To: Associate Professor Dr. Sonya Galcheva,
MD, PhD, University Hospital "Sv. Marina", Varna

Faculty of Medicine, Medical University-Varna

Chair of the scientific jury, appointed by an order of the Rector of the Medical University – Varna, based on the decision of the Departmental Council of the Department of Pediatrics at Medical University "Prof. Dr. Paraskev Stoyanov" – Varna on 26.10.2024.

Reviewer: Associate Professor Dr. Nartsis Nissim Kaleva-Hodzheva,

specialist in pediatrics and pediatric endocrinology, former head of the Endocrinology Unit at the Pediatrics Clinic, University Hospital "St. George" – Plovdiv until the end of 2021, and Associate Professor at the Department of Pediatrics and Medical Genetics, Faculty of Medicine, Medical University – Plovdiv

on the dissertation for the award of the educational and scientific degree "Doctor,"

Professional field 7.1 Medicine, Higher Education Field 7. Healthcare and Sport,

Doctoral Program in Pediatrics

Author: Dr. Nikolinka Yordanova Peykova

Form of Doctoral Study: Full-time

Department: "Pediatrics," Faculty of Medicine, Medical University – Varna

Title: **"Metabolic Risk and Body Composition in Children Born Small for Gestational Age Due to Genetic Syndromes (Prader-Willi and Silver-Russell and Others)"**

Scientific Supervisor: Professor Dr. Violeta M. Iotova, MD, PhD, DSc

1. General Overview of the Procedure and Doctoral Candidate

The submitted set of materials, both in paper and electronic format, complies with the Regulations of the Medical University – Varna for obtaining the educational and scientific degree "Doctor" and the scientific degree "Doctor of Science" in the Faculty of Medicine at MU-Varna. It includes the following documents:

1. Dissertation – original form in Word and PDF format
2. Abstract – original form in Word and PDF format

3. Curriculum vitae in European format
4. Diploma for the educational-qualification degree "Master" with an appendix
5. Copy of the specialist certificate
6. List of publications and scientific papers on the topic
7. Declaration of originality
8. Enrollment order in the doctoral program
9. Dismissal order with the right to defend
10. Transcript from the doctoral minimum exam in the specialty
11. Information cards from the National Center for Information and Documentation (NACID)
12. Declaration of authorship consent, in compliance with copyright laws
13. Application to the Rector to initiate the defense procedure

The presented documents meet the requirements of the Medical University – Faculty of Medicine – Varna.

1. Biographical Data and Procedural Requirements.

Dr. Nikolinka Yordanova was born on June 1, 1990, in Yambol, where she completed her secondary education in 2009 at Atanas Radev Mathematical High School. She graduated in Medicine from the Medical University – Varna in 2015. In March 2016, she began her specialization in pediatric endocrinology and metabolic diseases at the First Pediatric Clinic, University Hospital "St. Marina," Varna. In 2019, she won a competitive selection for a full-time Ph.D. position at the Department of Pediatrics at MU – Varna, with a dissertation on "Metabolic Risk and Body Composition in Children Born Small for Gestational Age Due to Genetic Syndromes (Prader-Willi, Silver-Russell, and others)." In 2020, she successfully obtained a specialization in pediatric endocrinology and metabolic diseases.

Dr. Yordanova's clinical and research interests are primarily focused on rare endocrine diseases. She leads a multidisciplinary team for the care of patients with Prader-Willi and Silver-Russell syndromes at the Expert Center for Rare Endocrine Diseases, part of the European Reference Network for Rare Endocrine Diseases (Endo-ERN). Since 2020, she has been an Honorary Assistant at the Department of Pediatrics at MU – Varna. In 2022, she completed a one-month specialization at the Erasmus MC Sophia Children's Hospital in Rotterdam, the Netherlands, focusing on specialized care for patients with rare imprinting syndromes, funded by the Endo-ERN. Dr. Yordanova has received three grants from the International Prader-Willi Syndrome Organization (IPWSO) to attend international conferences in Europe and the America. She is a member of the BPA, BNESPE, VAPED, and ESPE.

Dr. Yordanova has submitted a dissertation that includes the standard sections as required: Literature Review; Objectives; Materials and Methods; Results; Discussion; Conclusions; Contributions; Appendices; and References. The dissertation comprises 158 pages and is illustrated with 30 tables and 8 figures. The bibliography consists of 382 sources, 11 in Cyrillic and 371 in Latin script.

Her publication record and participation in scientific forums meet the requirements for defense as stipulated by the Medical University – Varna regulations. The dissertation work has resulted in two scientific publications, where Dr. Yordanova is the leading author of one, as well as two scientific presentations at the annual ESPE congresses in Athens and Rome.

In summary, the structure of the dissertation and Dr. Yordanova's independent role in its development are in full compliance with procedural requirements.

2. Relevance of the Topic

The dissertation is dedicated to two rare diseases—Prader-Willi Syndrome (PWS) and Silver-Russell Syndrome (SRS)—in the diagnosis and treatment of which Dr. Yordanova is actively involved as the head of the multidisciplinary team at the Expert Center for Rare Endocrine Diseases in Varna. She identifies unifying characteristics between these two disorders, including genomic imprinting inheritance, low birth weight, and early metabolic complications. In the literature review, she thoroughly discusses the fetal hypothesis, highlighting how low birth weight correlates with an increased risk of cardiovascular diseases, hypertension, dyslipidemia, obesity, insulin resistance, and type 2 diabetes.

Moreover, both PWS and SRS patients exhibit shorter stature and a lower percentage of muscle mass compared to healthy controls, providing a rationale for early initiation of recombinant growth hormone therapy in nearly all patients. Since there is insufficient data on the relationship between this treatment, muscle mass, and the extent of metabolic risk, Dr. Yordanova raises these controversial questions as part of the aims of her dissertation.

In recent years, the improvement of clinical care and knowledge in the field of rare diseases has become an increasingly interesting focus within contemporary science and practice. Furthermore, there has been a growing emphasis on studying and enhancing the quality of life of patients and devising ways to improve it. All of this underlines that the topic is relevant and that its selection reflects both the professionalism of Dr. Yordanova and her scientific supervisor.

3. Understanding of the Problem

The dissertation is written in clear Bulgarian, with a thorough and well-supported literature review, including the author's own publications, justifying the choice of topic. The number of patients and control groups is sufficient for robust statistical analysis. The logical conclusions, aligned with the objectives and derived from an in-depth statistical evaluation, demonstrate the candidate's excellent understanding of the problem.

4. Research Methodology

The clinical, laboratory, and statistical methods presented in the dissertation are appropriate to the objectives and tasks of the study, effectively supporting the author's thesis.

5. Characteristics and Evaluation of the Dissertation and Contributions

The dissertation is 158 standard typed pages, with 37 pages dedicated to the literature review, while the remaining sections discuss the objectives, tasks, methods, results, discussion, conclusions aligned with the objectives, contributions, appendices, and references. The literature review is purposefully structured, with focus on relevant aspects of the primary topic. Given the differences in issues posed by the two syndromes, the review is divided to address Prader-Willi Syndrome (PWS) and Silver-Russell Syndrome (SRS) separately.

The review provides a detailed presentation of clinical characteristics, inheritance patterns, nutritional features, hormonal abnormalities—including parameters such as leptin, adiponectin, and irisin associated with hyperphagia in PWS—thyroid function, and levels of gonadotropic, sex, and adrenal hormones. A special focus is given to the short stature observed in these patients and the rationale for growth hormone (GH) therapy, as well as the positive effects of this treatment on various clinical, metabolic, and psychological parameters. The review also devotes significant attention to obesity as a

leading pathological symptom in PWS and discusses opposing data regarding the presence of metabolic syndrome, examining these questions as part of the dissertation's objectives and the degree of metabolic risk in these patients.

In the SRS section, the review addresses unique conditions such as hypoglycemia, short stature, early puberty, and treatment with GnRH. Additionally, the method of Dual-energy X-ray Absorptiometry (DXA) is presented as a key measure for assessing body composition and the balance between fat, muscle, and bone tissue. The review also discusses bone health, which is further examined when presenting the patients and healthy controls as part of the overall metabolic profile.

The transition between the literature review and the original research material is highlighted by the persuasive and logical presentation of the rationale for the choice of topic. The objective is precisely formulated—to assess cardiovascular and metabolic risk in patients with PWS and SRS based on their fat and muscle mass—supported by four specific tasks designed to fulfill this objective.

Materials and Methods Used: The number of patients included in the study is sufficient. Given the examination of two distinct diseases, the research utilizes a "case-control" study design with two parallel cross-sectional substudies: 25 patients with genetically confirmed PWS, 18 with genetically confirmed or clinically diagnosed SRS, and 39 healthy controls matched by sex, age, and partially by BMI. Considering the rarity of both diseases, the number of included patients is sufficient to allow statistically valid conclusions. The selection of an obese control group rather than healthy controls for the PWS patients is logical and appropriate, although this choice partially explains the lack of statistically significant differences in some of the metabolic indicators studied.

Contributions of Dr. N. Yordanova's Dissertation

1. The dissertation examines a variety of parameters, some of which are novel for Bulgaria, as part of the assessment of the clinical and laboratory profile and body composition of significant groups of patients with Prader-Willi Syndrome (PWS) and Silver-Russell Syndrome (SRS). An example of this is the analysis of irisin levels in PWS and SRS patients and the correlations with metabolic indicators and body composition.

2. This study is the first in the available global literature to establish a link between increased abdominal circumference in both patient groups—PWS and SRS—and reduced levels of high-molecular-weight adiponectin in PWS, associated with a higher risk of arterial hypertension.

3. For the first time in Bulgaria, the study defines the frequency of metabolic and cardiovascular disorders in patients with PWS and SRS compared to gender-, age-, and partially BMI-matched healthy controls.

4. A correlation between leptin and insulin and HOMA-IR in PWS and SRS patients is identified, establishing leptin as an early indicator for the development of insulin resistance.

5. For both syndromes, delays in neuropsychological development are specific, and quality of life is lower than in healthy children. The presented assessment of quality of life in PWS and SRS patients, and the impact of growth hormone treatment (with IGF-I level used as an indicator), offers valuable guidance for future therapeutic strategies by a broad spectrum of healthcare providers.

6. From a practical standpoint, the inclusion of questionnaires, surveys, and quality of life tests in the "Appendices" section holds potential for application in routine practice and in other healthcare facilities.

These contributions are summarized in the doctoral candidate's conclusion: The results of this dissertation, both on their own and in affirmation of established patterns in the global literature, highlight the need for systematic and detailed assessment of anthropometric parameters, physical status, metabolic and hormonal profiles, body composition, and quality of life in clinical practice for these two rare endocrine disorders. This is essential for preventing obesity and its metabolic complications among PWS patients and for the early identification of risks in high-risk SRS patients.

6. Evaluation of Publications and Personal Contribution of the Doctoral Candidate

The presented publications and participation in scientific forums are directly related to the dissertation topic, with Dr. Nikolinka Yordanova being the lead author in most cases. The number of publications meets procedural requirements. While a dissertation typically reaches a limited readership and the dissertation summary is available to a slightly broader audience, scientific journal publications serve as a vital means of sharing the literature review and original findings with a wider audience, underscoring their value beyond purely academic merit. A future monograph covering rare diseases—such as Prader-Willi and Silver-Russell syndromes, including clinical presentation, metabolic features, treatment, and quality of life—would be a valuable resource for a broad range of clinicians, including pediatricians, pediatric endocrinologists, cardiologists, geneticists, and general practitioners.

At this stage, I have no further remarks.

7. Dissertation summary

The abstract is 100 pages long and effectively presents all original results—both verbally and through tables and charts—with detailed discussion that includes much of the literature review data, enabling logical interpretation of the author's results and conclusions. I have no further remarks regarding its content. It adequately corresponds to the dissertation and, even as a standalone document, provides sufficient information for readers regarding the results and references cited.

The abstract meets the requirements of the Medical University – Varna.

8. Conclusion

The dissertation contains scientific, applied-scientific, and practical results that constitute an original contribution to the field and meet all requirements of the Bulgarian Law on the Development of Academic Staff (ZRASRB), the Regulations for the Application of the ZRASRB, and the relevant regulations of the Medical University – Varna.

The dissertation demonstrates that the doctoral candidate possesses in-depth theoretical knowledge and professional skills in the field of pediatric endocrinology and metabolic diseases, showcasing the qualities and competencies necessary for independent scientific research.

For the reasons outlined above, I confidently provide a positive evaluation of the research presented in the dissertation, its summary, and the achieved results and contributions, and I recommend that the esteemed Scientific Jury award the educational and scientific degree of 'Doctor' to Dr. Nikolinka Yordanova.

12.11.2024

City of Plovdiv

Reviewer:

Associate Professor Dr. Narcis N. Kaleva-Khojeva, PhD