To the Chairman of the Scientific Jury
determined by order № P-109-85 / 11.03.2021
of the Rector of medical University Varna

REVIEW

By **Assoc. Prof. Dr. Rosen Kolarov, PhD**
Medical University of Varna, Faculty of Dental Medicine, Department of Oral Surgery
of a dissertation for the award of the educational and scientific degree "Doctor"

Field of higher education: 7. Health and sports
Professional field: 7.2. Dentistry
PhD program: "Oral surgery"

Author: Dr. Rosen Bozhidarov Tsolov,
Self study PhD student

Department: Oral Surgery

Subject:

"Treatment of medically-induced osteonecrosis of the jaws with the use of platelet-rich fibrin PRF"

Supervisor: Prof. Tihomir Dobrinov Georgiev, MD, PhD, Medical University - Varna, SPF, Department of Oral Surgery

1. General presentation of the procedure and the PhD student

This review is prepared on the basis of Order of the Rector of MU - Varna № P-109-85/11.03.2021. with an appointed Scientific Jury under a procedure for public defense of the described dissertation. The presented set of materials on paper and electronic media are in accordance with Art. 44 (3) of the Regulations for development of the academic staff in MU - Varna. They were provided to me within the statutory period.

The doctoral student has attached 3 publications related to the topic of the dissertation.

All documents are prepared and presented properly.

2. Brief biographical data about the doctoral student

Dr. Rosen Bozhidarov Tsolov was born on April 21, 1980.

In 2005 he completed a six-year training course in Dentistry at the Faculty of Dentistry of the Medical University of Plovdiv.

Since 2006 Dr. Rosen Tsolov has been working at the Clinic of Oral and Maxillofacial Surgery at St. George University Hospital – Plovdiv

Dr. Tsolov has a total work experience of 17 years. In 2012 he acquired the specialty "Oral Surgery". In 2016 he obtained a Master's degree in Public Health and Health Management.

Dr. Tsolov's latest attestation grade is "good." Dr. Tsolov is a member of the Bulgarian Dental Association (BDA) and the Bulgarian Scientific Dental Society (BNSD)

level of English language proficiency – good

3. Relevance on the subject and the expediency of the set goals and objectives

The topic of the dissertation deals with a current and important problem for dental science and practice. The goal is clearly formulated, the tasks are defined correctly and are performed with modern research methods.

4. Understanding of the problem

In his dissertation, Rosen Bozhidarov Tsolov shows in-depth knowledge of the topic, namely: "Treatment of drug-induced osteonecrosis of the jaws with the use of platelet-rich fibrin PRF."

He made an in-depth critical analysis of the literature, formulating unresolved issues on the topic, namely:

- The comparison with invasive versus non-invasive treatment in patients with induced osteonecrosis of the jaws;
- pain control in patients with induced osteonecrosis of the jaws;
- dealing with infection in patients with induced osteonecrosis of the jaws;
- stabilizing the progression of the disease in patients with induced osteonecrosis of the jaws;
- methods for closing the exposed bone in patients with induced osteonecrosis of the jaws;
- the choice of treatment with PRF, as the most optimal surgical procedure in patients with induced osteonecrosis of the jaws.

Dr. Rosen Bozhidarov Kolov shows skills for independent research.

Drug-induced osteonecrosis of the jaw is a disease characterized by an open necrotized bone that does not heal within 8 weeks of diagnosis, despite medical care.

These are mostly patients with cancer. They underwent long-term intravenous treatment with bisphosphonates as part of complex therapy. These drugs are metabolized intracellularly to ATP analogs. The intracellular accumulation of these metabolites in osteoclasts inhibits their function and induces apoptosis, most likely by inhibition of ATP-dependent enzymes.

Currently, the treatment of patients with drug-induced osteonecrosis of the jaw remains a problem. No effective treatment has been developed for the temporary cessation of bisphosphonates and no short-term benefits have been reported. On the other hand, long-term discontinuation (if systemic conditions allow) may be helpful in stabilizing osteonecrosis sites as well as reducing clinical symptoms.

The frequency of drug-induced osteonecrosis of the jaw worldwide is increasing, along with the increasing use of bisphosphonates. All this requires in-depth research and development of behavioral algorithms in these patients.

This enabled Dr. Tsolov to formulate clearly and precisely the purpose of his study, namely: "To make a comparative assessment of the results of conservative antibiotic treatment and surgical treatment with a platelet-rich fibrin membrane (PRFm) of drug-induced bone necrosis of the jaws".

The exposition is formulated in good scientific writing.

5. Research methodology

The goal is accomplishment through the implementation of four main tasks, namely:

Task 1: Study of the frequency of drug-induced bone necrosis of the jaws in Bulgaria.

Task 2: Study of the peculiarities of the radiological symptoms in bisphosphonate-induced osteonecrosis of the jaws.

Task 3: Comparative radiological evaluation of the results obtained after the treatment of patients with drug-induced bone necrosis of the jaws, conservatively (medically) and surgically with the help of PRF.

Task 4: Comparative clinical evaluation of the results obtained after the treatment of patients with drug-induced bone necrosis of the jaws, conservatively (medically) and surgically with the help of PRF.

The clinical material examined on the tasks of the dissertation is as follows:

By task 1.

The study of the frequency of drug-induced bone necrosis of the jaws in Bulgaria and the methods used for treatment are according to data from the National Health Insurance Fund (NHIF). The data are presented in spreadsheets in Excel, which show the number of patients with drug-induced osteonecrosis of the jaw by years in the period 2015-2018, as well as the specific drugs with which the patients were treated.

By task 2, 3 and 4.

The study included a total of 237 people who passed through the Clinic of Oral and Maxillofacial Surgery of MHAT "St. George" - Plovdiv in the period 2013-2017. Of these 237 people, 130 are women and 107 are men.

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

Units of observation: patients with drug-induced bone necrosis of the jaws.

The monitoring was performed in the selection of patients with the following criteria, namely:

Including criteria

- Established bisphosphonate osteonecrosis.
- Patients must be at least 18 years old.

Excluding criteria

- Presence of immunodeficiency.
- Patients taking anticoagulants and antiplatelets.
- Patients on steroid therapy for more than 10 days.
- Pregnant women with contraindications for surgical interventions.
- Presence of allergy to medications used during treatment.
- Presence of ulcerative colitis and enteritis.
- Identified diseases of the liver and kidneys.
- Patients smoking more than 10 cigarettes a day or taking drugs.

Monitoring time:

The study was conducted in the period 2013-2017.

The place of observation is the Clinic of Oral and Maxillofacial Surgery of UMHAT "St. George" - Plovdiv.

The following methods of research and analysis of the obtained data were used in the implementation of the set tasks:

Statistical methods for data analysis:

The data were processed using the statistical programs IBM SPSS, version 25 (2017), Minitab 18 (2017). The significance level is considered to be $p \leq 0.05$.

Used:

- Independent samples t-test for static comparison between two separate groups of patients or two types of treatment
- Paired samples t-test for related samples.
- Chi-square test, and for smaller samples it is applied
- Fisher's test.
- Alpha error tolerance test = 5% ($p < 0.05$),
- Independent samples-t-test.
- Accurate Fisher's test or Chi-Square test when analyzing the relationship between two qualitative variables

6. Characteristics and evaluation of the dissertation

The dissertation is written on 179 pages, illustrated by 16 tables, 61 figures, 20 tables and 5 appendices. The bibliography includes 200 sources, of which 14 in Cyrillic and 286 in Latin. It contains all the necessary sections for a dissertation.

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

The dissertation ends with conclusions based on the results, discussion and summaries of the study.

Final conclusions from the dissertation:

1. The frequency of drug-induced osteonecrosis of the jaw in Bulgaria in the period 2015-2018 has increased annually to 8.5%, which is in accordance with the norms established in other studies and adopted by the American Association of Dentists and Maxillofacial Surgeons.
2. There is a strong trend of statistical significance of drug-induced osteonecrosis of the jaw and ibandronic acid treatment. 92% of all cases were due to treatment with ibandronic acid.
3. PRF shows much better results in terms of bone density, bundle and lingual width than conservative treatment in the long term. In the process of time, it has significantly higher values in the PRF group, where there is a gradual increase, with the highest value reported on the 90th day. In the conservative treatment group, the mean bone density decreased and the lowest value was observed on the 90th day. Compared to the

conservative treatment group, the buccal width in the PRF group decreased on day 90. Lingual width has significantly higher values in the PRF group and changes differently in both treatments. In PRF treatment, a decrease in width was observed between day 45 and day 90, and in conservative treatment there was an increase between the two time points.

4. In the 6th month there are no statistically significant differences in the anatomical and topographic localization of the two types of treatment. There are some differences in size. With PRF treatment, a higher percentage of patients with a size of ≤ 1 mm was observed. The majority of the PRF group are in the first two size categories: ≤ 1mm and> 1 - 3mm. The majority of patients on conservative treatment are > 1 - 3 mm and> 3 - 5 mm in size.
5. PRF treatment showed much better overall long-term results in a follow-up computed tomography 6 months after compared to the conservative treatment. PRF treatment is characterized by a significantly higher percentage of patients with strong shadow intensity, while conservative treatment is dominated by a percentage with low intensity.
6. Comparing the two types of treatment in terms of homogeneity and contours, patients on PRF treatment have significantly better results after 6 months. In PRF treatment, homogeneity and sequestration predominate, while in conservative treatment, the percentage of homogeneity and sequestration is lower and inhomogeneity is present. In patients on PRF treatment, sharp and continuous outlines predominate, while in patients on conservative treatment, blurred and interrupted outlines are more common.
7. None of the patients on the PRF treatment group showed any change in the surrounding bone, in contrast to the conservative treatment group.
8. PRF shows good results in terms of healing: redness, as on the 90th day, it occurs faster and is statistically greater than that of the group with conservative treatment; healthy granulation tissue appears on the 7th day; the development of signs of epithelialization in the PRF group was rapid and by day 14 all patients showed signs of epithelialization. In the conservative group it is slower and more gradual and does not reach 100%.
9. Treatment of medically-induced osteonecrosis of the jaws with a platelet-rich fibrin membrane shows reliable results in both short and long term when it comes to healing of both soft and hard tissues. It shows much better results in epithelialization and bone density, as well as overall healing, compared to conservative treatment. It could be used as a modern alternative to drug treatment.

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

The dissertation outlines the following contributions of original and confirmatory nature, as follows:

1. For the first time in our country a study of the frequency of medically-induced osteonecrosis of the jaws and the bisphosphonates used for the treatment of patients is done.

2. In the present study, for the first time in our country, a membrane of platelet-rich fibrin (PRFm) is used for the treatment of medically-induced osteonecrosis of the jaws.

3. For the first time in our country a comparative clinical evaluation of the healing ability of the membrane of platelet-rich fibrin (PRFm) is made, in comparison with the conservative method of treatment of medically-induced osteonecrosis of the jaws.

4. For the first time in our country a comparative radiological assessment of the possibilities for bone density and regeneration of the membrane from platelet-rich fibrin (PRFm) is made, in comparison with the conservative method of treatment of medically-induced osteonecrosis of the jaws.

5. For the first time in our country a comparative computer-topographic study is performed for the possibilities of complete long-term healing of the membrane from platelet-rich fibrin (PRFm), in comparison with the conservative method of treatment of medically-induced osteonecrosis of the jaws.

The scientific and scientific-applied achievements in the dissertation are based on the results of the research, the conclusions made and the subsequent discussion. I believe that the results formed in this way shed light on a problem that is important for medical science. In addition, the main achievements in the work are important for scientific and applied research in this direction.

8. Evaluation of the publications on the dissertation

Three publications related to the dissertation are presented, and Dr. Gospodinov is the first author in all three publications, namely:

1. R Tsolov, G Yordanov, Case of treatment and follow-up of drug-induced osteonecrosis of the jaws with the use of PRF, Rare diseases and orphan drugs 11 (4)

2. R Tsolov, G Yordanov, STAGES AND PATHOPHYSIOLOGICAL MECHANISMS OF ACTION OF MRONJ, Knowledge International Journal 43 (4), 677-679

3. R Tsolov, G Yordanov, Use of platelet-rich plasma in the treatment of medication-related osteonecrosis of the jaw. A case report, Varna Medical Forum 9 (2)

They are published in prestigious magazines, two of them in Bulgarian and one in English. The results obtained in them satisfy quantitatively and qualitatively the legal requirements for dissertation work.

9. Personal participation of the doctoral student

I accept the conducted research and observations of patients and the resulting conclusions and contributions in the dissertation as a personal work of the author.

10. Abstract

The abstract contains 82 pages, illustrated with 61 figures and 20 tables. It correctly reflects the essence of the research and the results achieved in the dissertation. The conclusions are reflected in it.

It has been prepared in accordance with the requirements of the Law for the Development of the Academic Staff in the Republic of Bulgaria and the legislation of MU - Varna. It reflects the content of the dissertation.

11. Critical remarks and recommendations

The set of materials from the dissertation provided to me is complete and in accordance with the Regulations for its application, as well as with the Regulations of MU - Varna. I have no remarks or recommendations.

12. Personal impressions

I have no personal impressions of the author of the dissertation. The text presented to me shows thorough knowledge of the problem and gives grounds for accepting it as his personal work.

13. Recommendations for future use of dissertation contributions and results

I recommend Dr. Tsolov form and publish his dissertation as a monograph. This would make his work more accessible to colleagues who work on this issue on a daily basis.

CONCLUSION

The topic chosen by Dr. Rosen Tsolov for the dissertation **"Treatment of drug-induced osteonecrosis of the jaws with the use of platelet-rich fibrin PRF"** treats a current problem in dental science and practice. In this sense, the topic is relevant and well chosen.

The literature review is comprehensive and gives a clear idea of the current state of the problem. It ends with a critical analysis, which is a good basis for the research.

The clinical material and the studies performed are of interest to dental science and practice. The results obtained in the dissertation, their interpretation, as well as the presented publications related to it, I accept as a personal work of the author.

The dissertation shows that the doctoral student Dr. Tsolov **has in-depth theoretical knowledge** and professional skills in the specialty of Oral Surgery by **demonstrating** qualities and skills for independent research.

Based on everything noted here, I acknowledge that the requirements of the Law for the Development of the Academic Staff in the Republic of Bulgaria, the Regulations for implementation of the LDASRB and the respective Rules of MU - Varna are met. The presented materials and dissertation results **fully** comply with the specific requirements of MU - Varna.

In conclusion: I confidently give a positive assessment of the dissertation on "**Treatment of drug-induced osteonecrosis of the jaws with the use of platelet-rich fibrin PRF"** and I will vote with "**YES**" for awarding the scientific and educational degree "**Doctor**" in the scientific specialty "**Oral Surgery**" to **Dr. Rosen Bozhidarov Tsolov**.

05.04. 2021 г.

Reviewer: .....

(Prof. Dr. Rosen Gospodinov Kolarov, PhD)