IMC) hospital with high fever > 38 C, chill, fatigue, body pain during the period from 1/1/2018 to 31/12/2018.

3.1.5. Inclusion criteria:

All multination patients pass by International Medical Center Hospital(IMC), Jeddah with high fever > 38C, physicians request to them complete blood count, C-reactive protein (CRP) and procalcitonin (PCT) along with blood culture & sensitivity.

3.1.6. Exclusion criteria:

1- Any known patients under antibiotic therapy before taking the blood samples for hs-CRP & PCT.
2- Cancer patients.

3.1.7. Ethical consideration:

Research purpose and objectives was explained to (IMC) hospital administration board.

Ethical approval was sought from (IMC) Hospital Administration Board, and Sudan University of Sciences and Technology.

(IMC) Hospital Administration Board had the right to voluntary informed consent.

The patients had the right to no harm (Privacy and confidentiality by using coded data).

3.2. Methodology:

3.2.1. Data collection:

The data collection started immediately after the approval was obtained, (IMC) Hospital provided access to the Hospital Information System (HIS) in which the questionnaire was filled using it, then serum samples prepared from each participant blood.

3.2.2. Measurement of PCT and CRP:

Cobase 601module which is highly innovative and patented Electrochemiluminescence (ECL) technology had been used to measured PCT and CRP.

PCT principle is depending on the Sandwich technique with a Total duration of the assay about 18 minutes.

- 1st incubation: Antigen in the sample (30 µL), a biotinylated monoclonal PCT-specific antibody, and a monoclonal PCT-specific antibody labeled with a ruthenium complexa) react to form a sandwich complex.
- 2nd incubation: After addition of streptavidin-coated microparticles, the complex becomes bound to the solid phase via interaction of biotin and streptavidin.
- The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell/ProCell M. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.
- Results are determined via a calibration curve which is instrument specifically generated by 2-point calibration and a master curve provided via the reagent barcode.

Hs-CRP Principle start with erythrocytes of the capillary or venous blood

sample are separated from the plasm by centrifugation. Then ,the plasma samples is diluted with HEPES buffer and transferred into a reaction chamber where it is mixed with CRP antibody on the latex particle. The concentration of CRP is calculated as a function of the changed absorbance measured at 525nm and 625 nm which is in relation to the amount of agglutination.

3.2.3. Quality Control Method:

Control sera were used with all samples to ensure the validity of the results long with the calibration curve.

3.2.3. Statistical analysis

Data obtained were analysis using SPSS version 25.0, the frequency distributions for independent variable and dependent variable was generated Student t-test

The sensitivity (true positive/ true positive + false negative), specificity (true negative/ true negative + false positives) were calculated for both PCT and CRP.

Chapter Four

4. Result

The study included 123 patients; come to the hospital with fever and other symptom .The physicians request to them complete blood count ,C-reactive protein (CRP) and procalcitonin (PCT) along with blood culture.

The results obtained were illustrated in tables and figures as follows

Table (4.1) Demographic and clinical data of the study group.

Table (4.2) Comparison between median of serum hs- CRP, PCT among study group according to Blood culture result.

Table (4.3) hs-CRP and PCT Sensitivity and Specificity with cut off 5mg/l for hs-CRP and 2ng/ml

Figure (4.1) ROC curves for hs-CRP and PCT. For hs-CRP; AUC=0.646, with CI(0.547-745), P- value 0.006, for PCT; AUC= 0.984 with CI(0.954-1.015), P.value 0.00.

Table (3.1): Demographic and clinical data of the study group.

Variable		Frequency	Percentage
Age / years	less than 30	6	4.9
	30-59	20	16.3
	≥ 60	97	78.9
Sex	Male	69	65.1
	Female	54	43.9
Sepsis	Yes	73	59.3
	No	50	40.7
hs-CRP mg/L	0-5	6	4.9
	>5	117	95.1
PCT ng/mL	0.5- 2	49	39.8
	>2	74	60.2
Bacterial sp	gram +ve		
	gram -ve		

Table (3.2): Comparison between median of serum hs- CRP, PCT among study group according to Blood culture result.

Variable	Infection absent (n=50) Median (IQR-3QR)	Infection present (n=73) Median (IQR-3QR)	P.Value
hs-CRP mg/L	90.4 (23.2- 145.2)	127.1 (63.1-228.2)	0.003
PCT ng/mL	0.33 (0.12- 0.84)	8.7 (3.6-24.6)	0.000

-Mann–Whitney U test was used to compared between the median of test
-P. value considered significant at level≥0.05.

Table (3.3): hs-CRP and PCT Sensitivity and Specificity with cut off 5mg/l for hs-CRP and 2ng/ml

	infection			
Variable	present	absent	Sensitivity	Specificity
hs-CRP mg/L ≤5	0	6	100%	88%
< 5	73	44		
PCT ng/mL <2.0	0	49	100%	98%
≥2.0	73	1		

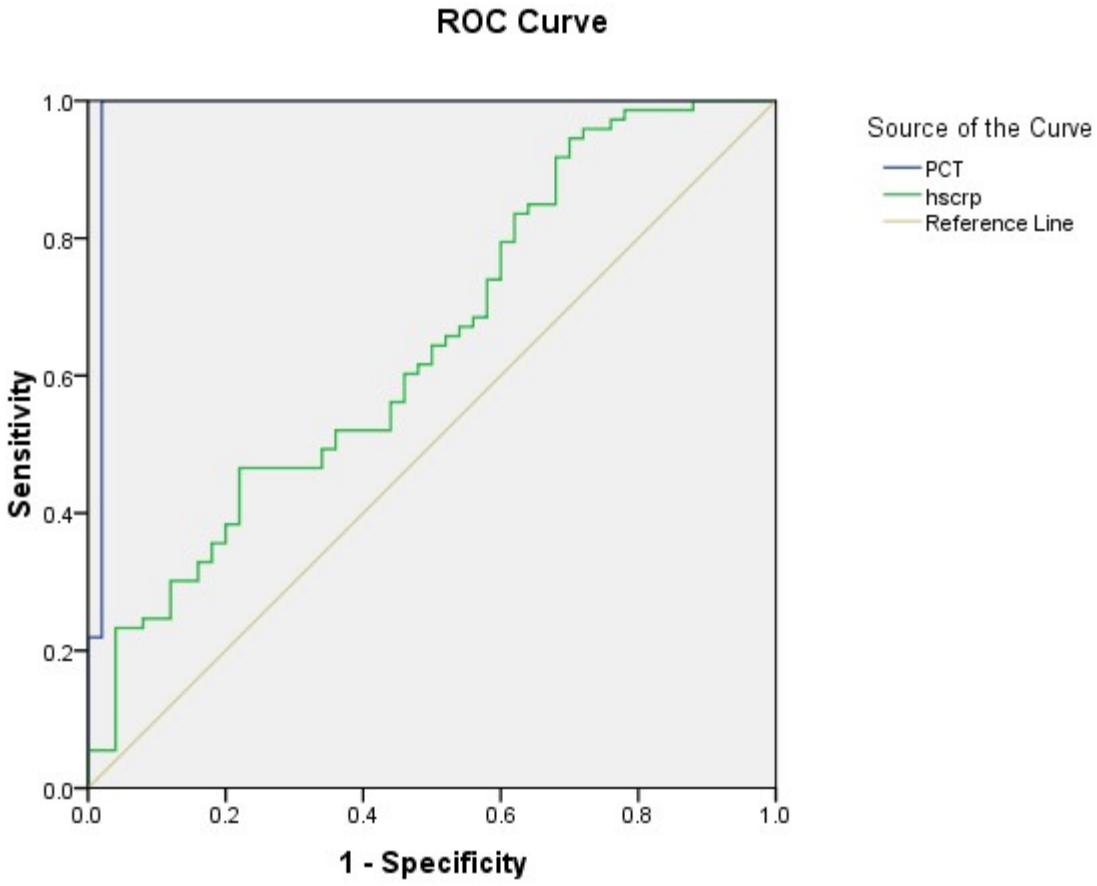

Fig (3.1): ROC curves for hs-CRP and PCT. For hs-CRP; AUC=0.646, with CI(0.547-745), P-value 0.006, for PCT; AUC= 0.984 with CI(0.954-1.015), P.value 0.00.

Area Under the Curve

Test Result Variable(s)	Area	Std. Error[a]	Asymptotic Sig.[b]	Asymptotic 95% Confidence Interval	
				Lower Bound	Upper Bound
PCT	.984	.016	.000	.954	1.015
hs-CRP	.646	.051	.006	.547	.745

a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

Chapter Five

5.1. DISCUSSION:

In this study both PCT and CRP values were higher in the positive than in the negative blood culture group. Previous studies have compared biomarkers in patient groups categorized according to PCT level (Harbarth S 2000 and Muller B., 2001) The present study demonstrated that the PCT levels of patients in the infected-group were significantly higher than those of non-infected group, while CRP levels were insignificantly higher on the non-infected group, this agree with (Kruif MD *et al.,* 2010 and Simon L *etal* .,2004) in the part concerning PCT and disagree in the part concerning CRP because they were demonstrated that the PCT and CRP levels of generalized pustular psoriasis (GPP) patients in the infected-group were significantly higher than those of non-infected group.,

It has been a controversial issue whether PCT or CRP was significantly correlated with age in the infected or non-infected population, the present study implied that PCT/CRP was not correlated with age which disagree with (Siyu Wang et al 2019) study which showed moderately positively correlated with age in the infected group, but not in the non-infected group .

In the present results sensitivity of procalcitonin was 92% and specificity was 82.5% while sensitivity of C-reactive protein was 99.0% and specificity was 13.5%. Other study showed relative results of PCT level was sensitive (88%)vs.(75%)(Simon *et al*.,2004).

A cross sectional study conducted in Sudan in the efficacy of procalcitonin in early diagnosis of neonatal sepsis. The results showed that CRP have 93.5% sensitivity and 50% specificity , on the other hand PCT show better sensitivity and specificity 96.8%,85.7 respectively (El-Amin et al.,2017).

A systematic review of 17 articles stated that the concentrations of both PCT

and CRP were higher in the serious bacterial infections (SBI) group than in the non- bacterial infection group. Sensitivity for differentiating bacterial infections from nonbacterial infections was higher for PCT compared with CRP(Hu *et al*.,2017).

Another study with 382 subjects (mean age, 78.9 years) were consecutively enrolled and stratified in two groups at the time of the admission based on the absence or presence of chronic kidney disease CKD, these two groups were further divided according to the presence or absence of sepsis and the serum PCT was analyzed. It was detected that PCT was highly sensitive and specific in patients presenting with sepsis and no CKD. (Lo *et al*.,2016).

A study included 276 cases and assessed procalcitonin (PCT) and C-reactive protein (CRP) for the prediction of subclinical intrauterine infection in patients with premature rupture of membranes (PROM). the sensitivity of PCT was significantly better than CRP (Li *et al.,*2016). Other study discovered unique results that the combination of two biomarkers of PCT and CRP was 0.694 for diagnosis of sepsis which was higher than the either biomarkers alone with AUC of 0.628 for PCT and 0.627 for CRP (Min *et al*.,2018). Another study revealed the area under the receiver operating characteristics curve in the diagnosis of sepsis was 0.97 for PCT ($p < 0.001$) with sensitivity of 100% and specificity of 89.3%. Non-survivors had a mean PCT level significantly higher than that of survivors (Barati *et al*., 2008).CRP sensitivity was of 78 % and specificity of 70 %. On the other hand, CRP specificity was (70%). Results showed that similar finding was found using CRP and PCT combination (Fattah MA.,2017).The results showed that CRP have 93.5% sensitivity and 50% specificity,

The results of this study go with the international studies that PCT have much higher Specificity level than CRP and CRP have more Sensitivity.

The diagnostic specificity of PCT in detecting bacterial infection is superior to CRP in GPP patients. It may be attributed to the fact, proven by previous studies, that PCT did not appear to be pivotally influenced by viral infections ,autoimmune or allergic disorders, immunosuppressives, or steroids, None the less, CRP is a biomarker of inflammation rather than a biomarker of infection. Its level rises in most pathological cases associated with inflammation, such as bacterial/viral infections, trauma, systemic disease flare and post-surgical period.

5.2 CONCLUSION:

- Procalcitonin has high level of sensitivity 92% and specificity82.5%.
- The C reactive protein test has very low level of specificity13.5 %.
- There is no relation between levels of PCT and different diagnosis.
- There is relation between levels of PCT and the causative organism if the organism is either Gram negative bacilli or Gram positive coci, this demand could be studied intensively on another study with a higher number of participants.
- Significant organism more likely to give abnormal level of PCT.
-

5.3 RECOMMENDATION:

- PCT test should be the test of choice for early detection of infection inpatients.
- Negative C reactive protein is conclusive for elimination of disease.
- Ministry of health should conduct lectures for medical staff to update them with these results.
- Further studies should be carried out on larger number of patients for detection of specific organisms groups.

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Appendix

1-Inform consent

2- Questionnaire

3-Method insert sheet

المركز الطبي الدولي
International Medical Center

To : Ms. Nashwa Mirghani Ahmed
Principal Investigator

Project: Diagnostic Value of Procacitonin in Comparison with C-Reactive Protein for Detecting bacterial blood Infection (Sepsis)

IMC-IRB #: 2019-11-116

From : Research Center, International Medical Center

Date : 27 November 2019

International Medical Center (IMC) IRB[1] has reviewed and approved your retrospective study proposal submission titled "**Diagnostic Value of Procacitonin in Comparison with C-Reactive Protein for Detecting bacterial blood Infection (Sepsis)**" and the below related documents:

1. IMC Retrospective Application form

The submission got the full approval with a super majority of members (without conflicts of interest) voted in favor of projects with no major/minor concerns.

Good luck and we wish you all the success.

Best Regards,

Prof. Ezzeldin Ibrahim
Chairman, Oncology Center
Academic Affairs & Research Executive Director

Prof. Ezzeldin M. Ibrahim

Executive Director, Research Center

[1]International Medical Center IRB is an Institutional Review Board, established in accordance with 7 CFR lc.107, 10 CFR 745.107, 14 CFR 1230.107, 15 CFR 27.107, 16 CFR 1028.107, 21 CFR 56.107, 22 CFR 225.107, 24 CFR 60.107, 28 CFR46.107, 32 CFR 219.107, 34 CFR 97.107, 38 CFR 16.107, 40 CFR 26.107, 45CFR 46.107, 45 CFR 690.107, or 49 CFR 11.107 and in compliance to ICH GCP.

[2]Independent affiliated IRB member

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ص.ب ٢١٧٢ جدة ٢١٤٥١ المملكة العربية السعودية، هاتف: ٩٠٠٠ ٦٥٠ ١٢ ٩٦٦+ فاكس: ٩٠٠١ ٦٥٠ ١٢ ٩٦٦+

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APPENDIX 1

INTERNATIONAL MEDICAL CENTER (IMC) IRB MEMBERS

S.no	Name	Position/Title
1	**Abdul Hameed Hassan**	Consultant, Family Medicine
2	**Ezzeldin Ibrahim**	Consultant, Oncology
3	**Ibrahim Mansoor**	Consultant, Anatomical Pathology & Clinical Chemistry
4	**Mohammad A. Albar**[4]	Consultant, Islamic Medicine
5	**Mohammed Janish**	Clinical Research Coordinator
6	**Raheel Shariff**	Consultant, Orthopedic
7	**Nashaat S. Hamza**	Consultant, Infection Control
8	**Hatem S. Bayoumy**	Clinical Pharmacist
9	**Abdullah E. Khalil**	Consultant, Emergency Medicine

[4]Independent affiliated IRB member from other institution, consulted on the applicable Islamic sharia

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ص.ب ٢١٧٢ جدة ٢١٤٥١ المملكة العربية السعودية، هاتف: ٩٠٠٠ ٦٥٠ ١٢ ٩٦٦+ فاكس: ٩٠٠١ ٦٥٠ ١٢ ٩٦٦+

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Sudan University of Science and Technology

College Of Graduate Studies

"Diagnostic Value of Procalcitonin in Comparison with C- reactive protein for detecting bacterial blood Infection"

(Study has been done on Saudi Arabia)

Questionnaire No ( )

Participant name &MRN.....

Age ……. years

Gender:

Male ( ) Female ( )

What is the type of organism/s have been discovered from the blood Culture?

……

Is the growth consider Significant? yes ( ) No ( )

Is the diagnosis of the case correlate with culture ?

yes ( ) No ( )

Laboratory Investigations:

PCT …… ng/ml

CRP……mg/l

Participant signature

cobas CRP Test

REF 08024669119 Σ 10 SYSTEM **cobas b** 101

English

Intended use

The Roche **cobas b** 101 is an in vitro diagnostic test system designed to quantitatively determine the C-reactive protein (CRP) in human capillary whole blood and serum, EDTA K2/K3 and lithium heparin anticoagulated whole blood and plasma by photometric measurement. Measurement of CRP is of use for the evaluation of inflammatory disorders and associated diseases, infection and tissue injury. The system is intended for use in point-of-care (PoC) settings such as pharmacies, physician offices, physician office laboratories, clinics and hospitals, and clinical laboratory settings.

Note: Please note that the catalogue number appearing on the package insert retains only the first 8 digits of the licensed 11-digit catalogue number: 08024669190 for the cobas CRP test. The last 3 digits -190 have been replaced by 119 for logistic purposes.

Summary

Most tissue-damaging processes such as infections, inflammatory diseases and malignant neoplasms are associated with a major acute phase response of the C-reactive protein (CRP) and other acute phase reactants (e.g. AAT, AAGP, C3C, C4, HAPT). The CRP response frequently precedes clinical symptoms, including fever. In healthy individuals CRP is a trace protein with a range up to 5 mg/L. After onset of an acute phase response the serum CRP concentration rises rapidly and extensively. Alterations are detectable within 6 to 8 hours and the peak value is reached within 24 to 48 hours. Levels of up to a thousandfold the normal value are associated with severe stimuli such as myocardial infarction, major trauma, surgery, or malignant neoplasms. CRP activates the classical complement pathway. CRP has a half-life of only a few hours, making it an ideal tool for clinical monitoring. Postoperative monitoring of CRP levels of patients indicates either the normal recovery process (decreasing levels to normal) or unexpected complications (persisting high levels). Measuring changes in the concentration of CRP provide useful diagnostic information about how acute and how serious a disease is. It also allows the assessment of complications during the disease and judgements about the disease genesis. Persistence of a high CRP concentration is usually a grave prognostic sign which generally indicates the presence of an uncontrolled infection. CRP determination may replace the classical determination of Erythrocytes Sedimentation Rate (ESR), due to its prompt response to changes in disease activity and its good correlation to ESR.[1,2,3,4]

Test principle

The erythrocytes of the capillary or venous blood sample are separated from the plasma by centrifugation. Then, the plasma sample is diluted with HEPES buffer and transferred into a reaction chamber where it is mixed with CRP antibody-latex reagent. The CRP in the diluted plasma binds with the CRP antibody on the latex particle. The concentration of CRP is calculated as a function of the changed absorbance measured at 525 nm and 625 nm which is in relation to the amount of agglutination.[5,6,7]

Reagents

One test contains:

HEPES buffer: 1.79 mg

Anti-human CRP antibody (goat) Latex-conjugate: 41.84 µg

Precautions and warnings

For in vitro diagnostic use.
Exercise the normal precautions required for handling all laboratory reagents.
Disposal of all waste material should be in accordance with local guidelines.
Safety data sheet available for professional user on request.

Reagent handling

Carefully tear open the foil pouch at the tear notch until one side is open.

Discard the disc if the foil pouch is found open or damaged, or if the disc is damaged, or the desiccant is missing, or loose desiccant particles or any other dirt or particles especially at the blood application zone are found.

Use **cobas** CRP Control in the same way as a blood sample.

Storage and stability

Store at 2-30 °C until the expiration date printed on the pouch. Do not freeze. If stored in a refrigerator, allow the disc to warm up in the closed pouch for at least 20 minutes before use. Once the pouch is opened, use the disc within 20 minutes. Protect the disc from direct sunlight. Do not store opened pouches in a refrigerator.

Specimen collection and preparation

For specimen collection and preparation only use suitable tubes or collection containers.

Use fresh human capillary whole blood or serum, EDTA K2/K3 or lithium heparin anticoagulated whole blood or plasma.

Do not use other anticoagulants or other additives.

For EDTA K2/K3 and lithium heparin anticoagulated whole blood and plasma samples, test within 8 hours of sample collection if stored at room temperature. If stored in the refrigerator plasma samples may be used up to 14 days and whole blood samples up to 3 days. Frozen serum and plasma samples stored at -20 °C may be used up to 30 days. Freeze only one time. Mix sample thoroughly before use. Do NOT use frozen whole blood to avoid the risk of hemolysis.

The marking on the disc shows where to apply the sample. If samples are used from a venipuncture or control material, use a standard pipette or dropper to form a drop. The disc is self-filling. Do not push the sample into the disc. Do not use syringes. Assure that the disc is free from blood outside the sample application zone and the hinge cover.

Sample volume: 12 µL

Sample stability on disc

ⓘ After sample application, the disc must be inserted immediately (in ≤ 120 seconds). Please follow the instructions in the **cobas b** 101 Operator's Manual.

Assay

Instructions for use

- Wash hands with soap. Warm water helps to stimulate the blood flow. Rinse the fingers extensively. Dry hands.
- Disinfect the fingertip by wiping three times the area to be lanced with a cotton swab or sterile gauze pad impregnated with 70 %-100 % isopropanol emollient free or 70 %-100 % ethanol emollient free; repeat the procedure with a second cotton swab or sterile gauze pad impregnated with 70 %-100 % isopropanol emollient free or 70 %-100 %ethanol emollient free, then dry with a cotton swab or sterile gauze pad.
- Prick the patient's finger by applying a single-use disposable lancing device (e.g. Accu-Chek Safe-T-Pro Plus). Make sure to follow the lancing device instructions for obtaining a blood sample.
- Wipe off the first drop of blood with a swab.
- With the imprinted side of the disc facing upwards, position the disc's suction point above the drop of blood. The disc is self-filling.
- Apply blood and observe that it has filled the marked area. Check the sample volume: turn the disc on its backside. The area marked in blue has to be filled completely with blood. Do not push the blood into the disc.
- Press hinge cover down firmly to close the disc.
- Assure that the disc is free from blood outside the sample application zone and the hinge cover.
- Insert the disc into the **cobas b** 101 instrument. Close the lid.
- The measurement starts automatically.

For more details, please refer to the **cobas b** 101 Quick Reference Guide or **cobas b** 101 Operator's Manual.

Materials provided

- REF 08024669190, **cobas** CRP Test, 10 tests

Materials required (but not provided)

- Single use disposable lancing device (e.g. Accu-Chek Safe-T-Pro Plus)
- REF 08024723190, **cobas** CRP Control
- REF 06378668190, **cobas b** 101 instrument
- Optical check disc
- General laboratory equipment (e.g., sample transfer pipette for venous blood or alcohol wipes for disinfection of the finger)

Calibration
This method has been standardized against the ERM DA 472/IFCC reference material. Each disc lot of the **cobas** CRP Test is traceable to ERM DA 472/IFCC reference material.[8,9]

The instrument automatically reads in the lot-specific calibration data from the barcode information printed on the disc, eliminating the need for calibration by the user.

Quality control
For quality control, use **cobas** CRP Control.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

QC info disc
Every **cobas** CRP Control kit contains a lot-specific QC info disc for quality control. This QC info disc contains the target values and ranges for the **cobas** CRP Test.

The instrument display prompts the user to insert the QC info disc. The **cobas b** 101 instrument reads the disc providing the lot specific target ranges.

Display of results
At the end of the automatic determination, the **cobas b** 101 instrument shows the result within 3-4 minutes. The concentration of CRP will be displayed in mg/L or in mg/dL.

Limitations - interference
Hematocrit levels between 20 % and 60 % do not affect results.

Criterion: Recovery within ± 10 % of initial values at CRP concentrations of 10.0 mg/L and 40.0 mg/L.

Hemolysis: No significant interference up to a hemoglobin concentration of 500 mg/dL.

Icterus:[10] No significant interference up to a conjugated/unconjugated bilirubin concentration of 50 mg/dL.

Lipemia (Intralipid):[10] No significant interference up to an Intralipid and Triglyceride concentration of 750 mg/dl.

Glycemia:[10] No significant interference up to a glucose level of 1000 mg/dL. A fasting sample is not required.

Rheumatoid factors: No significant interference up to 1200 IU/mL.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[11]

Drug interferences are measured based on recommendations given in CLSI guidelines EP07 and EP37 and other published literature. Effects of concentrations exceeding these recommendations have not been characterized.

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

Limits and ranges
Measuring range
3.0-400 mg/L or 0.30-40.0 mg/dL

Expected values
Adults: < 5.0 mg/L (< 0.5 mg/dL)[2,12]

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Specific performance data
Representative performance data on the instruments are given below. Results obtained in individual laboratories may differ.

Precision
Precision was determined using controls in a CLSI EP5-A3 protocol. Precision was measured with 3 lots of **cobas** CRP Test using 5 different serum samples at the medical decision points and 2 **cobas** CRP Control solution levels over 21 days with 2 runs per day and duplicate measurements per run and specimen. The following results were obtained for a representative lot:

	Repeatability			Intermediate precision		
Sample	Mean mg/L	SD mg/L	CV %	Mean mg/L	SD mg/L	CV %
Sample Healthy	5.1	0.13	2.5	5.1	0.17	3.3
Sample Cut off	10.0	0.23	2.3	10.0	0.24	2.4
Sample Decision	39.9	0.93	2.3	39.9	0.98	2.5
Sample Acute	93.4	1.62	1.7	93.4	1.84	2.0
Sample Acute High	351	7.99	2.3	351	8.42	2.4
Control Level 1	9.7	0.29	2.9	9.7	0.30	3.1
Control Level 2	39.2	0.79	2.0	39.2	1.09	2.8

Method comparison
CRPNX
A comparison experiment using 3 different reagent lots with serum samples measured with **cobas** CRP Test on the **cobas b** 101 instrument (y) and CRP-latex X2 "Seiken" NX reagent on the **cobas c** 501 analyzer (x) gave the following correlation for a representative lot (Weighted Deming regression method).[13,14]

Sample size: 140
Slope: 1.00
Intercept: 0.0934
Pearson r: 0.998
Mean bias in the range 3.0-200 mg/L: 0.5 %
Mean bias in the range > 200-400 mg/L: 3.5 %
Bias at 5.0 mg/L: 1.9 %
Bias at 10.0 mg/L: 0.9 %

CRPLX
A second comparison experiment using 3 different reagent lots with serum samples measured with **cobas** CRP Test on the **cobas b** 101 instrument (y) and CRPLX C- Reactive Protein (Latex) reagent on the **cobas c** 501 analyzer (x) gave the following correlation for a representative lot (Weighted Deming regression method).[13,14]

Sample size: 122
Slope: 1.05
Intercept: 0.08
Pearson r: 0.996
Mean bias in the range 3.0-200 mg/L: 6.41 %
Mean bias in the range > 200-400 mg/L: 1.79 %
Bias at 5.0 mg/L: 6.82 %
Bias at 10.0 mg/L: 6.02 %

CRPNX or CRPLX are possibly not commercially available in all regions.

References

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For further information, please refer to the appropriate Operator's Manual for the instrument concerned, and the Method Sheets of all necessary components.

A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see dialog.roche.com for definition of symbols used):

CONTENT	Contents of kit
	Analyzers/Instruments on which reagents can be used
REAGENT	Reagent
CALIBRATOR	Calibrator
→	Volume after reconstitution or mixing
GTIN	Global Trade Item Number

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com

ms_05056888003V11.0

Elecsys BRAHMS PCT

cobas®

Procalcitonin

REF	Σ	SYSTEM
		Elecsys 2010 MODULAR ANALYTICS E170
05056888 003	100	**cobas e** 411 **cobas e** 601 **cobas e** 602

English

Intended use

Immunoassay for the in vitro quantitative determination of PCT (procalcitonin) in human serum and plasma.

The Elecsys BRAHMS PCT assay can be used to aid in the early detection of clinically relevant bacterial infections.

The **e**lectro**c**hemiluminescence **i**mmuno**a**ssay "ECLIA" is intended for use on Elecsys and **cobas e** immunoassay analyzers.

Summary

Procalcitonin (PCT) is a 116 amino acid prohormone with a molecular weight of approximately 12.7 kD. PCT is expressed by neuroendocrine cells (C cells of the thyroid, pulmonary and pancreatic tissues) and successively enzymatically cleaved into (immature) calcitonin, katacalcin, and an N-terminal region. The blood of healthy individuals contains only low levels of PCT.[1,2] It was discovered that PCT increases during bacterial infection.

It is probable that multiple tissues express PCT throughout the body in response to sepsis as was shown in an animal model.[3] PCT circulating in septic patients consists of only 114 amino acids lacking the N-terminal dipeptide Ala-Pro.[4]

Increased PCT levels are often found in patients suffering from bacterial sepsis, especially severe sepsis and septic shock.[5,6,7,8,9,10] PCT is considered as a prognostic marker to support outcome prediction in sepsis patients.[8,11,12,13]

In acute pancreatitis PCT was found to be a reliable indicator of severity and of major complications.[14,15]

In patients suffering from community-acquired respiratory tract infections or ventilator-induced pneumonia PCT has been proposed as a guide for the decision of antibiotic treatment necessity and to monitor treatment success.[16,17]

Test principle

Sandwich principle. Total duration of assay: 18 minutes.

- 1st incubation: Antigen in the sample (30 µL), a biotinylated monoclonal PCT-specific antibody, and a monoclonal PCT-specific antibody labeled with a ruthenium complex[a)] react to form a sandwich complex.
- 2nd incubation: After addition of streptavidin-coated microparticles, the complex becomes bound to the solid phase via interaction of biotin and streptavidin.
- The reaction mixture is aspirated into the measuring cell where the microparticles are magnetically captured onto the surface of the electrode. Unbound substances are then removed with ProCell/ProCell M. Application of a voltage to the electrode then induces chemiluminescent emission which is measured by a photomultiplier.
- Results are determined via a calibration curve which is instrument-specifically generated by 2-point calibration and a master curve provided via the reagent barcode.

a) Tris(2,2'-bipyridyl)ruthenium(II)-complex ($Ru(bpy)_3^{2+}$)

Reagents - working solutions

The reagent rackpack (M, R1, R2) is labeled as PCT.

M Streptavidin-coated microparticles (transparent cap), 1 bottle, 6.5 mL:
Streptavidin-coated microparticles 0.72 mg/mL; preservative.

R1 Anti-PCT-Ab~biotin (gray cap), 1 bottle, 9 mL:
Biotinylated monoclonal anti-PCT antibody (mouse) 2.0 µg/mL; phosphate buffer 95 mmol/L, pH 7.5; preservative.

R2 Anti-PCT-Ab~$Ru(bpy)_3^{2+}$ (black cap), 1 bottle, 9 mL:
Monoclonal anti-PCT antibody (mouse) labeled with ruthenium complex 5.6 µg/mL; phosphate buffer 95 mmol/L, pH 7.5; preservative.

PCT Cal1 PCT calibrator 1 (white cap), 1 bottle (lyophilized) for 4 mL:
PCT (recombinant) approximately 0.10 ng/mL in a human serum matrix; preservative.

PCT Cal2 PCT calibrator 2 (black cap), 1 bottle (lyophilized) for 4 mL:
PCT (recombinant) approximately 54 ng/mL in a human serum matrix; preservative.

PC PCT1 PreciControl PCT 1 (beige cap), 2 bottles (lyophilized) each for 4 mL:
PCT (recombinant) approximately 0.50 ng/mL in a human serum matrix; preservative.

PC PCT2 PreciControl PCT 2 (brown cap), 2 bottles (lyophilized) each for 4 mL:
PCT (recombinant) approximately 10 ng/mL in a human serum matrix; preservative.

Calibrators: The exact lot-specific calibrator values are encoded in the barcoded labels of the test-specific reagent.

Controls: The exact lot-specific target values and ranges are encoded in the barcodes as well as printed on the enclosed (or electronically available) value sheet.

Precautions and warnings

For in vitro diagnostic use.
Exercise the normal precautions required for handling all laboratory reagents.
Disposal of all waste material should be in accordance with local guidelines.
Safety data sheet available for professional user on request.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

2-methyl-2H-isothiazol-3-one hydrochloride

EUH 208 May produce an allergic reaction.

Product safety labeling primarily follows EU GHS guidance.

All human material should be considered potentially infectious. All products derived from human blood are prepared exclusively from the blood of donors tested individually and shown to be free from HBsAg and antibodies to HCV and HIV. The testing methods applied were FDA-approved or cleared in compliance with the European Directive 98/79/EC, Annex II, List A.

However, as no testing method can rule out the potential risk of infection with absolute certainty, the material should be handled with the same level of care as a patient specimen. In the event of exposure, the directives of the responsible health authorities should be followed.[18,19]

Avoid foam formation in all reagents and sample types (specimens, calibrators and controls).

Reagent handling

The reagents in the kit (M, R1 and R2) are ready for use and are supplied in bottles compatible with the system.

Calibrators and controls

Carefully dissolve the contents of one bottle by adding exactly 4 mL of distilled or deionized water and allow to stand closed for 15 minutes to

reconstitute. Mix carefully, avoiding foam formation. Transfer the reconstituted calibrators/controls into empty labeled snap-cap bottles.

Unless the entire volume is necessary for calibration and quality control on the analyzer, transfer aliquots of the freshly reconstituted calibrators and controls into empty snap-cap bottles (CalSet Vials/ControlSet Vials). Attach the supplied labels to these additional bottles. Store the aliquots at -20 °C for later use. Perform **only one** calibration or control procedure per aliquot.

Note: Do not combine bottles from different lots. Use only control bottles out of one lot with each other.

All information required for correct operation is read in from the respective reagent barcodes.

Please note: Both the vial labels, and the additional labels (if available) contain 2 different barcodes. The barcode between the yellow markers is for **cobas** 8000 systems only. If using a **cobas** 8000 system, please turn the vial cap 180° into the correct position so the barcode can be read by the system. Place the vial on the instrument as usual.

Storage and stability

Store at 2-8 °C.

Do not freeze.

Store the Elecsys reagent kit **upright** in order to ensure complete availability of the microparticles during automatic mixing prior to use.

Stability of the reagent rackpack	
unopened at 2-8 °C	up to the stated expiration date
after opening at 2-8 °C	12 weeks
on the analyzers	4 weeks

Stability of the calibrators and controls	
lyophilized calibrators/controls	up to the stated expiration date
reconstituted calibrators/controls on the analyzers	2 hours (use only once)
reconstituted calibrators/controls at -20 °C	3 months (freeze only once)

Store the calibrators and controls **upright** in order to prevent the solution from adhering to the snap-cap.

Specimen collection and preparation

Only the specimens listed below were tested in a sufficient number and found acceptable.

Serum collected using standard sampling tubes or tubes containing separating gel.

Li-heparin, K_2-EDTA and K_3-EDTA plasma.

Criterion: Slope 0.9-1.1 + intercept within < ± 2 x analytical sensitivity (LDL) + coefficient of correlation > 0.95.

Stable for 24 hours at 2-8 °C, 3 months at -20 °C. Freeze only once.

After drawing the blood, measure samples within 24 hours or freeze at -20 °C.

Frozen samples can lead to a lower recovery of up to 8 %.

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Centrifuge samples containing precipitates before performing the assay.

Do not use samples and controls stabilized with azide.

Ensure the samples, calibrators and controls are at 20-25 °C prior to measurement.

Due to possible evaporation effects, samples, calibrators and controls on the analyzers should be analyzed/measured within 2 hours.

Materials provided

See "Reagents – working solutions" section for reagents.

- 2 barcode cards
- control barcode sheet
- 2 x 8 bottle labels (calibrators)
- 2 x 14 bottle labels (controls)
- 6 empty labeled snap-cap bottles

Materials required (but not provided)

- REF 11776576322, CalSet Vials, 2 x 56 empty snap-cap bottles
- REF 03142949122, ControlSet Vials, 2 x 56 empty snap-cap bottles
- General laboratory equipment
- Elecsys 2010, MODULAR ANALYTICS E170 or **cobas e** analyzer

Accessories for Elecsys 2010 and **cobas e** 411 analyzers:

- REF 11662988122, ProCell, 6 x 380 mL system buffer
- REF 11662970122, CleanCell, 6 x 380 mL measuring cell cleaning solution
- REF 11930346122, Elecsys SysWash, 1 x 500 mL washwater additive
- REF 11933159001, Adapter for SysClean
- REF 11706802001, Elecsys 2010 AssayCup, 60 x 60 reaction vessels
- REF 11706799001, Elecsys 2010 AssayTip, 30 x 120 pipette tips

Accessories for MODULAR ANALYTICS E170, **cobas e** 601 and **cobas e** 602 analyzers:

- REF 04880340190, ProCell M, 2 x 2 L system buffer
- REF 04880293190, CleanCell M, 2 x 2 L measuring cell cleaning solution
- REF 03023141001, PC/CC-Cups, 12 cups to prewarm ProCell M and CleanCell M before use
- REF 03005712190, ProbeWash M, 12 x 70 mL cleaning solution for run finalization and rinsing during reagent change
- REF 12102137001, AssayTip/AssayCup Combimagazine M, 48 magazines x 84 reaction vessels or pipette tips, waste bags
- REF 03023150001, WasteLiner, waste bags
- REF 03027651001, SysClean Adapter M

Accessories for all analyzers:

- REF 11298500316, ISE Cleaning Solution/Elecsys SysClean, 5 x 100 mL system cleaning solution

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator's manual for analyzer-specific assay instructions.

Resuspension of the microparticles takes place automatically prior to use. Read in the test-specific parameters via the reagent barcode. If in exceptional cases the barcode cannot be read, enter the 15-digit sequence of numbers.

Bring the cooled reagents to approximately 20 °C and place on the reagent disk (20 °C) of the analyzer. Avoid foam formation. The system automatically regulates the temperature of the reagents and the opening/closing of the bottles.

Place the reconstituted calibrators (in the system-compatible bottles with barcoded labels) in the sample zone.

All the information necessary for calibrating the assay is automatically read into the analyzer.

After calibration has been performed, discard the calibrators.

Analyze the controls PC PCT1 and PC PCT2. The information on the barcoded label of the control serum bottle is read in automatically. After the control procedure has been performed, discard the controls.

Calibration

Traceability: This method has been standardized against the BRAHMS PCT LIA assay.

Every Elecsys BRAHMS PCT reagent set has a barcoded label containing specific information for calibration of the particular reagent lot. The predefined master curve is adapted to the analyzer using PCT Cal1 and PCT Cal2.

Elecsys BRAHMS PCT

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Calibrator sequence on all systems: Always measure PCT Cal2 before PCT Cal1.

Calibration frequency: Calibration must be performed once per reagent lot using fresh reagent (i.e. not more than 24 hours since the reagent kit was registered on the analyzer). Renewed calibration is recommended as follows:

- after 8 weeks when using the same reagent lot
- after 7 days (when using the same reagent kit on the analyzer)
- as required: e.g. quality control findings outside the defined limits

Quality control

For quality control, use PC PCT 1 and PC PCT 2.

In addition, other suitable control material can be used.

Controls for the various concentration ranges should be run individually at least once every 24 hours when the test is in use, once per reagent kit, and following each calibration.

The control intervals and limits should be adapted to each laboratory's individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

Note: When using two reagent kits with different lots in the same run, the controls will be measured with both reagent lots. Use only control values measured with the corresponding lots.

Calculation

The analyzer automatically calculates the analyte concentration of each sample in ng/mL.

Limitations - interference

The assay is unaffected by icterus (bilirubin < 428 µmol/L or < 25 mg/dL), hemolysis (Hb < 0.559 mmol/L or < 0.900 g/dL), lipemia (Intralipid < 1500 mg/dL) and biotin (< 123 nmol/L or < 30 ng/mL).

Criterion: Recovery within ± 15 % of initial value.

Samples should not be taken from patients receiving therapy with high biotin doses (i.e. > 5 mg/day) until at least 8 hours following the last biotin administration.

No interference was observed from rheumatoid factors up to a concentration of 1500 IU/mL.

There is no high-dose hook effect at PCT concentrations up to 1000 ng/mL.

In vitro tests were performed on 18 commonly used and 10 special pharmaceuticals. No interference with the assay was found.

In rare cases, interference due to extremely high titers of antibodies to analyte-specific antibodies, streptavidin or ruthenium can occur. These effects are minimized by suitable test design.

PCT levels can be increased in certain situations without infectious origin. These include, but are not limited to:[20]

- prolonged or severe cardiogenic shock
- prolonged severe organ perfusion anomalies
- small cell lung cancer or medullary C-cell carcinoma of the thyroid
- early after major trauma, major surgical intervention, severe burns
- treatments which stimulate the release of pro-inflammatory cytokines
- neonates (< 48 h after birth)[21]

For diagnostic purposes, the results should always be assessed in conjunction with the patient's medical history, clinical examination and other findings.

Limits and ranges

Measuring range

0.02-100 ng/mL (defined by the lower detection limit and the maximum of the master curve). Values below the lower detection limit are reported as < 0.02 ng/mL. Values above the measuring range are reported as > 100 ng/mL.

Lower limits of measurement

Lower detection limit of the test

Lower detection limit: ≤ 0.02 ng/mL

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying two standard deviations above that of the lowest standard (master calibrator, standard 1 + 2 SD, repeatability study, n = 21).

Dilution

Samples with PCT concentrations above the measuring range can be diluted manually with PCT negative human serum or plasma. The recommended dilution is 1:4. The concentration of the diluted sample must be > 1.0 ng/mL. After manual dilution, multiply the result by the dilution factor.

Expected values

Reference range

A study performed with the Elecsys BRAHMS PCT assay using 492 samples from apparently healthy males (245) and females (247) revealed the following normal value: 0.046 ng/mL (95th percentile).

Clinical cut-off

Results obtained with the Elecsys BRAHMS PCT assay are in agreement with the literature.[20] A study performed on samples from patients admitted to an ICU (intensive care unit) showed that PCT values:

< 0.5 ng/mL represent a low risk of severe sepsis and/or septic shock

> 2.0 ng/mL represent a high risk of severe sepsis and/or septic shock

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Clinical performance

Clinical studies were conducted on samples from 283 ICU patients. The patients were classified into categories based on the ACCP/SCCM (American College of Chest Physicians/Society of Critical Care Medicine) consensus criteria on their first day of ICU admission: SIRS (systemic inflammatory response syndrome), sepsis, severe sepsis and septic shock.[22]

The PCT values of the patients with SIRS (n = 95) or sepsis (n = 71) compared to patients with severe sepsis (n = 60) or septic shock (n = 57) were as follows:

Results with a cut-off at 0.5 ng/mL

	Clinical classification		
Elecsys BRAHMS PCT	SIRS	Severe sepsis/ septic shock	Total
< 0.5 ng/mL	63	5	68
≥ 0.5 ng/mL	32	112	144
Total	95	117	212

Based on the above data the sensitivity was 96 %, the specificity 66 %, the positive predictive value 78 % and the negative predictive value 93 %.

	Clinical classification		
Elecsys BRAHMS PCT	SIRS	Sepsis	Total
< 0.5 ng/mL	63	25	88
≥ 0.5 ng/mL	32	46	78
Total	95	71	166

Based on the above data the sensitivity was 65 %, the specificity 66 %, the positive predictive value 59 % and the negative predictive value 72 %.

Results with a cut-off at 2 ng/mL

	Clinical classification		
Elecsys BRAHMS PCT	SIRS	Severe sepsis/ septic shock	Total
< 2 ng/mL	88	18	106
≥ 2 ng/mL	7	99	106
Total	95	117	212

Based on the above data the sensitivity was 85 %, the specificity 93 %, the positive predictive value 93 % and the negative predictive value 82 %.

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	Clinical classification		
Elecsys BRAHMS PCT	SIRS	Sepsis	Total
< 2 ng/mL	88	55	143
≥ 2 ng/mL	7	16	23
Total	95	71	166

Based on the above data the sensitivity was 23 %, the specificity 93 %, the positive predictive value 70 % and the negative predictive value 62 %.

Specific performance data

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined using Elecsys reagents, pooled human serum/plasma and controls in a protocol (EP5-A2) of the CLSI (Clinical and Laboratory Standards Institute): 2 runs per day in duplication each for 21 days (n = 84). The following results were obtained:

Elecsys 2010 and **cobas e** 411 analyzers					
		Repeatability		**Intermediate precision**	
Sample	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
Human plasma 1	0.060	0.005	8.8	0.010	16.3
Human plasma 2	0.622	0.013	2.1	0.026	4.2
Human plasma 3	41.2	0.879	2.1	2.02	4.9
PreciControl PCT1	0.520	0.007	1.3	0.019	3.7
PreciControl PCT2	10.2	0.096	0.9	0.404	4.0

MODULAR ANALYTICS E170, **cobas e** 601 and **cobas e** 602 analyzers					
		Repeatability		**Intermediate precision**	
Sample	Mean ng/mL	SD ng/mL	CV %	SD ng/mL	CV %
Human serum 1	0.080	0.006	7.1	0.007	8.7
Human serum 2	0.431	0.008	1.8	0.011	2.6
Human serum 3	54.4	0.618	1.1	0.895	1.6
PreciControl PCT1	0.491	0.013	2.6	0.016	3.2
PreciControl PCT2	9.59	0.181	1.9	0.222	2.3

Method comparison

A comparison of the Elecsys BRAHMS PCT assay (y) with the BRAHMS PCT LIA (x) using human heparin plasma gave the following correlations (ng/mL):

Number of samples measured: 152

Passing/Bablok[23]	Linear regression
y = 1.065x - 0.090	y = 1.143x - 0.194
τ = 0.856	r = 0.981

The sample concentrations were between approximately 0.3 and approximately 82 ng/mL.

A comparison of the Elecsys BRAHMS PCT assay (y) with the BRAHMS PCT sensitive KRYPTOR (x) using human heparin plasma gave the following correlations (ng/mL):

Number of samples measured: 185

Passing/Bablok[23]	Linear regression
y = 0.850x - 0.035	y = 1.090x - 0.709
τ = 0.953	r = 0.988

The sample concentrations were between approximately 0.04 and approximately 85 ng/mL.

Analytical specificity

The Elecsys BRAHMS PCT assay does not show any significant cross reactions with the following substances, tested with PCT concentrations of approx. 0.4 ng/mL and 1.5 ng/mL (max. tested concentration):

Substances	Non-interfering concentrations (ng/mL)
Human katacalcin	30
Human calcitonin	10
Human alpha-CGRP[b)]	10000
Human beta-CGRP	10000

b) Calcitonin Gene-Related Peptide

Functional sensitivity

≤ 0.06 ng/mL

The functional sensitivity is the lowest analyte concentration that can be reproducibly measured with an intermediate precision CV of 20 %.

Concordance with BRAHMS PCT LIA/BRAHMS PCT sensitive KRYPTOR

A comparison study was performed with the Elecsys BRAHMS PCT assay and the BRAHMS PCT LIA. Cut-off values of 0.5 ng/mL and 2 ng/mL have been evaluated.

	BRAHMS PCT LIA		
Elecsys BRAHMS PCT	< 0.5 ng/mL	≥ 0.5 ng/mL	Total
< 0.5 ng/mL	104	49	153
≥ 0.5 ng/mL	6	370	376
Total	110	419	529

	BRAHMS PCT LIA		
Elecsys BRAHMS PCT	< 2 ng/mL	≥ 2 ng/mL	Total
< 2 ng/mL	266	10	276
≥ 2 ng/mL	11	242	253
Total	277	252	529

The concordance between both assays was 90 % at the cut-off value of 0.5 ng/mL and 96 % at the cutoff-value of 2 ng/mL.

The Elecsys BRAHMS PCT assay was also compared to the BRAHMS PCT sensitive KRYPTOR. Cut-off values of 0.5 ng/mL and 2 ng/mL have been evaluated.

	BRAHMS PCT sensitive KRYPTOR		
Elecsys BRAHMS PCT	< 0.5 ng/mL	≥ 0.5 ng/mL	Total
< 0.5 ng/mL	183	20	203
≥ 0.5 ng/mL	2	392	394
Total	185	412	597

	BRAHMS PCT sensitive KRYPTOR		
Elecsys BRAHMS PCT	< 2 ng/mL	≥ 2 ng/mL	Total
< 2 ng/mL	312	24	336
≥ 2 ng/mL	1	260	261
Total	313	284	597

The concordance between both assays was 96 % at the cut-off value of 0.5 ng/mL and 96 % at the cutoff-value of 2 ng/mL.

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For further information, please refer to the appropriate operator's manual for the analyzer concerned, the respective application sheets, the product information and the Method Sheets of all necessary components (if available in your country).

A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Reagent developed in collaboration with B·R·A·H·M·S.

B·R·A·H·M·S PCT

ELECSYS

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard.

CONTENT	Contents of kit
SYSTEM	Analyzers/Instruments on which reagents can be used
REAGENT	Reagent
CALIBRATOR	Calibrator
→	Volume after reconstitution or mixing
GTIN	Global Trade Item Number

CE

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