

To the Chairman of the Scientific Jury
determined by order No. R-109-410/26.10.2022
of the Rector of Medical University – Varna city

R E V I E W

by Prof. Rosen Gospodinov Kolarov, DMD, PhD

Varna city, Faculty of dental medicine, Department of Oral surgery
of a dissertation paper for the award of educational and scientific degree "Doctor"

Field of higher education: 7. "Health and sports";

Professional direction: 7.1. "Medicine";
"Surgery"

Author: Yanko Georgiev Yankov, MD

Independent doctoral student

Department of General and operative surgery

Subject:

"Procalcitonin and delta neutrophil index levels in the surgery of head and neck
inflammatory diseases"

Scientific Directors:

Prof. Nikola Yordanov Kolev, MD, PhD, DSc

Assoc. Prof. Yana Dimitrova Bocheva, MD, PhD

1. General presentation of the procedure and the doctoral student

The present review was prepared based on the Order of the Rector of the Medical University – Varna city No. R-109-410/26.10.2022, with an appointed Scientific Jury under the procedure for public defense of the described dissertation work.

The presented set of materials on paper and electronic media are in accordance with Art. 24, para. 6 and Art. 30, para. 3 of PPZRASRB, art. 68, para. 1 of the Regulations for the development of the academic staff of the Medical University – Varna city and were provided to me within the statutory period.

The doctoral student has submitted a total of 4 scientific works related to the topic of the dissertation published in scientific publications and presented at scientific forums.

All documents are prepared and presented properly.

2. Brief biographical data for the doctoral student

Yanko Georgiev Yankov, MD was born on 17.04.1989 in the town of Shumen.

In 2014, he graduated from the Medical University "Prof. Dr. Paraskev Stoyanov" - city of Varna with excellent (6.00) success and was awarded the "Golden Hippocrates" award, as well as the "First Degree" award from the Bulgarian Medical Union for excellent success.

From 2015 to the present, he has been working as a medical doctor in the Clinic of Maxillofacial Surgery at University General Hospital (UGH) "Sveta Marina" – Varna city.

In 2016, he graduated with a master's degree in Health Management at the Medical University "Prof. Dr. Paraskev Stoyanov" – city of Varna.

In the period February - October 2019, he worked as a part-time assistant in Oral and Maxillofacial Surgery at the Department of Oral and maxillofacial surgery at the Faculty of Dental Medicine and at the Department of General and operative surgery at the Faculty of Medicine of Medicine University "Prof. Dr. Paraskev Stoyanov" – city of Varna.

In 2019, after winning a competition, he was appointed full-time assistant in Maxillofacial surgery at the Department of General and operative surgery in the same university.

In July 2020, he acquired a specialty in Maxillofacial surgery.

From September 2021, he is a doctoral student of an independent form of study in the doctoral program in "Surgery" at the Department of General and operative surgery at the Medical University of Varna city.

Yanko Yankov, MD has a total work experience of 8 years, of which 4 as a teacher.

Yanko Yankov, MD is a member of the Bulgarian medical union (BLS).

He speaks English at a good level.

3. Actuality of the topic and appropriateness of the set goals and tasks

The topic of the dissertation examines a current and important problem for medical science and practice. The goal is clearly formulated, the tasks are defined correctly and are carried out using modern research methods.

The dissertation work was discussed, accepted and referred for defense before a scientific jury by the Departmental Council - protocol No. 8 of 30.09.2022 of the Department of General and operative surgery at the Medical University "Prof. Dr. Paraskev Stoyanov" - city of Varna.

4. Knowing the problem

Inflammatory diseases of the face and neck are common diseases. They are of extremely great social and psycho-emotional significance for the patient. Their complications are often protracted and severe, with long stays in intensive care units and wards, and sometimes have an unfavorable prognosis. These diseases are one of the conditions that burden the health system the most.

Laboratory parameters known to us, such as the value of leukocytes, neutrophils, ESR and CRP do not always increase their absolute value during the course of the infectious process. For this reason, they may prove to be insufficiently reliable and definitive markers for the diagnosis, treatment, follow-up and prediction of the outcome of the disease.

PCT is one of the most promising biomarkers of inflammation in the diagnosis of infectious diseases. Its increased values in the blood plasma indicate the degree of bacterial

inflammation and appear to be an earlier marker for the diagnosis of inflammatory diseases than the classic indicators of inflammation. Data in the literature indicate that PCT as a marker has a very good diagnostic profile in their diagnosis. PCT has been defined as an early marker of inflammation. It can detect the development of the inflammatory process even before the values of WBC, neutrophils and CRP have increased. However, a number of causes other than inflammation can induce increased PCT synthesis.

All this indicates the need for further studies to clarify its predictive value in the diagnosis of inflammatory diseases. This fact forms a large part of the rationale for the present study.

In order to establish the clinical applicability of PCT and DNI as indicators of inflammation, it is necessary to evaluate their diagnostic reliability and the effectiveness of their methodologically derived cut-off values in the studied patients with head and neck abscesses of odontogenic and non-odontogenic origin.

Yanko Yankov, MD has done a thorough critical analysis of the literature, formulating the unresolved problems on the subject.

In his work, Yanko Yankov, MD shows skills for independent conduct of scientific research.

On the basis of the analysis of the scientific research conducted so far on the problem, the goal of the scientific development has been logically deduced, namely:

"To determine the diagnostic value of procalcitonin (PCT) and delta neutrophil index (DNI), their predictability, sensitivity and specificity in inflammatory diseases of the head and neck and to analyze their advantages and disadvantages in the diagnostic-therapeutic plan of these diseases relative to already established markers of inflammation such as WBC, neutrophils and CRP."

The presentation is written in good scientific language.

5. Research methodology

The realization of the goal was achieved through the implementation of five main tasks, namely:

1. Determination of the average values of PCT, DNI, CRP, WBC and neutrophils in the studied group of patients with head and neck abscesses, the subgroups of patients with odontogenic abscesses, with non-odontogenic abscesses and the control group of healthy people;
2. Determination and analysis of gender differences regarding the studied markers PCT, DNI, CRP, WBC and neutrophils in the studied population;
3. Determination and analysis of correlations between PCT, DNI, CRP, WBC and neutrophils in the studied group of patients with head and neck abscesses, in the subgroups with odontogenic and non-odontogenic abscesses and in the control group of healthy people;
4. Determination of cut-off diagnostic and reference values of PCT and DNI in the studied groups of patients with odontogenic and non-odontogenic abscesses of the head and neck;
5. Determination of sensitivity, specificity and predictability of PCT, DNI, CRP, WBC and neutrophils in the studied group of patients with odontogenic and non-odontogenic head and neck abscesses.

Formulated in this way, the tasks enable the objective to be demonstrated.

6. Patient selection and study design:

81 patients with head and neck abscesses were studied, 50 of them with odontogenic and 31 with non-odontogenic origin. PCT, CRP, WBC and Neutr values were examined and DNI was calculated.

Included in the study were 48 men (59.26%) and 33 women (40.74%). The average age is 43 years (42 years for men and 44 years for women).

One 80-year-old female patient with non-odontogenic abscess was excluded from the study. The cause is a newly discovered oncohematological disease.

The remaining 80 patients were divided into two groups – 50 with odontogenic abscesses and 30 with non-odontogenic abscesses.

Their gender distribution is 48 (60%) men and 32 (40%) women. Patients had a mean age of 43 years (42 years for men and 44 years for women), ranging from 18 to 84 years.

As a control group, 51 healthy individuals (30 men and 21 women) of age and gender corresponding to the study group were used.

Patients who met the following inclusion criteria were examined in the study:

- presence of an abscess or phlegmon in the region of the head and neck, proven during the clinical examination and/or through additional imaging studies such as echography, nuclear magnetic resonance and computed tomography;
- confirmation of the clinical diagnosis during the surgical intervention (incision, lavage, drainage), during which the available exudate is evacuated;
- age over 18 years;
- confirmation by patients that they wish to participate in the study, evidenced by signing an informed consent to this effect.

The study did not examine patients with a history of the following diseases and conditions in which false positive results are possible. Exclusion criteria were defined:

- multiple injuries;
- recent major surgical interventions;
- burns;
- cardiogenic shock;
- overheating;
- parasitic infections;
- systemic mycotic infection;
- known lung carcinoma (including small cell);

- known carcinoma of the thyroid gland (including medullary C-cell carcinoma);
- known tumors with paraneoplastic hormone production;
- prolonged severe organ hypoperfusion;
- conditions and diseases reducing the content of oxygen in the tissues - bronchial asthma, pneumonia, etc.;
- intake of medications that stimulate the release of cytokines.

The number of logical units required for a clinical study for a doctoral dissertation is sufficient for the formulation of conclusions of good scientific value.

Clinical, paraclinical, medical and statistical research methods were used in the dissertation work. Through the chosen methods, the goal was achieved and an answer to the set tasks was obtained.

The clinical material selected for the implementation of the set main goal and tasks is completely sufficient for the development of a dissertation work.

The units of observation are:

- patients with odontogenic abscesses;
- patients with non-odontogenic abscesses;
- a control group of healthy individuals.

The signs of observation are:

- gender differences regarding the studied markers PCT, DNI, CRP, WBC and neutrophils in the studied population;
- correlations between PCT, DNI, CRP, WBC and neutrophils in the studied group of patients with head and neck abscesses, in the subgroups with odontogenic and non-odontogenic abscesses and in the control group of healthy people;

- cut-off diagnostic and reference values of PCT and DNI in the studied groups of patients with odontogenic and non-odontogenic head and neck abscesses;
- sensitivity, specificity and predictability of PCT, DNI, CRP, WBC and neutrophils in the studied group of patients with odontogenic and non-odontogenic head and neck abscesses.

Observation time:

The patients were examined and treated in the period from June 2021 to December 2021.

The places of observation: Clinic of Maxillofacial Surgery at University General Hospital (UGH) "Sveta Marina" – Varna city and the Central clinical laboratory at the same medical institution.

7. Statistical methods for the data analysis:

Column graphics with clusters, pie charts with individual segments in 3D and SmartArt graphics of the software program "Microsoft Word" (2010) were used to present the results.

For the purposes of this study, the following statistical methods were applied:

- In the descriptive statistics of the studied groups and the control group, frequency tables with absolute numbers and percentages, arithmetic mean, median, standard deviation, minimum and maximum values were used. Graphically, mean measurements are presented with box-plot graphics and categorical data with bar graphics. "Jamovi software 2.2.0" was used to visualize the obtained data.
- In the intergroup comparison of the differences in the studied indicators between the individual studied groups (with odontogenic abscesses, with non-odontogenic abscesses and control group), variation analysis "ANOVA" was used. In them, the graphical visualization of the compared numerical values of the investigated markers is presented with "Error Bar Graphics". "Post-Hoc" analysis using Tukey and Games-Howell methods was used to check between which of the studied groups there were statistically significant differences.

- For the intragroup comparison of the differences between the studied indicators, a parametric "Independent T-test" was used for symmetrically distributed data and its corresponding non-parametric "Mann-Whitney U-test" for asymmetrically distributed data. Graphically, the differences in mean measurements of the studied markers are presented with "Error Bar Graphs".

- ROC curves were used to determine the predictive accuracy of the indicators we investigated in the study (CRP, WBC, Neutr, PCT and DNI). Graphically, they show the sensitivity and specificity of these markers. The predictability of these indicators is determined by their area under the curve (AUC - area under the curve) and 95% confidence interval (95% CI - confidence interval). Using rock curves, cut-off values of the same markers were determined.

- Correlation analysis was used to study the dependencies between the studied clinical indicators and to establish the strength of their influence on each other. Pearson's coefficient (r) was used to determine the strength of correlation between these indicators.

All differences between the values at the $p < 0.05$ value accepted for biological experiments were considered statistically significant (reliable).

All tables were created on the software program "Microsoft Word" (2010).

8. Characterization and evaluation of the dissertation work:

The dissertation contains a total of 104 pages and is illustrated with 49 graphics, 43 tables and 2 diagrams. The bibliographic reference includes 135 literary sources, 2 of them in Cyrillic and 133 in Latin alphabet. The numbers of the graphs in the abstract do not correspond to those in the dissertation.

The results are comprehensively and correctly described, analyzed and interpreted.

The dissertation concludes with conclusions based on the results, discussion and generalizations made from the study.

9. Contributions and significance of the development for science and practice

The following contributions are outlined in the dissertation:

1. For the first time in Bulgaria, PCT and DNI are used as markers in the management of odontogenic and non-odontogenic head and neck abscesses;

2. For the first time in Bulgaria, a correlation between PCT, DNI, CRP, WBC and neutrophils as markers of inflammation in odontogenic and non-odontogenic head and neck abscesses was made;

3. Determination and follow-up of PCT and DNI in routine practice in patients suspected of odontogenic and non-odontogenic head and neck abscesses may lead to earlier diagnosis and more precise treatment of these diseases;

4. The introduction of DNI as a diagnostic marker of odontogenic and non-odontogenic head and neck abscesses has positive economic dimensions;

5. PCT and DNI cut-off values for the diagnosis of odontogenic and non-odontogenic head and neck abscesses were derived.

10. Evaluation of publications on the dissertation work

Four scientific papers are presented, of which two articles were published in scientific publications and two reports were presented at scientific forums, with Yanko Yankov, MD being the first author in all four of them, namely:

Scientific publications:

1) Y. Yankov, Y. Bocheva. The Value of Delta Neutrophil Index in The Odontogenic and Non-odontogenic Abscesses' Surgery of Head and Neck. Varna Medical Forum. 2022;

2) Y. Yankov, Y. Bocheva. Procalcitonin and C-reactive Protein in The Inflammatory Surgery of Head and Neck - Diagnostic Significance and Correlations. Varna Medical Forum. 2022.

Participation in scientific forums:

1) Y. Yankov, Y. Bocheva, N. Kolev. Evaluation of Both Procalcitonin and Delta Neutrophil Index in Hospitalized Patients with Odontogenic and Non-odontogenic Abscesses. Clin Chem Lab Med 2021; 59. Special Suppl: pp S94 – S998. Nov/Dec 2021;

2) Y. Yankov, Y. Bocheva. Correlation between the serum levels of procalcitonin and CRP in patients with odontogenic and non-odontogenic abscesses in maxillofacial surgery.

Clinica Chimica Acta (530): S212-S213.

They have been published in prestigious journals, and the results obtained in them satisfy quantitatively and qualitatively the legal requirements for a dissertation work.

11. Personal participation of the doctoral student

I consider the researches and observations of patients and the resulting conclusions and contributions in the dissertation to be the personal work of the author.

12. Abstract

The abstract contains 71 pages, illustrated with 48 graphs and 43 tables. It correctly reflects the essence of the research and the results achieved in the dissertation. The conclusions drawn are reflected in it.

It was prepared in accordance with the requirements of the RSASR and the regulations of the Ministry of Education – Varna city. Reflects the content of the dissertation work.

13. Critical remarks and recommendations

The set of materials from the dissertation provided to me are complete and in accordance with the Law of the development of the academic staff and the rules for its implementation, as well as with the Rules of the Medical university – Varna city. I have no comments or recommendations.

14. Personal impressions

Yanko Yankov, MD is built as a teacher and doctor of medicine with a marked clinical thinking. Possesses in-depth theoretical knowledge and professional skills in the specialty. The text provided to me shows a thorough knowledge of the problem and gives me reason to take it as her personal case.

15. Recommendations for future use of dissertation contributions and results

I recommend Yanko Yankov, MD to continue his scientific research on the topic by expanding his search for more new scientific contributions.

CONCLUSION

The dissertation submitted to me for review by Yanko Georgiev Yankov, MD - a PhD student of independent study on the topic "Procalcitonin and delta neutrophil index levels in the surgery of head and neck inflammatory diseases" I take it as complete. Meets the requirements of the Higher Education Institutions, the Law of higher education, the Law of the development of the academic staff, the Regulations for the implementation of the Law of the development of the academic staff and the Regulations of the Medical university – Varna city.

The topic of the work is current and well chosen.

The literature review is comprehensive and gives a clear idea of the current state of the problem under consideration. It concludes with a critical analysis that is a good foundation for the research conducted.

Based on an analysis of scientific research carried out so far, the purpose of the scientific development has been deduced. The assigned tasks give an opportunity to prove it.

The clinical material and the studies carried out are of interest to medical science and practice. The obtained results in the dissertation work, their interpretation, as well as the presented publications related to it, I consider being the personal work of the author.

The dissertation shows that the doctoral student Yanko Yankov, MD possesses in-depth theoretical knowledge and professional skills in the specialty, demonstrating qualities and skills for independent conduct of scientific research.

Based on everything noted here, I accept that the requirements of the Law of the Development of the Academic Staff in the Republic of Bulgaria, the Regulations for the Implementation of Law of the development of the academic staff and the relevant Regulations of the Medical University – Varna city have been met. The presented materials and dissertation results **fully** correspond to the specific requirements of the Medical University – Varna city

In conclusion: I confidently give a positive assessment of the dissertation work on the topic **"Procalcitonin and delta neutrophil index levels in the surgery of head and neck inflammatory diseases"** and I will vote **"YES"** for awarding the scientific and educational degree **"Doctor"** in a scientific specialty **"Surgery"** by **Yanko Georgiev Yankov, MD**.

07.11.2022

Reviewer:

(Prof. Rosen Gospodinov Kolarov, DMD, PhD)

