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The Journal of Pediatric Research

Original Articles

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Chest Wall Tumors
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Surgical Factors in Megameatus Intact Prepuce
Bağcı et al.

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Editorial

Dear Readers of The Journal of Pediatric Research,

We are delighted to present to you the second issue of 2024. Within this edition, you will find a collection of eight original articles, each offering valuable insights. Topics explored in this edition include, "Position of the Newborn During Transition", "Significant Risk Factors for Children Obesity", "Pediatric Ketoacidosis : A Turkish PICU", "Olive Oil Supplement and Preterm Infants", "Phagocyte Functions of Cystic Fibrosis Patients", as well as three topics from pediatric surgery: "Chest Wall Tumors", "Advanced Thoracic Pediatric Robotic Surgery", and "Surgical Factors in Megameatus Intact Prepuce".

We would like to highlight that The Journal of Pediatric Research is indexed in Emerging Sources Citation Index (ESCI), Embase, Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, CABI, Gale/Cengage Learning, Ulakbim TR Dizin, TurkMedline, J-GATE IdealOnline, Hinari, GOALI, ARDI, OARE, AGORA and Türkiye Citation Index.

We express our gratitude to the authors, referees, editorial team, and Galenos Publishing House for their valuable support in assembling this edition. We eagerly anticipate your continued contributions to our forthcoming publications.

Sincerely,

Assoc. Prof. Dr. Gulhadiye Avcu

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Original Article

Examining the Potential of Advanced Robotic-Assisted Thoracic Surgery in Pediatric Cases

Ülgen Çeltik[1], Cengiz Şahutoğlu[2], Zafer Dökümcü[1], Coşkun Özcan[1], Ata Erdener[1]

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ABSTRACT

Aim: Robotic-assisted surgery has demonstrated safety and feasibility in numerous pediatric cases. Nevertheless, there is a scarcity of literature regarding advanced pediatric thoracic robotic surgery (APTRS). The objective of this study was to present our experiences with APTRS in 31 patients.

Materials and Methods: From October, 2020 to December, 2023, a total of 31 APTRS procedures were conducted at our institution. A retrospective analysis was carried out, encompassing demographics, indications for surgery, console time, complication rates, length of hospital stay, and postoperative complications.

Results: Twenty-one patients (M/F: 13/17) underwent robotic-assisted surgery, with procedures including thoracic mass excision in 17 cases, esophageal surgery in 8 cases, and various other pathologies in 5 patients. The average age at the time of surgery was 8.4±5.2 years (10 months-17 years), and the average weight was 29.6±18.4 kg (10-65 kg). The mean console time was 165.6±124.8 minutes, with no instances of conversion. The median length of hospital stay was 3.5 days (1-30 days). Postoperative complications occurred in eight patients (25.8%).

Conclusion: Our experience in pediatric robotic thoracic surgery reinforces its suitability even for complex cases. Robotic thoracic surgery appears to offer benefits, particularly in posterior mediastinal mass excision and esophagectomy for corrosive esophageal strictures, when compared to thoracoscopy.

Keywords: Robotic-assisted surgery, thoracoscopy, neuroblastoma, esophageal atresia

Introduction

Robotic surgical procedures in pediatric patients have been becoming more popular, albeit at a slower pace compared to adults, over the years. Robotic-assisted surgery (RAS) brings various advantages such as three-dimensional (3D) vision, enhanced maneuverability, and tremor filtration (1,2). However, there are also notable drawbacks and limitations, with one of the most significant being the inappropriate size of the instruments for small children and neonates.3 Additionally, higher costs and longer operation times can be considered as the other main limitations. Nevertheless, despite these drawbacks, RAS has seen a growing adoption in pediatric surgery in recent years (3). A common concern surrounding pediatric robotic surgeries is the potential limitations posed by the patient's weight and age. However, the literature has reported that this is not an absolute contraindication for RAS (4,5).

RAS has been utilized across various pediatric subspecialties, with pediatric urology being the most commonly reported one (2,3,6). On the other hand, robotic-

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assisted thoracoscopic surgeries (RATS's) have been seldom reported on in the literature (6). Takazawa et al. (7) developed a RATS model specifically for infants and found that robotic suturing was faster than the conventional thoracoscopic approach. Also, there were some reports about esophageal surgeries, thoracic tumoral excision, and lobectomies in the literature (8).

Despite the limited data and the small number of reported cases for RATS, since 2020, we have been conducting robot-assisted thoracoscopic surgery for select cases. This study aimed to share our experience with RATS and delve into the technical aspects of these cases.

Materials and Methods

The present study was conducted in the pediatric surgery department in accordance with international ethical standards and the World Health Organization Helsinki Declaration. Ethics committee approval, confirming that the data collected for this research adhered to ethical guidelines, was obtained from the Ege University Medical Research Ethics Committee (approval no: 24-3.1T/22, date: 21.03.2024). Informed consent was obtained from all participants.

Robotic surgery typically comprises three primary surgical phases. The first step involves thoracoscopic exploration, during which the primary pathology is assessed using thoracoscopy, and port placements are planned. Following this, the second phase is docking, where the robotic arms are positioned and prepared for the surgical procedure. Subsequently, the console phase commences. Finally, the last step is undocking. In some cases, additional steps may be necessary after undocking, such as cervical dissection for procedures such as gastric pull-up.

A retrospective review of the hospital records was conducted for those patients who underwent robot-assisted thoracoscopic surgery between the years 2020 and 2023. The data collected included the patients' demographics, their weight, surgical indications, operational strategies, docking times, and console times, as well as their intraoperative, and postoperative complications.

The patients were categorized into three groups for discussion: thoracic masses, esophageal pathologies, and miscellaneous diseases (Figure 1).

Results

Thirty patients underwent RATS. The mean age at operation was 8.4±5.2 years (range: 1-17 years), and the mean weight at operation was 29.6±18.4 kg (range: 10-65

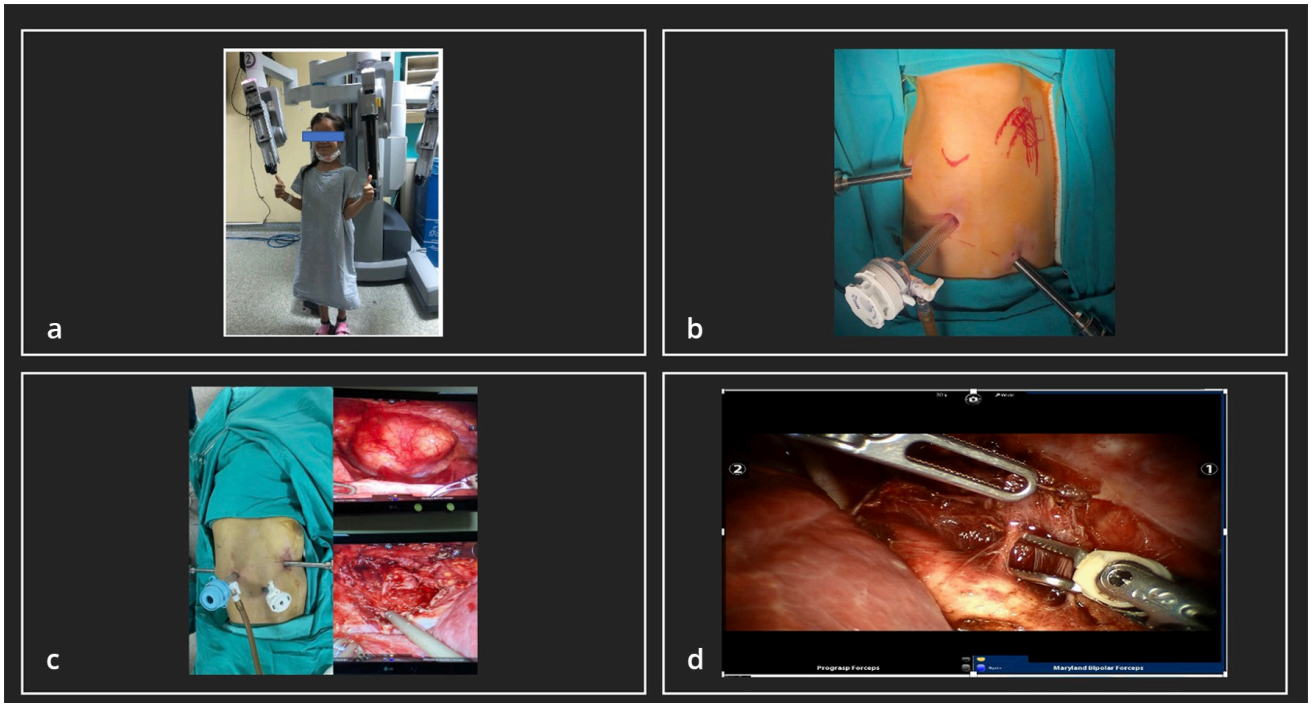

Figure 1. Figures of the robotic system and intraoperative images, a) da Vinci® Si™ Surgical System with patient number 6th. b) port placement of a posterior mediastinal neurogenic tumor; c) port placement and intraoperative views of esophageal duplication, d) the dissection of the neurogenic tumor from the aorta

Table I. Indications for surgery	
Group 1: Thoracic masses (n=17)	
Neurogenic tumors	10
Metastasis	3
Congenital cyst	2
Ewing sarcoma	1
Mature cystic teratoma	1
Group 2: Esophageal pathologies (n=8)	
Corrosive esophageal stricture (CES)	3
Esophageal atresia (EA)	2
Achalasia	2
Hiatal hernia	1
Group 3: Miscellaneous (n=5)	
Morgagni hernia	1
Pulmonary hydatid cyst	1
Abnormal pulmonary venous return	1
CPAM	1
Bronchiectasis	1
CPAM: Congenital pulmonary airway malformation	

kg). The primary diseases were categorized into three groups (Table I). The mean console time was 165.6±124.8 minutes. There were no intraoperative complications related to RATS. However, a tracheal injury occurred during the complementary step after undocking in a patient who had undergone a gastric pull-up for a corrosive esophageal stricture. The majority of patients underwent surgery for thoracic masses, primarily neurogenic tumors. Two cases involved metastases, one from osteosarcoma and the other from hepatocellular carcinoma, requiring rib resection and thoracic wall reconstruction in a Ewing sarcoma patient.

Additionally, there were two cases of congenital cysts: one bronchogenic and the other an esophageal duplication cyst. Among replacement techniques, gastric pull-up (GPU) is the only method which can be performed using minimally invasive techniques. Five patients (3 with corrosive esophageal stricture and 2 with esophageal atresia) underwent robot-assisted GPU. Heller myotomy with Dorr fundoplication were performed for achalasia patients, and the myotomy was facilitated by robotic vision, offering greater reliability compared to laparoscopy. Re-do Nissen fundoplication for recurrent hiatal hernia was deemed more feasible with robotic assistance due to its superior maneuvering capabilities. Lobectomy was performed on two patients with congenital pulmonary airway malformation and bronchiectasis.

Postoperatively, eight patients experienced various complications, including Horner syndrome after neurogenic tumor excision, swallowing dysfunction following gastric transposition, atelectasis, pleural effusion, prolonged air leakage in a lung cyst hydatid case, and one recurrence following Heller myotomy.

The operations were concluded with tube thoracostomy in 20 patients, with a median time of 3 (1-25) days for tube insertion and a median hospital stay of 3.5 (1-30) days.

Discussion

Robotic systems were originally developed for adults. In 2001, robotic surgery was adapted for children for the first time (9). Since then, RAS's have been performed across various subspecialties in pediatric surgery (8). However, there is limited information available specifically regarding thoracic robotic surgery in the pediatric population. A literature review by Saxena et al. (8) identified only seven relevant studies (four on thoracic surgery and three on esophageal surgery) on thoracic robotic surgery published between 2017 and 2022. This highlights the scarcity of data and series focused on thoracic robotic surgery in pediatric patients during that period (8).

Robot-assisted surgery (RAS) brings numerous advantages, including 3D visualization, tremor filtration, and the use of articulating instruments, all of which enhance surgical procedures (3,10). However, in pediatric cases, there are certain challenges (1). The primary limitation is the size difference between the large instruments used in robotic surgery and the small body size of pediatric patients (11). This challenge can be addressed by adjusting the placement of ports or the patient's position during surgery (12,13).

The literature has discussed whether patient-weight poses a limitation for robotic surgery. Molinaro et al. (5) categorized patients into two groups based on their weight and found that operations took significantly longer in those patients weighing less than 15 kg. However, there was no difference in conversion or complication rates between the groups. This led to the conclusion that patient-weight is not an absolute contraindication for robotic surgery (5). In our series, the smallest patient weighed 10 kg and successfully underwent gastric transposition using RAS. This experience supports the feasibility of robotic surgery even in small pediatric patients.

RAS in pediatric surgical oncology has recently gained popularity. However, thoracic tumor cases are rarely reported on in the literature. A systematic review covering the years 2012-2021 identified 10 patients with thoracic

tumors out of 134 patients (14). Blanc et al. (15) reported their experience with 100 oncologic patients and formulated a guideline for patient selection. They suggested that RAS can be considered for paravertebral tumors, tumors limited to the thymic bed, and single lung metastasis. However, they noted that patients younger than 2 years of age are a relative contraindication, while encasement of vessels and extension to the median mediastinum are considered formal contraindications (15). The majority of patients in our series had a posterior mediastinal mass.

Surgeons also perform robotic-assisted esophageal surgery. There are several articles in the literature about gastroesophageal reflux (GER) with or without hiatal hernia and achalasia. Laparoscopic Nissen fundoplication remains the gold standard for the treatment of GER. However, robotic-assisted Nissen fundoplication can be considered in certain cases (16). In our opinion, RAS can be considered for redo procedures. Additionally, robotic-assisted Heller myotomy in achalasia is safer than the laparoscopic technique with regards to mucosal perforation (17).

In our series, we performed five gastric pull-ups. Three patients had corrosive esophageal strictures, and two had esophageal atresia. We performed thoracic esophageal resection and gastric pull-up using robotic assistance. This technique had not been reported on in the literature before. We plan to discuss this technique in subsequent studies.

Li et al. (18) compared the surgical outcomes of thoracoscopic and robotic-assisted pulmonary resection in the pediatric population. They found no difference in perioperative complications, hospital stays, or drainage lengths between the two techniques. However, they noted that while the total operative time was longer, the pure operative time was shorter in RAS (18).

Study Limitations

This study was conducted in order to summarize our experiences with RATS. The operation indications and patients' demographics were varied. Therefore, it was not possible to describe the techniques or port placements for all of the procedures in this study. These will be described separately for each procedure in future publications.

Conclusion

RATS has emerged as a valuable and increasingly utilized approach in pediatric surgery. Our experience and the existing literature suggest that RATS offers several advantages, including improved visualization and greater precision in complex procedures. However, further research is needed to fully understand the optimal applications and outcomes of RATS in pediatric patients. As technology advances and surgical techniques evolve, RATS is likely to play an even more significant role in the management of thoracic conditions in the pediatric population.

Ethics

Ethics Committee Approval: Ethics committee approval, confirming that the data collected for this research adhered to ethical guidelines, was obtained from the Ege University Medical Research Ethics Committee (approval no: 24-3.1T/22, date: 21.03.2024).

Informed Consent: Informed consent was obtained from all participants.

Authorship Contributions

Surgical and Medical Practices: C.Ş., Z.D., C.Ö., Concept: C.Ş., Z.D., Design: Z.D., Data Collection and/or Processing: Ü.Ç., Z.D., Analysis and/or Interpretation: Ü.Ç., Z.D., C.Ö., A.E., Literature Search: Ü.Ç., Z.D., Writing: Ü.Ç., Z.D.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the publication of this article.

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References

1. Shen LT, Tou J. Application and prospects of robotic surgery in children: a scoping review. World J Pediatr Surg 2022; 5:1-6.
2. Krebs TF, Schnorr I, Heye P, Häcker FM. Robotically assisted surgery in children—a perspective. Children (Basel) 2022; 9:836.
3. Boscarelli A, Giglione E, Caputo MR, et al. Robotic-assisted surgery in pediatrics: what is evidence-based?-a literature review. Transl Pediatr 2023; 12:271-9.
4. Meehan JJ, Sandler A. Pediatric robotic surgery: A single-institutional review of the first 100 consecutive cases. Surg Endosc 2008; 22:177-82.
5. Molinaro F, Angotti R, Bindi E, et al. Low weight child: can it be considered a limit of robotic surgery? experience of two centers. J Laparoendosc Adv Surg Tech A 2019; 29:698-702.
6. Richards HW, Kulaylat AN, Cooper JN, McLeod DJ, Diefenbach KA, Michalsky MP. Trends in robotic surgery utilization across tertiary children's hospitals in the United States. Surg Endosc 2021; 35:6066-72.
7. Takazawa S, Ishimaru T, Harada K, et al. Evaluation of surgical devices using an artificial pediatric thoracic model: a comparison between robot-assisted thoracoscopic suturing versus conventional video-assisted thoracoscopic suturing. J Laparoendosc Adv Surg Tech A 2018; 28:622-7.
8. Saxena AK, Borgogni R, Escolino M, D'Auria D, Esposito C. Narrative review: robotic pediatric surgery-current status and future perspectives. Transl Pediatr 2023; 12:1875-86.

9. Meininger DD, Byhahn C, Heller K, Gutt CN, Westphal K. Totally endoscopic Nissen fundoplication with a robotic system in a child. Surg Endosc 2001; 15:1360.

10. Vinit N, Vatta F, Broch A, et al. Adverse events and morbidity in a multidisciplinary pediatric robotic surgery program. a prospective, observational study. Ann Surg 2023; 278:E932-E8.

11. Denning N-L, Kallis MP, Prince JM. Pediatric robotic surgery. Surg Clin North Am 2020; 100:431-43.

12. Kim SJ, Barlog JS, Akhavan A. Robotic-assisted urologic surgery in infants: positioning, trocar placement, and physiological considerations. Front Pediatr 2018; 6:411.

13. Durand M, Musleh L, Vatta F, et al. Robotic lobectomy in children with severe bronchiectasis: A worthwhile new technology. J Pediatr Surg 2021; 56:1606-10.

14. Vatta F, Gazzaneo M, Bertozzi M, Raffaele A, Avolio L, Riccipetitoni G. Robotics-assisted pediatric oncology surgery-a preliminary single-center report and a systematic review of published studies. Front Pediatr 2021; 9:780830.

15. Blanc T, Meignan P, Vinit N, et al. Robotic surgery in pediatric oncology: lessons learned from the first 100 tumors-a nationwide experience. Ann Surg Oncol 2022; 29:1315-26.

16. Binet A, Fourcade L, Amar S, et al. Robot-assisted laparoscopic fundoplications in pediatric surgery: experience review. Eur J Pediatr Surg 2019; 29:173-8.

17. Ballouhey Q, Dib N, Binet A, et al. How robotic-assisted surgery can decrease the risk of mucosal tear during Heller myotomy procedure? J Robot Surg 2017; 11:255-8.

18. Li S, Luo Z, Li K, et al. Robotic approach for pediatric pulmonary resection: preliminary investigation and comparative study with thoracoscopic approach. J Thorac Dis 2022; 14:3854-64.

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Original Article

Position Effect on Cerebral Oxygenation in Neonates During Transition After Birth

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ABSTRACT

Aim: According to delivery room guidelines, an optimal position is not specified for the stabilization of the baby. This study aimed to define the positions effects on postnatal adaptation parameters and cerebral oxygenation in non-resuscitated neonates.

Materials and Methods: A total of 60 neonates delivered by cesarean section stabilized randomly in the supine, right-side, left-side, or prone positions were enrolled. Apgar scores, heart rates (HR), arterial oxygen saturations via pulse oximetry, and perfusion indexes (PI) at the 2nd, 5th and 10th minutes were recorded. Cerebral regional oxygen saturation (SpO_2) of the patients was monitored by near-infrared spectroscopy.

Results: In the prone position, the 1st minute Apgar score was significantly lower than other groups, but no difference was observed at the 5th minute Apgar scores (1st min Apgarprone, p=0.05). Although there was no statistically significant difference, the prone position had the lowest HR at the 2nd minute, while the supine posture had the greatest HR at the 5th and 10th minutes. While the groups' SpO_2 values were similar, the left-side group's perfusion rates increased at the 5th and 10th minute marks (5th and 10th min Plleft-side, p=0.67, p=0.21, respectively). Regional cerebral oxygen saturation ($rScO_2$) and cerebral fractional oxygen extraction did not differ significantly between groups at the 5th and 10th minute time intervals. Although right and left $rScO_2$ were found to be higher in the first 5 minutes in the prone position, this elevation did not lead to a statistically significant difference, and right and left $rScO_2$ values were found to be similar in all groups at the 10th minute.

Conclusion: Adaptation parameters were not affected by position, except for lower 1st minute Apgar scores in the prone group and higher perfusion indices in the left lateral position. Cerebral perfusion was similar in all groups. The left-side position, which results in a higher PI, may be a good alternative. Studies with larger case series may provide further information.

Keywords: Apgar, cerebral oxygenation, delivery room, neonate, position

Introduction

For postnatal adaptation, the first "golden hour", and especially the first "golden minute", after birth are very important. In the delivery room, routine care and resuscitation of newborn infants should adhere to international management guidelines (1,2). Although these are very detailed, it is unclear which position is optimal for the baby immediately after birth. The 2000 Neonatal Resuscitation Program (NRP) guidelines suggested that a supine or lateral position was appropriate (3), but these recommendations were excluded from the updated guidelines (1,2,4,5).

In neonates, the position of the body may affect cardiopulmonary functions (6). Positions other than the

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supine position can provide a faster rising heart rate (HR), easier cleaning of airway secretion, and therefore possibly better cerebral oxygenation (7). Several papers on the newborn's cord clamping time and the necessity of non-invasive breathing have been published in the literature which look at how position affects cerebral regional oxygenation and hemodynamic changes (8,9). When delayed cord clamping occurred after birth, there was no discernible difference in hematocrit between the supine and prone positions at the 30-hour mark. However, prone positioning was observed to increase arterial and cerebral oxygenations in preterm babies undergoing nasal non-invasive ventilation (NIV) for mild-to-moderate respiratory distress.

The Apgar score, developed by Virgina Apgar in 1953, is the most widely used instrument for evaluating a neonate (10). This clinical score takes into account breathing, muscular tone, skin tone, HR, and reflex irritation. However, there are significant inter-observer fluctuations with this score (11). More cardio vascular monitoring could mitigate this restriction. The most recent guidelines for the assistance of baby transition and resuscitation from the European Resuscitation Council recommended employing electrocardiography (ECG) and/or pulse oximetry to check HR as the only cardio circulatory monitoring in the delivery room (12). To assess the neonate's cardio-circulatory state, HR may not be sufficient on its own. Furthermore, cardiac output (CO) may be an important variable in the first few weeks following birth. Thermodilution is the gold standard method for obtaining CO, however its utility and viability in the neonatal population are limited. Thus, the primary purpose of echocardiography in this population is to evaluate CO. However, during the immediate transition period, this is not a practical approach. When echocardiography is combined with cerebral-regional-oxygen-saturation, which may be measured using near-infrared spectroscopy (NIRS), it is useful in evaluating cardio-circulation as well as cerebral autoregulation (13). Delivery and consumption of oxygen are necessary for NIRS measurements. The arterial oxygen saturation (SpO_2), brain perfusion, and hemoglobin concentration of the blood all influence the amount of oxygen given. For cerebral perfusion, systemic vascular resistance and CO are required, as well as brain vascular resistance and cerebral perfusion pressure.

The present study assessed the impact of four different positions, namely supine, right-side, left-side, and prone, on cerebral oxygenation and postnatal adaptability while stabilizing newborns who did not require resuscitation.

Materials and Methods

This study, which lasted two months and was conducted in the delivery room of the department of obstetrics and gynecology by the newborn department, where about 3,500 babies are born every year, was prospective, randomized, and observational. Stabilization was carried out in compliance with NRP guidelines by three pediatric residents and a neonatologist who was certified by the NRP. The study participants were babies with Apgar scores greater than 7 and those who were single, term, and appropriate for gestational age (GA) and did not require resuscitation. Only infants delivered by cesarean section (CS) and who were unable to have early skin-to-skin contact were included, while vaginal births were excluded. Babies with major congenital anomalies, preterm infants, and those requiring positive pressure ventilation were excluded from this study.

The women who were about to undergo CS were placed under spinal anesthesia; oxygen therapy was not started before the baby was delivered. Written consent was acquired and the parents were informed about this study before the baby was born. After the inclusion criteria were met, randomization was performed, and closed-envelope randomization was applied. The infants (15 in each group) were positioned supine, on the right or left side, or prone after randomly opening envelopes with positional codes.

Infant Positioning

When the baby was born, the first care was carried out under a radiant heater. Those babies who required positive pressure ventilation were placed in a supine position and removed from the study population.

Medical Records and Vital Parameters

Thirty seconds after delivery, each evaluation began, and they ended after 10 minutes. The same individual scored each infant's Apgar scores at the first and fifth minutes. They also recorded the infant's HR, SpO_2, and perfusion index (PI) pulse oximetry at the second, fifth, and tenth minutes using a Masimo Radical-7 pulse co-oximeter. The difference in change between the mean values were assessed. After positioning the probe preductally (on the right wrist), the oximeter was attached to it. During every stabilization period, the fraction of inspired oxygen was only 0.21, or "room air". The NRP 2015 recommendation states that aspiration should not be carried out in the delivery room on a regular basis. The number of requirements and duration of tactile stimuli were recorded.

Cerebral Oxygenation

Cerebral oxygenation was assessed using noninvasive NIRS; a Covidien INVOS 5100C device was used to record the mean values at the five and ten-minute marks (14-16). The NIRS sensors were disposable and designed for single-use. Therefore, a separate NIRS probe was used for each patient, following the company's instructions. The NIRS probe was positioned on either side of the frontal lobe.

The baseline value was the first measured value. Regional cerebral oxygen saturation ($rScO_2$) variations were then collected for 60 seconds at 2-second intervals, and the mean value was computed. Normally, the $rScO_2$ ranges from 50 to 80%, but it can fluctuate by 10 to 20% at any time (17). Cerebral ischemia is indicated by a reduction of more than 20% in patients whose basal cerebral $rScO_2$ value is greater than 50%, and a decrease of more than 15% in patients whose basal cerebral $rScO_2$ value is less than 50% (18). After application, data were gathered for ten minutes at a rate of 0.1 Hz for each position. In an animal model, cerebral fractional oxygen extraction (cFOE), which represents the equilibrium between oxygen delivery and consumption in the tissue, was computed as $(SpO_2\text{-}rScO_2)/SpO_2$ for each position for the 5th and 10th minute averages (19).

No financial support was obtained for this study.

Statistical Analysis

The analysis of the variables was conducted using the programs PAST 3 (Hammer, Harper, D.A.T., Ryan, P.D. 2001. Paleontological Statistics) and SPSS 25.0 (IBM Corporation, Armonk, New York, United States). The Levene test was used to assess homogeneity of variance, while the Shapiro-Wilk-Francia test was used to assess conformance of univariate data to the normal distribution. The homogeneity of variance was assessed using the Box-M test, and the conformance of multivariate data to the normal distribution was assessed using the Mardia (Dornik and Hansen Omnibus) test. Post-hoc analyses were conducted using Dunn's test; the Kruskal-Wallis H test, a non-parametric parametric method, was applied to the Monte Carlo simulation results; and Fisher's Least Significant Difference tests were utilized for one-way ANOVA post-hoc analysis when comparing more than two groups based on quantitative data. The Fisher-Freeman-Holton test was used to compare categorical variables with one another using the Monte Carlo Simulation method. Categorical variables are given as n (%) in the tables, while quantitative variables are expressed as mean (standard deviation) and median (25th percentile/75th percentile). The variables were examined with a 95% confidence level, and a p-value of less than 0.05 was accepted as significant.

Results

We recorded 178 live births evaluated by the same delivery room team for 2 months. We included only cesarean-born infants. Totally, 118 infants were excluded from this study, of whom 87 were vaginally born, 22 were preterm, 3 had major congenital anomalies, and 6 required positive pressure ventilation (Figure 1). All other infants were placed in their assigned positions. All groups were similar in terms of gestational weeks and birth weight (Table I). There were no significant differences among the groups in terms of their requirements for aspiration or tactile stimulation (Table I). In the prone position group, the 1st min Apgar score, especially the grimace "reflex response" score, was significantly lower than those of the other groups [1st min Apgar $score_{median}$, supine: 9 (8/9), prone: 8 (8/9), right-side: 9 (8/9), left-side: 9 (9/9); p=0.05] (Table II). The 5th minute Apgar scores were similar in all groups (Table II). The 2nd minute HR was the lowest in the prone position; the 5th and 10th min HRs were highest in the supine position, but there was no statistically significant difference [2nd min

Table I. Characteristics of the study group

	Supine (n=15)	Prone (n=15)	Right-side (n=15)	Left-side (n=15)	p value
Gender (Female), n (%)	7 (46.7)	10 (66.7)	8 (53.3)	10 (66.7)	0.678[f]
Gestational age, median (q1/q3)	39 (39/39)	39 (38/39)	39 (39/40)	39 (38/39)	0.476[k] 0.476[k]
Birth weight (g), mean (SD)	3,304.0 (413.3)	3,174.7 (378.5)	3,295.3 (495.6)	3,307.3 (462.9)	0.813[a]
Aspiration, median (q1/q3)	1 (1/2)	1 (1/1)	1.5 (1/2)	1.5 (1/3)	0.327[k]
Aspiration (yes), n (%)	10 (66.7)	9 (60)	10 (66.7)	6 (40)	0.437[f]
Tactile stimulation (yes), n (%)	10 (66.7)	12 (80)	11 (73.3)	8 (53.3)	0.516[f]

[a]One-Way ANOVA (Robuts Statistic: Brown-Forsythe) - (Method: Bootsrap), [k]Kruskal-Wallis H test (Monte Carlo), [f] Fisher-Freeman-Halton test (Monte Carlo), SD: Standard deviation, q1: Percentile 25, q3: Percentile 75

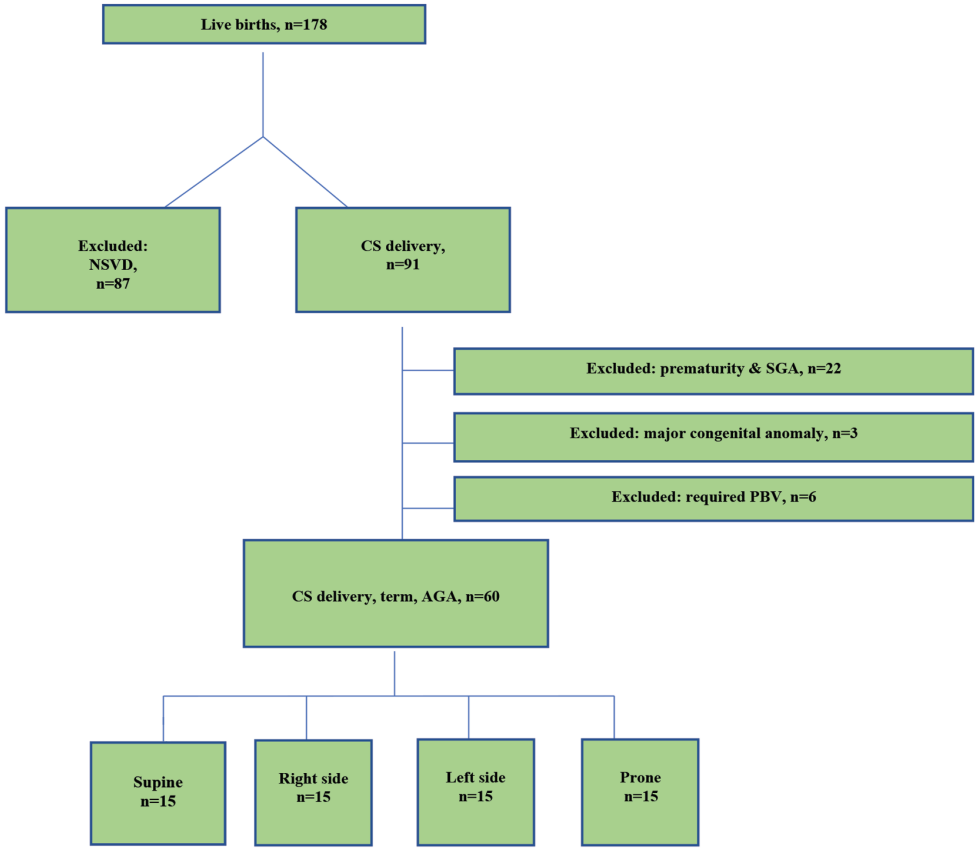

Figure 1. Flow chart of included and excluded term neonates who do not require resuscitation for the first lying position on postnatal adaptation and cerebral oxygenation

NSVD: CS: Cesarean section, SGA: Small for gestational age, AGA: Appropriate for gestational age

Table II. Adaptation parameters

	Supine (n=15)	**Prone (n=15)**	**Right-side (n=15)**	**Left-side (n=15)**	**p value**
APGAR, median (q1/q3)					
1st min.	9 (8/9)	8 (8/9)	9 (8/9)	9 (9/9)	**0.051[k]**
5th min.	10 (9/10)	10 (9/10)	10 (9/10)	10 (10/10)	0.320[k]
Difference (5th-1st) min.	1 (1/2)	1 (1/1)	1 (1/1)	1 (1/1)	0.419[k]
p value for (1st vs 5th min.)	**<0.001[w]**	**<0.001[w]**	**<0.001[w]**	**<0.001[w]**	
Heart rate (/min.), median (q1/q3)					
2nd min.	138 (126/152)	131 (126/145)	139 (130/152)	144 (128/158)	0.539[k]
5th min.	168 (153/176)	161 (147/165)	158 (138/168)	159 (149/165)	0.379[k]
10th min.	170 (152/179)	166 (146/170)	167 (141/172)	169 (166/178)	0.172[k]
p (Heart rate * group)					**<0.001[fr]**
Difference (5th-2nd) min.	18 (14/40)	19 (9/35)	13 (3/24)	18 (5/25)	0.339[k]
Difference (10th-2nd) min.	24 (16/41)	24 (12/36)	19 (7/27)	32 (18/38)	0.289[k]
Difference (10th-5th) min.	4 (2/11)	8 (-2/10)	1 (-8/10)	8 (4/16)	0.100[k]
p value for (2nd vs 5th vs 10th min.)	**<0.001[fr]**	**<0.001[fr]**	0.09[fr]	**<0.001[fr]**	

Table II. Continued

	Supine (n=15)	Prone (n=15)	Right-side (n=15)	Left-side (n=15)	p value
Pairwise comparison for heart rate					
(2^{nd} vs 5^{th}) min.	**0.019**	**0.019**	**ns.**	0.085	
(2^{nd} vs 10^{th}) min.	**<0.001**	**<0.001**	**ns.**	**<0.001**	
(5^{th} vs 10^{th}) min.	0.301	0.820	**ns.**	**0.008**	
Pulse oximetry (SpO_2, %), median (q1/q3)					
2^{nd} min.	70 (67/80)	70 (69/80)	72 (71/80)	75 (70/78)	0.634[k]
5^{th} min.	88 (82/92)	86 (82/91)	86 (82/93)	88 (81/92)	0.948[k]
10^{th} min.	96 (94/99)	95 (92/97)	95 (94/98)	96 (93/98)	0.680[k]
p (Pulse oximetry * group)					0.941[fr]
Difference (5^{th}-2^{nd}) min.	14 (10/18)	14 (6/18)	13 (9/16)	11 (8/15)	0.607[k]
Difference (10^{th}-2^{nd}) min.	25 (16/27)	23 (12/27)	23 (16/25)	21 (19/27)	0.824[k]
Difference (10^{th}-5^{th}) min.	8 (4/12)	8 (4/14)	9 (5/14)	8 (6/15)	0.980[k]
p value for (2^{nd} vs 5^{th} vs 10^{th} min.)	**<0.001[fr]**	**0.001[fr]**	**<0.001[fr]**	**<0.001[fr]**	
Pairwise comparison for Pulse oximetry					
(2^{nd} vs 5^{th}) min.	**0.01**	**0.019**	**0.01**	**0.01**	
(2^{nd} vs 10^{th}) min.	**<0.001**	**<0.001**	**<0.001**	**<0.001**	
(5^{th} vs 10^{th}) min.	**0.032**	**0.019**	0.053	0.053	
Perfusion index, median (q1/q3)					
2^{nd} min.	2.9 (1.8/5.4)	3 (2.1/3.6)	3.8 (2.1/6.1)	2.5 (2.1/3.1)	0.558[k]
5^{th} min.	2.6 (2.4/4.8)	3.2 (2.6/4.2)	3.3 (2.4/5.2)	3.6 (2.6/4.7)	0.671[k]
10^{th} min.	3.4 (2.3/4.6)	2.6 (1.9/6.4)	2.4 (2.1/4.4)	4.1 (3.5/5.6)	0.211[k]
Difference (5^{th}-2^{nd}) min.	-0.1 (-1.2/0.8)	0.5 (-0.8/1.6)	0.3 (-2.2/1)	0.9 (-0.2/2.1)	0.136[k]
Difference (10^{th}-2^{nd}) min.	1.3 (-1.8/1.9)	0.3 (-0.9/3.4)	0.3 (-4/1.3)	2 (-0.5/3.5)	0.083[k]
Difference (10^{th}-5^{th}) min.	0.8 (-0.7/1.3)	-0.2 (-1.1/1.9)	-0.2 (-0.6/0.4)	0.1 (-0.6/2.2)	0.307[k]
p value for (2^{nd} vs 5^{th} vs 10^{th} min.)	**0.469[fr]**	**0.715[fr]**	**0.852[fr]**	**0.066[fr]**	

[k]Kruskal-Wallis H test (Monte Carlo), [w]Wilcoxon signed-rank test (Monte Carlo), [fr]Friedman test (Monte Carlo), q1: Percentile 25, q3: Percentile 75, [A]Expresses significance according to supine group, [B]Expresses significance according to prone group, [C]Expresses significance according to right-side group, [D]Expresses significance according to left-side group

HR, prone: 131 (126/145); p=0.53, 5^{th} and 10^{th} min HR, supine: 168 (153/176); p=0.37, 170 (152/179); p=0.17, respectively] (Table II). The SpO_2 values did not differ between the groups, the 5^{th} and 10^{th} minute perfusion rates were higher in the left-side group [5^{th} and 10^{th} min PI, left-side: 3.6 (2.6/4.7); p=0.67, 4.1 (3.5/5.6); p=0.21, respectively] (Table II). There was no significant difference between the groups in terms of cerebral oxygenation as measured by $rScO_2$ and cFOE at 5^{th} and 10^{th} min [$left_{rScO2}$, p=0.39, $right_{rScO2,}$ p=0.13, $left_{cFOE}$, p=0.58, $right_{cFOE}$, p=0.68] (Table III). Although right and left $rScO_2$ in the prone position did not demonstrate a statistically significant difference, it was found to be higher at the 5^{th} minute [5^{th} min $rScO_2$ in the prone position, left frontal: 76.40 (8.80); p=0.75, right frontal: 77.27 (10.85); p=0.51, respectively], and the right and left $rScO_2$ values at the 10^{th} minute were similar in all groups [10^{th} min $rScO_2$ in all positions, left frontal; p=0.97, right frontal; p=0.84]. None of the study infants required further resuscitation. However, following stabilization, one infant in the supine position and one infant in the right-side position developed respiratory distress. The respiratory distress of the infant in the supine position regressed within 1 hour, and the baby was the given to the mother. However the other infant in the right-side position group had to be transported to the neonatal intensive care unit.

Table III. Cerebral oxygenation measurements

	Supine (n=15)	Prone (n=15)	Right-side (n=15)	Left-side (n=15)	p value
Right frontal NIRS ($rScO_2$, %), mean (SD)					
5th min average	72.53 (10.59)	77.27 (10.85)	72.47 (9.51)	73.13 (9.56)	0.516[a]
10th min average	82.80 (8.26)	82.27 (8.61)	80.20 (8.25)	82.27 (8.73)	0.842[a]
Difference (10th-5th) average	10.27 (7.03)	5.00 (6.02)	7.73 (5.75)	9.13 (6.33)	0.132[ra]
p value for (5th vs 10th min)	**0.002[v]**	**0.009[v]**	**0.002[v]**	**0.001[v]**	
Left frontal NIRS ($rScO_2$, %), mean (SD)					
5th min average	72.87 (8.66)	76.40 (8.80)	75.67 (9.63)	74.47 (10.51)	0.752[a]
10th min average	83.20 (5.03)	83.00 (6.15)	82.53 (7.43)	83.53 (6.48)	0.978[a]
Difference (10th-5th) average	10.33 (5.75)	6.60 (6.21)	6.87 (7.75)	9.07 (7.57)	0.392[ra]
p value for (5th vs 10th min)	**0.001[v]**	**0.006[v]**	**0.006[v]**	**0.002[v]**	
Right FOE, mean (SD)					
5th min average	17.5 (9.7)	11.9 (6.1)	16.1 (7.8)	14.4 (6.9)	0.227[a]
10th min average	15.0 (8.2)	12.6 (7.5)	15.5 (8.3)	13.5 (7.6)	0.732[a]
Difference (10th-5th) average	-2.5 (9.6)	0.7 (4.0)	-0.6 (7.5)	-0.9 (7.2)	0.687[ra]
p value for (5th vs 10th min)	**0.316[v]**	**0.508[v]**	**0.769[v]**	**0.621[v]**	
Left FOE, mean (SD)					
5th min average	15.9 (8.9)	12.0 (5.8)	14.3 (9.2)	13.3 (8.5)	0.605[a]
10th min average	13.3 (3.5)	12.9 (4.8)	13.6 (7.0)	12.4 (4.9)	0.933[a]
Difference (10th-5th) average	-2.7 (7.1)	0.9 (5.1)	-0.7 (9.1)	-0.9 (6.1)	0.582[ra]
p value for (5th vs 10th min)	**0.168[v]**	**0.464[v]**	**0.751[v]**	**0.593[v]**	

[ra]General Linear Model Repeated Anova (Wilks' Lambda), post-hoc test: Fisher's least significant difference (LSD), [v]Paired Samples t-test (Bootstrap), [a]One-Way ANOVA (Robuts Statistic: Brown-Forsythe) - (Method: Bootsrap), SD: Standard deviation, [A]Expresses significance according to supine group, [B]Expresses significance according to prone group, [C]Expresses significance according to right-side group, [D]Expresses significance according to left-side group

Discussion

The international delivery room management guidelines do not give clear information on the initial lying position of those infants who do not require resuscitation. Although positioning may affect cerebral oxygenation in neonates, there are very few studies investigating the effect of positioning on cerebral oxygenation.

The mixed tissue saturation value that cerebral rSO_2 provides allows information on the equilibrium between brain oxygen delivery and oxygen usage. Cerebral rSO_2 is affected by cerebral blood flow, hemoglobin concentration, and SpO_2 (20-22). When contrasted with SpO_2 levels or HR, $rScO_2$ rises and achieves a plateau during the fetal-to-neonatal transition considerably more quickly, suggesting that the brain may receive preferential oxygen delivery (20). The burden of cerebral hypoxia was found to be reduced in the $rScO_2$+SpO_2 group when decisions about providing extra oxygen to newborns during the first 15 minutes after delivery were made based on $rScO_2$+SpO_2 rather than SpO_2 alone (23). Therefore, during immediate transition and resuscitation after birth, $rScO_2$ monitoring to guide respiratory and supplemental oxygen support is feasible.

In our study, there were no differences between the groups in terms of $rScO_2$, cFOE, and SpO_2 in different lying positions in healthy newborns who did not need resuscitation. In certain studies where infants requiring resuscitation were included, differences in NIRS values according to position were observed.

Stabilization in the lateral or prone positions allows for the easy removal of secretions and better oxygenation (24). In both animal studies and adult intensive care patients, arterial blood oxygenation was better when infants were placed prone (25-28). In premature infants receiving NIV, Barsan Kaya et al. (29) showed that prone positions

improved arterial and cerebral oxygenations more than supine positions. In severely preterm newborns (GA 24-28 weeks) at one week of age, Shepherd et al. (30) found increased cerebral FOE but no change in $rScO_2$ and higher SPO_2 values in the prone position during active and calm sleep, suggesting lower cerebral blood flow. Despite the fact that the prone position may increase oxygenation by enhancing the ventilation/perfusion ratio, it is difficult to evaluate the baby's color, HR, reflex response, muscle tone, and respiration rate in this position.

There were no differences in the HR and SpO_2 parameters between the positions in our study. In the prone position, the grimacing reflex response and the 1st minute Apgar score was lower, and the need for tactile stimulation was non-significantly higher, but the statistical difference in Apgar scores between the groups disappeared by the 5th minute. O'Donnell et al. (11) compared the supine and left lateral positions during postnatal adaptation in 82 preterm infants born <32 gestational weeks, and those in the left lateral position respired more effectively as measured by 5th minute pulse oximetry (11). Konstantelos et al. (7) found that babies stabilized in the lateral position were less agitated than those in the supine position as SpO_2 and HR increased more rapidly in these infants.

The pulsatile-to-static blood flow ratio in peripheral tissue, known as PI, is largely based on the volume of blood present at the monitoring point and significantly correlates with left ventricular output in healthy infants (31). Another study conducted in the Republic of Korea evaluated the association between hemodynamic parameters and left ventricle position in a pig model during cardiopulmonary resuscitation. Their results showed that there were substantial relationships between the two (32). In our study, the 5th and 10th minute PI values were insignificantly higher in the left-side lying group. This finding may be related to increased left ventricular CO in the left-sided position. These effects of left- or right-side positioning need to be confirmed by further studies with larger series.

Study Limitations

To date, this was the first study evaluating the effects of position on cerebral oxygenation in healthy-term infants who did not need resuscitation during the transition period in the delivery room. The limitations of this study were its small number of study patients and the use of pulse oximetry instead of ECG in the delivery room. We preferred pulse oximeter measurements to enable a comparison with cerebral oxygenation measurements with NIRS. The third limitation was the impossibility of performing a blind intervention due to the nature of this study.

Conclusion

We found no significant major effects of the initial lying position on postnatal adaptation parameters or cerebral oxygenation during the first 10 minutes of life in term newborns who did not require resuscitation. Only the 1st minute Apgar score was lower in the prone position, but no difference was observed for 5th minute Apgar scores between the groups. The left-side position, which resulted in a higher PI, may be a good alternative. Studies with larger case series may provide further information on this issue.

Ethics

Ethics Committee Approval: This study was approved by the University of Health Sciences Turkey, Dr. Behçet Uz Children's Diseases Training and Research Hospital, Clinical Research Ethics Committee (approval no.: 2018/04-03, date: 22.02.2018).

Informed Consent: Written consent was acquired and the parents were informed about this study before the baby was born.

Authorship Contributions

Surgical and Medical Practices: E.Y.E., R.Ç., Concept: Ö.A.K., Ş.Ç., M.Y., N.K., Design: E.Y.E., R.Ç., Ö.A.K., Ş.Ç., M.Y., N.K., Data Collection and/or Processing: E.Y.E., R.Ç., D.T., Analysis and/or Interpretation: R.Ç., Literature Search: E.Y.E., R.Ç., Writing: E.Y.E., R.Ç.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the publication of this article.

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References

1. Kattwinkel J, Perlman J, Aziz K, et al. Neonatal resuscitation: 2010 American Heart Association Guidelines for Cardiopulmonary Resuscitation and Emergency Cardiovascular Care. Pediatrics 2010; 126:e1400-13.
2. Perlman J, Wyllie J, Kattwinkel J, et al. Part 11: Neonatal resuscitation: 2010 International Consensus on Cardiopulmonary Resuscitation and Emergency Cardiovascular Care Science With Treatment Recommendations. Circulation 2010; 122:S516-38.
3. Niermeyer S, Kattwinkel J, Van Reempts P, et al. International Guidelines for Neonatal Resuscitation: An excerpt from the Guidelines 2000 for Cardiopulmonary Resuscitation and Emergency Cardiovascular Care: International Consensus on Science. Contributors and Reviewers for the Neonatal Resuscitation Guidelines. Pediatrics 2000; 106:E29.

4. American Heart Association. 2005 American Heart Association (AHA) guidelines for cardiopulmonary resuscitation (CPR) and emergency cardiovascular care (ECC) of pediatric and neonatal patients: pediatric basic life support. Pediatrics 2006; 117:e989-1004.
5. Aziz K, Lee HC, Escobedo MB, et al. Part 5: Neonatal Resuscitation: 2020 American Heart Association Guidelines for Cardiopulmonary Resuscitation and Emergency Cardiovascular Care. Circulation 2020; 142:S524-50.
6. Gouna G, Rakza T, Kuissi E, Pennaforte T, Mur S, Storme L. Positioning effects on lung function and breathing pattern in premature newborns. J Pediatr 2013; 162:1133-7.
7. Konstantelos D, Gurth H, Bergert R, Ifflaender S, Rüdiger M. Positioning of term infants during delivery room routine handling - analysis of videos. BMC Pediatr 2014; 14:33.
8. Bensouda B, Mandel R, Mejri A, Tordjman L, St-Hilaire M, Ali N. Supine versus Prone Position during Delayed Cord Clamping in Infants ≥36 Weeks: A Randomized Trial. Am J Perinatol 2023.
9. Barsan Kaya T, Aydemir O, Tekin AN. Prone versus supine position for regional cerebral tissue oxygenation in preterm neonates receiving noninvasive ventilation. J Matern Fetal Neonatal Med 2021; 34:3127-32.
10. Apgar V. A Proposal for a New Method of Evaluation of the Newborn Infant. Originally published in July 1953, volume 32, pages 250-259. Anesth Analg 2015; 120:1056-9.
11. O'Donnell CP, Kamlin CO, Davis PG, Carlin JB, Morley CJ. Interobserver variability of the 5-minute Apgar score. J Pediatr 2006; 149:486-9.
12. Madar J, Roehr CC, Ainsworth S, et al. European Resuscitation Council Guidelines 2021: Newborn resuscitation and support of transition of infants at birth. Resuscitation 2021; 161:291-326.
13. Meng L, Hou W, Chui J, Han R, Gelb AW. Cardiac Output and Cerebral Blood Flow: The Integrated Regulation of Brain Perfusion in Adult Humans. Anesthesiology 2015; 123:1198-208.
14. Mutoh T, Guest RJ, Lamm WJ, Albert RK. Prone position alters the effect of volume overload on regional pleural pressures and improves hypoxemia in pigs in vivo. Am Rev Respir Dis 1992; 146:300-6.
15. Brodsky JB. What intraoperative monitoring makes sense? Chest 1999; 115:101S-5S.
16. Wolf M, Greisen G. Advances in near-infrared spectroscopy to study the brain of the preterm and term neonate. Clin Perinatol 2009; 36:807-34.
17. Baikoussis NG, Karanikolas M, Siminelakis S, Matsagas M, Papadopoulos G. Baseline cerebral oximetry values in cardiac and vascular surgery patients: a prospective observational study. J Cardiothorac Surg 2010; 5:41.
18. Samra SK, Dy EA, Welch K, Dorje P, Zelenock GB, Stanley JC. Evaluation of a cerebral oximeter as a monitor of cerebral ischemia during carotid endarterectomy. Anesthesiology 2000; 93:964-70.
19. Naulaers G, Meyns B, Miserez M, et al. Use of tissue oxygenation index and fractional tissue oxygen extraction as non-invasive parameters for cerebral oxygenation. A validation study in piglets. Neonatology 2007; 92:120-6.
20. Bruckner M, Pichler G, Urlesberger B. NIRS in the fetal to neonatal transition and immediate postnatal period. Semin Fetal Neonatal Med 2020; 25:101079.
21. Pichler G, Schmölzer GM, Urlesberger B. Cerebral Tissue Oxygenation during Immediate Neonatal Transition and Resuscitation. Front Pediatr 2017; 5:29.
22. Garvey AA, Dempsey EM. Applications of near infrared spectroscopy in the neonate. Curr Opin Pediatr 2018; 30:209-15.
23. Pichler G, Urlesberger B, Baik-Schneditz N, et al. Cerebral Oxygen Saturation to Guide Oxygen Delivery in Preterm Neonates for the Immediate Transition after Birth: A 2-Center Randomized Controlled Pilot Feasibility Trial. J Pediatr 2016; 170:73-8.
24. Gillies D, Wells D, Bhandari AP. Positioning for acute respiratory distress in hospitalised infants and children. Cochrane Database Syst Rev 2012; 2012:CD003645.
25. Mezidi M, Guérin C. Effects of patient positioning on respiratory mechanics in mechanically ventilated ICU patients. Ann Transl Med 2018; 6:384.
26. Beck KC, Vettermann J, Rehder K. Gas exchange in dogs in the prone and supine positions. J Appl Physiol (1985) 1992; 72:2292-7.
27. Lamm WJ, Graham MM, Albert RK. Mechanism by which the prone position improves oxygenation in acute lung injury. Am J Respir Crit Care Med 1994; 150:184-93.
28. Mure M, Glenny RW, Domino KB, Hlastala MP. Pulmonary gas exchange improves in the prone position with abdominal distension. Am J Respir Crit Care Med 1998; 157:1785-90.
29. Barsan Kaya T, Aydemir O, Tekin AN. Prone versus supine position for regional cerebral tissue oxygenation in preterm neonates receiving noninvasive ventilation. J Matern Fetal Neonatal Med 2021; 34:3127-32.
30. Shepherd KL, Yiallourou SR, Odoi A, et al. Effects of Prone Sleeping on Cerebral Oxygenation in Preterm Infants. J Pediatr 2019; 204:103-10.
31. Corsini I, Cecchi A, Coviello C et al. Perfusion index and left ventricular output correlation in healthy term infants. Eur J Pediatr 2017; 176:1013-8.
32. Jung YH, Jeung KW, Lee DH et al. Relationship Between Left Ventricle Position and Haemodynamic Parameters During Cardiopulmonary Resuscitation in a Pig Model. Heart Lung Circ 2018; 27:1489-97.

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Original Article

Identification of Significant Risk Factors for Obesity Among Children Aged Five to Nine Years: A Cross-Sectional Analysis

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ABSTRACT

Aim: Being overweight or obese are characterized by excess fat accumulation and pose substantial health risks representing significant public health challenges in the 21st century. Overweight or obese children will potentially be overweight or obese adults in the future. Individuals who are obese during childhood are more likely to develop non-communicable chronic diseases at an earlier age compared to those who are not obese. Therefore, this study aimed to investigate the obesity rates and the associated risk factors for obesity of primary school students in one of İzmir's districts.

Materials and Methods: This study focused on primary school students aged five to nine years in the Bayraklı district of the İzmir province. A total of 535 children were initially included in this study. We excluded 128 children who had chronic diseases or were taking medicine associated with obesity or those who did not agree to participate. The dependent variable was obesity and the independent variables were defined by socio-demographic and natal features, physical activities, eating habits, screen time (television and computer usage), family structure and parental weight. We administered a questionnaire to the parents and measured the children's weight and height thus calculating their body mass index. Chi-square analysis, t-test and the Mann-Whitney U test were used for statistical calculations. A probability rate was used for obesity and overweight associated factors.

Results: The mean age was 8±1.1 years for the 407 children. The overweight rate was 12% and the obesity rate was 12%. We detected that gender was not associated with obesity or being overweight (p=0.486). Birth weight (p<0.05), irregular meal times (p=0.007), eating snacks (p=0.027), high monthly income (p=0.026), maternal (p=0.03) and paternal (p<0.05) obesity or being overweight and being an only child (p=0.031) were found to be associated with obesity or being overweight for the child.

Conclusion: The findings of this study revealed elevated frequencies of being overweight or obese among children aged five to nine years in the Bayraklı district, comparable to rates observed in developed nations. High monthly income, being an only child, and parental obesity or being overweight were associated with the children's obesity or being overweight. These results underscore the necessity for education among parents and caregivers regarding healthy and balanced nutrition practices.

Keywords: Pediatric obesity, obesity risk factors, obesity frequency

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Introduction

Being overweight or obese are defined as abnormal or excessive fat storage which poses a risk to health (1). Childhood obesity remains one of the major public health challenges of the 21st century (2). According to the World Health Organization (WHO) European Regional Obesity Report 2022, the prevalence of overweight or obese individuals increases in the 5-9 year old age group, with approximately one in eight children (11.6%) afflicted by obesity and nearly one in three (29.5%) classified as being overweight (including obesity) (3).

The main cause of childhood obesity is the energy imbalance between calories consumed and expended per day. WHO recognizes that the prevalence of childhood obesity is due to societal changes. Changing diets (such as sweetened beverages, junk food and fast food) increase the daily calorie intake, while a sedentary life (prolonged screen time, decreased physical activity) reduces energy expenditure leading children to becoming overweight or obese (1).

Childhood obesity is strongly linked to polygenic factors. While genetic factors play a role in adipocyte variations, research suggests that a healthy home environment can counteract their effects (4-6). Although some studies have found that socio-economic status is related to childhood obesity, this relationship is still controversial (7,8).

Obese children and adolescents suffer from both short-term and long-term health problems. It is known that most of those who are overweight or obese in childhood are also likely to be overweight or obese in adulthood. Childhood-onset overweight and obesity increase the risk of cardiovascular diseases, diabetes, bone-joint diseases (such as osteoarthritis) and some cancers (such as endometrial, breast and colon cancer) in adulthood. The risk for obesity-related chronic diseases was found to be associated with the duration of obesity and age at onset (9). Therefore, this study aimed to evaluate the effects of socio-economic status on childhood obesity and its risk factors.

Materials and Methods

Study Population

The data of this cross-sectional study were collected between November, 2019 and January, 2020. The number of children aged 5-9 going to primary school in the metropolitan central districts of İzmir province was 139,026 (BIMER). The central districts of İzmir were stratified based on their number of students, and their share of the total was calculated. Districts with 10% or more of the student population were included. A multistage sampling method was used in this study. Each district was accepted as a cluster and the district to be sampled was determined using the cluster sampling method. The selection of districts for the sample was conducted using a random numbers table. The Bayraklı district was chosen as the study site, in which 15,546 children aged 5-9 attended 29 primary schools.

This study aimed to achieve a sample size of at least 485 individuals with a 95% confidence level and a 5% margin of error (assuming a design effect of 2). To accommodate potential contingencies, the sample size was determined to be 534, including a 10% reserve.

The socio-economic status of primary schools in the Bayraklı district was determined through consultation with the Bayraklı District Health Directorate. Schools were classified into three socio-economic categories: high, moderate, and low. The total weight of all three layers in Bayraklı's population aged 5-9 was calculated, and the number of samples to be obtained from each socio-economic status was determined based on their respective weights, resulting in 134 individuals (25%) from a high socio-economic status, 187 individuals (35%) from a moderate socio-economic status, and 214 individuals (40%) from a low socio-economic status.

This study was approved by the Dokuz Eylül University Non-invasive Research Ethics Committee (approval no.: 2019/24-37, date: 30.09.2019). And informed consent was obtained from all children and their families for publication.

Statistical Analysis

The schools were visited in order to measure the heights and weights of the children aged 5-9. The parents were administered a 32-question survey, which assessed their children's birth characteristics, duration of breastfeeding, dietary habits, physical activities, screen time, the parents' anthropometric data, monthly income per capita, and details regarding medications regularly used by the child and the presence of any past-chronic diseases. The children's anthropometric measurements were evaluated using Centers for Disease Control and Prevention growth charts. Being overweight or obese were categorized based on body mass index (BMI). Individuals with a BMI falling between the 85th and 94.9th percentiles were classified as being overweight, while those at or above the 95th percentile were categorized as obese (9).

The data obtained in this research were coded into a database created in the Statistical Package for Social

Sciences program and statistical analyses were made. In descriptive statistics, categorical variables were expressed as frequency (n) and percentage (%). The chi-square test was used in the analysis of categorical variables. The Levene test was used for the normal distribution of the data determined by measurement, and data with normal distributions were tested with the t-test, and those data without normal distribution were tested with the Mann-Whitney U test. A p-value <0.05 was accepted as statistical significance.

Results

Of the 407 children participating in this study, 217 (53.3%) were girls and 190 (46.7%) were boys. The socio-demographic characteristics of the children are given in Table I.

When all the children in our study group were examined, the mean duration of breastfeeding was 15.1 (±10.1) months, and the mean age of starting complementary feeding was 6 (±3.2) months. 39.5% of the children had regular meal times while 60.5% of them had irregular meal times. It was determined that 97.5% of the children consumed junk food and 74.9% consumed fast food, and 41.5% did regular sports. The nutritional characteristics of the children participating in this study are given in Table II.

86% of the children participating in this study were term and 14% were preterm. When the birth weights were evaluated according to the weeks at birth, 7.8% were small for gestational age, 80.6% were found to be appropriate for gestational age, and 11.6% were large for gestational age.

Of the children participating in this study, 5.5% (n=22) were underweight, 70.5% (n=287) had normal weight, 12% (n=49) were overweight, and 12% (n=49) were obese.

When examining the relationship between the socio-demographic characteristics and obesity, significant associations were found between the number of siblings and per capita monthly income. Similarly, when investigating the relationship between the birth characteristics and obesity, significant correlations emerged with birth weight and birth weight adjusted for gestational age. Additionally, in the analysis of the relationship between nutritional characteristics and obesity, meal sequence and the consumption of junk food were found to be significantly associated with obesity. Moreover, a significant relationship was observed between high maternal and paternal BMI and obesity in the child (Table III).

In this study, no statistically significant relationships were found between obesity and gender, duration of breastfeeding, transition to complementary feeding, fast-food consumption, regular sports, or screen time.

Table I. The socio-demographic characteristics of the children

	n	**(%)**
Gender	407	
Female	217	53.3
Male	190	46.7
Age (mean ± SD, years)	8±1.1	
Family health insurance	385	
Present	342	88.8
Absent	43	11.2
Consanguineous marriage	396	
Yes	69	17.4
No	327	82.6
Maternal education status	400	
Illiterate	12	3
Literate	12	3
Primary school graduate	124	31
Secondary school graduate	85	21.2
High school graduate	116	29
Graduated from university	51	12.8
Paternal education status	399	
Illiterate	7	1.8
Literate	5	1.3
Primary school graduate	119	29.8
Secondary school graduate	74	18.5
High school graduate	143	35.8
Graduated from university	51	12.8
Per capita monthly income	371	
Low income	66	17.8
Low-moderate income	106	28.6
High-moderate income	96	25.9
High income	103	27.8
Siblings	405	
Present	327	80.7
Absent	78	19.3

SD: Standard deviation

Table II. The nutritional characteristics of the children		
	n	**%**
Duration of breastfeeding mean (SD), months	15.1±10.1	
Age at initiation of complementary feeding, mean (SD), months	6 (±3.2)	
Regular meals	403	
Yes	244	39.5
No	159	60.5
Junk food consumption	406	
Yes	391	96.3
No	15	3.7
Fast-food consumption	407	
Yes	291	74.8
No	98	25.2
SD: Standard deviation		

Table III. Factors associated with obesity			
	Underweight+Normal n (%)	**Overweight+Obese n (%)**	**p value**
Siblings			
Present	257 (83.4)	70 (72.2)	**0.012**
Absent	51 (16.6)	27 (27.8)	
Per capita monthly income			
Low or moderate income	214 (75.1)	54 (62.8)	**0.026**
High income	71 (24.9)	32 (37.2)	
Birth weight (mean, grams)	3,216±585	3,486±567	**<0.05**
Birth weight by week of birth			
SGA	27 (9.2)	3 (3.2)	**0.001**
AGA	241 (82.3)	71 (75.5)	
LGA	25 (8.5)	20 (21.3)	
Regular meals			
Yes	174 (57.0)	70 (71.4)	**0.007**
No	131 (43.0)	28 (28.6)	
Junk food consumption			
Yes	293 (95.1)	98 (100)	**0.027**
No	15 (4.9)	0 (0)	
Maternal BMI	25±4	27±5	**0.03**
Paternal BMI	26±3	28±4	**<0.05**
SGA: Small for gestational age, AGA: Appropriate for gestational age, LGA: Large for gestational age, BMI: Body mass index			

Discussion

Obesity constitutes one of the significant health challenges in the current era. According to WHO data, the prevalence of being overweight or obese among children and adolescents aged 5-19 years increased from 4% in 1975 to 18% in 2016. In European countries, roughly one in five children and adolescents were found to be overweight or obese, while in the United States, one-third

were overweight and one-fifth were obese. In this study, conducted in the İzmir province of Turkey, the prevalences of being overweight or obese were found to be 12% and 12% for each. Compared to the United States, this rate of being overweight or obese appears lower. This could be explained by the different eating habits in Turkey. However, our obesity prevalence exceeds the average of European countries. Notably, European countries were categorized into four regions and our country was placed in the Mediterranean Region, where the obesity prevalence aligns closely with ours (10).

In our study, we observed a higher prevalence of being overweight among girls (13.4%) compared to boys (10.5%), whereas obesity was more prevalent among boys (15.3%) than girls (9.2%). When being overweight and obesity are considered together, the frequency was higher in males (female: 22.6%, male: 25.8%), although no significant difference was found between gender and obesity (p=0.45). Although there are studies with similar results to our study in our country, there are also studies in which the prevalence of obesity in girls is higher than in boys. In the studies conducted by Dündar and Öz (4) on secondary school students in Samsun and by Özilbey and Ergör (11) on first-grade students in İzmir, the prevalence of being overweight and obese in boys was significantly higher than in girls. Conversely, a study by Yuca et al. (12) among children and adolescents aged 6-18 in the Van province of Turkey revealed higher frequencies of being overweight or obese in girls compared to boys. The variation in prevalence across regions could stem from differing societal attitudes toward gender, potentially resulting in unequal opportunities for physical activity among girls in the eastern part of the country. In contrast, boys in the western part may experience higher rates due to prolonged screen time (such as computer and tablet games) and families with low socio-economic status show differences in care between genders.

A study evaluating the 10-year trend of being overweight or obese in Canada noted higher frequencies among boys, attributing this difference to biological factors such as gender-based fat distribution and energy needs, as well as social and cultural reasons such as food choices and body image differences. It has been suggested that gender-based norms may also play a role (13). These findings parallel those observed in our study in the İzmir province, suggesting a higher development level compared to other Turkish provinces.

Breast milk intake and the duration of transition to complementary feeding were not significantly associated with being overweight or obese. However, a significant relationship was observed between meal patterns and being overweight or obese. ESPGHAN recommends that children should eat at least four meals daily, including breakfast. Literature reviews have revealed that children who eat three or fewer meals and skip breakfast are associated with being overweight or obese (4,5,14). This underscores the importance of both meal frequency and content in relation to being overweight or obese.

Another nutrition-related factor is the consumption of junk food and fast food (p=0.065), with a significant relationship found between the consumption of junk food and being overweight or obese (p=0.027). Various studies have reported that the consumption of obesogenic foods such as sugary drinks, processed foods, fast food, and confectionery increases the rates of being overweight or obese. In one study, it was determined that fast food consumption led to being overweight or obese, and the educational status of the mothers of children who consumed fast food was lower than that of other children (15). However, in our study, no significant relationship was observed between fast food consumption and being overweight or obese. This may be attributed to the fact that the dietary choices for those children in the primary school age group are primarily influenced by their parents and the sale of fast food products in school canteens and the surrounding areas may be restricted due to obesity prevention programs in schools.

In our study, socio-economic status was assessed by investigating the parents' education levels, their monthly income per capita and the occupation of the person who provided the household income. Our initial hypothesis was that quality food and sports activities are expensive, it would be easier for families with a high socio-economic levels to access healthy food and sports activities; therefore, being overweight or obese would be less common in this group. However, contrary to our hypothesis, we found no significant relationship between parental education levels (mother: p=0.22, father: p=0.99) and the occupation of the primary income provider (p=0.66) and being overweight or obese. Notably, a significant association was observed between high monthly income per capita and being overweight or obese (p=0.026).

When reviewing the existing literature, most studies concluded that a low socio-economic level was associated with being overweight or obese (16,17), although some put forward the opposite view (7,18). Furthermore, an increase in maternal education levels were found to be

inversely proportional to both the child and the mother being overweight or obese (19). When other studies were examined in detail, low socio-economic status in developed countries tended to be associated with being overweight or obese, while the opposite held true in underdeveloped or developing countries. However, in our country, it appears that the accessibility of sugary foods often consumed as fast food and snacks, is higher among individuals with higher monthly income per capita. Additionally, the persistent belief that overweight children are healthier, which is prevalent in our country, leads families with high socio-economic status to offer their children calorie-dense foods and larger portions, potentially contributing to higher rates of being overweight or obese in this group.

In our study, both maternal (p=0.03) and paternal (p<0.05) BMI values were found to be associated with being overweight or obese. At the same time, no significant relationship was observed between the presence of consanguinity between the parents and being overweight or obese (p=0.92). While some have found only maternal BMI to be associated with the child's being overweight or obese (20), some studies have found the significance of both parents on being overweight or obese (7). The lifestyles and dietary habits of parents determine the child's lifestyle with the BMI of the parents influencing the child's BMI trajectory in parallel.

In our study, a significant relationship was observed between the presence of siblings and being overweight or obese (p=0.012). Reviewing the literature, there are studies suggesting that being an only child increases the risk of obesity (21,22). For instance, one study examining the effects of the one-child policy on obesity in the People's Republic of China found that children without siblings had higher BMI values (23). Moreover, the educational status of the mother (p=0.001) and father (p=0.015) and the monthly income per capita (p=0.015) in families with one child were found to be higher. These findings suggest that families with many children may have a lower socio-economic status than those families with one child; this may show that they can not meet their children's daily energy requirements according to their age, and therefore, the children are weaker.

Conclusion

This cross-sectional study aimed to investigate the prevalence of being overweight or obese among healthy children aged 5-9 years in the Central Districts of İzmir Metropolitan Province and to identify potential risk factors.

Study Limitations

The limitation of this study was the difficulty of finding a causal relationship with variables such as dietary habits, physical activity, and screen time due to the cross-sectional design of the study. This study showed both similar and different results when compared with the obesity risk factors in the literature. The general conclusion is that socio-economic status and the parents' knowledge of nutrition affect childhood obesity. For this reason, regular and balanced nutrition education is necessary for both the parents and their children for every socio-economic status.

Ethics

Ethics Committee Approval: This study was approved by the Dokuz Eylül University Non-invasive Research Ethics Committee (approval no.: 2019/24-37, date: 30.09.2019).

Informed Consent: Informed consent was obtained from all children and their families for publication.

Authorship Contributions

Surgical and Medical Practices: E.T.T.K., Concept: E.T.T.K., O.T.İ., A.A., T.G., Design: E.T.T.K., O.T.İ., A.A., T.G., Data Collection and/or Processing: E.T.T.K., Analysis and/or Interpretation: E.T.T.K., O.T.İ., A.A., T.G., Literature Search: E.T.T.K., Writing: E.T.T.K.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the publication of this article.

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References

1. Obesity and overweight. https://www.who.int/dietphysicalactivity/childhood_what/en/. Accessed: 01.10.2021.
2. Report on the fifth round of data collection, 2018-2020: WHO European Childhood Obesity Surveillance Initiative (COSI). Copenhagen: WHO Regional Office for Europe; 2022.
3. WHO European Regional Obesity Report 2022. Copenhagen: WHO Regional Office for Europe; 2022. Licence:CC BY-NC-SA 3.0 IGO.
4. Dündar C, Öz H. Obesity-related factors in Turkish school children. Sci World J 2012; 2012:353485
5. Ciardullo S, Salvatore MA, Mandolini D, et al. Trend in Breakfast Consumption among Primary School Children in Italy. Nutrients 2023; 15:4632.
6. Calcaterra V, Cena H, Magenes VC, et al. Sugar-Sweetened Beverages and Metabolic Risk in Children and Adolescents with Obesity: A Narrative Review. Nutrients 2023; 15:702.
7. Yardim MS, Hilal Özcebe L, Araz OM, et al. Prevalence of childhood obesity and related parental factors across socioeconomic strata in Ankara, Turkey. East Mediterr Heal J 2019; 25:374-84.

8. Paalanen L, Levälahti E, Mäki P, et al. Association of socioeconomic position and childhood obesity in Finland: a registry-based study. BMJ Open 2022; 12:e068748.

9. Noncommunicable diseases: Childhood overweight and obesity. https://www.who.int/news-room/q-a-detail/noncommunicable-diseases-childhood-overweight-and-obesity. Accessed: 06/03/2024

10. Garrido-Miguel M, Cavero-Redondo I, Álvarez-Bueno C, et al. Prevalence and Trends of Overweight and Obesity in European Children from 1999 to 2016: A Systematic Review and Meta-analysis. JAMA Pediatr 2019; 173:e192430.

11. Özilbey P, Ergör G. Determining the prevalence of obesity in primary school students and eating habits in Izmir. Turk J Public Health 2015; 13:30-9.

12. Yuca SA, Yilmaz C, Cesur Y, Doğan M, Kaya A, Başaranoğlu M. Prevalence of overweight and obesity in children and adolescents in Eastern Turkey. J Clin Res Pediatr Endocrinol 2010; 2:159-63.

13. Rao DP, Kropac E, Do MT, Roberts KC, Jayaraman GC. Childhood overweight and obesity trends in Canada. Health Promot Chronic Dis Prev Can 2016; 36:194-8.

14. Andre´ M. Toschke, Helmut Ku¨chenhoff, Berthold Koletzko and RVK. Diet and Physical Activity Meal Frequency and Childhood Obesity. Obes Res. 2005; 13:1932–8.

15. Zhao Y, Wang L, Xue H, Wang H, Wang Y. Fast food consumption and its associations with obesity and hypertension among children: results from the baseline data of the Childhood Obesity Study in China Mega-cities. BMC Public Health 2017; 17:933.

16. Rogers R, Eagle TF, Sheetz A, et al. The Relationship between Childhood Obesity, Low Socioeconomic Status, and Race/Ethnicity: Lessons from Massachusetts. Child Obes 2015; 11:691-5.

17. Springer AE, Li L, Ranjit N, Delk J, Mehta K, Kelder SH. School-level economic disadvantage and obesity in middle school children in central Texas, USA: A cross-sectional study. Int J Behav Nutr Phys Act 2015; 12(Suppl 1):S8.

18. Fruhstorfer BH, Mousoulis C, Uthman OA, Robertson W. Socio-economic status and overweight or obesity among school-age children in sub-Saharan Africa - a systematic review. Clin Obes 2016; 6:19-32.

19. Oude Groeniger J, De Koster W, Van Der Waal J. Time-varying Effects of Screen Media Exposure in the Relationship between Socioeconomic Background and Childhood Obesity. Epidemiology 2020; 31:578-86.

20. Xu RY, Zhou YQ, Zhang XM, Wan YP, Gao X. A two-year study of parental obesity status and childhood obesity in China. Nutr Metab Cardiovasc Dis 2019; 29:260-7.

21. Datar A. The more the heavier? Family size and childhood obesity in the U.S. Soc Sci Med 2017; 180:143-51.

22. Chen AY, Escarce JJ. Family Structure and Childhood Obesity: An Analysis Through 8th Grade. Matern Child Health J 2014; 18:1772-7.

23. Min J, Xue H, Wang VHC, Li M, Wang Y. Are single children more likely to be overweight or obese than those with siblings? The influence of China's one-child policy on childhood obesity. Prev Med (Baltim) 2017; 103:8-13.

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Original Article

Effects of Enteral Olive Oil Supplement on Weight Gain, Length of Hospital Stay, and the Development of Some Complications in Preterm Infants: A Randomized Controlled Trial

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ABSTRACT

Aim: The objective of the present research was to compare the nutritional status, weight gain, length of hospital stay, and the development of some complications in very low birth-weight (VLBW) infants who received and those who did not receive olive oil supplementation enterally.

Materials and Methods: This study was a single-blind, randomized controlled trial with 96 VLBW infants (intervention: 48, control: 48) in a neonatal intensive care unit. In this study, those infants who met the inclusion criteria for the study were divided into two groups by using a random number table. The same feeding protocol (breast milk and/or formula milk) was applied to the infants in both groups. From the seventh day of life (after starting to take 25-30 mL/kg/day orally), 0.5 cc/30 mL of olive oil was added to the milk at each feeding of those infants in the intervention group.

Results: In comparison with the control group, the infants in the intervention group had a higher daily weight gain rate in the first month and a higher weight on the tenth day, a shorter transition time to full enteral feeding, a higher amount of calories on the day of transition to full enteral feeding, and a shorter length of hospital stay ($p<0.05$). Furthermore, the need for rectal enema and the prevalence of sepsis, gastrointestinal system intolerance, and bronchopulmonary dysplasia were significantly lower in the intervention group in comparison with the control group ($p<0.05$).

Conclusion: These findings suggest that olive oil supplementation administered enterally to preterm infants can be recommended since it positively affects the development of infants. Trial registration: This study was registered in ClinicalTrials.gov with the following ID: NCT05815849. This study was retrospectively registered on the 14th of April, 2023.

Keywords: Preterm infant, olive oil, neonatal intensive unit, nursing care

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Introduction

It is known that nutrition for very low birth-weight (VLBW, <1,500 g) neonates is as crucial as the treatment they receive in the neonatal intensive care unit in order to prevent problems which may develop in the long-term follow-ups and so to increase their quality of life. In order to support optimal growth, VLBW newborns require parenteral nutrition as a source of energy, and lipid emulsion is a crucial component of total parenteral nutrition (1). Due to preterm birth, VLBW neonates have a weakened immune system and antioxidant defense, which makes them susceptible to oxidative stress. Oxidative stress plays a significant role in the development of diseases such as retinopathy of prematurity (ROP), intraventricular hemorrhage, chronic lung disease, and necrotizing enterocolitis (NEC), all of which can increase the risk of morbidity (2,3). Prematurity also causes an insufficient supply of long-chain polyunsaturated fatty acids (LC-PUFAs), including eicosapentaenoic acid and docosahexaenoic acid, most of which are transferred to the fetus during the third trimester of pregnancy (4,5).

Providing parenteral and enteral feeding support in the early stages and maintaining this treatment regularly may allow the continuation of intrauterine growth and development in the extrauterine period (6). Research has shown that the best nutrition for newborns in all circumstances is breast milk. Unsupplemented breast milk can, however, be nutritionally insufficient for premature newborns for a variety of reasons, despite its many advantages. First, the nutritional contents in breast milk can change over time (7-9), and some nutrients needed to support preterm infants' fast growth are not present in sufficient amounts (7). Of these, fat is one of the most variable nutrients (10). When breast milk alone cannot meet these needs, it is recommended that special nutritional supplements in the form of powder or liquid be added to breast milk and given to the infant (11).

Due to its content, olive oil has antioxidant, cell regenerative, and anti-carcinogenic properties which help digestion (12). There are clear pieces of evidence indicating that parenterally administered oil emulsions can be well tolerated by VLBW and even extremely low birth-weight infants from the first day and even from the first 1-2 hours of life (13-15). The objective of the current research was to compare the nutritional status, weight gain, length of hospital stay, and the development of some complications [bronchopulmonary dysplasia (BPD), ROP, gastrointestinal system (GIS) intolerance, etc.] in preterm neonates who received and those who did not receive olive oil enterally for calorie support.

Materials and Methods

Study Design

The sample of this experimentally designed study consisted of all premature babies (n=387) hospitalized in the Neonatal Care Unit of a hospital in between June, 2020 and March, 2021. The sample consisted of 96 preterm infants (intervention: 48, control: 48).

Sample selection criteria: Preterm infants between the 28th and 36th weeks of gestation at the time of delivery determined by the date of the mothers' last menstruation and obstetric evaluation results, weighing over 1,000 g during the study period, with stable vital signs and being able to consume 75% of the total protein and energy through an orogastric tube, fed with breast milk and breast milk fortifiers, not having any severe neurological condition, not using inotropic, muscle relaxants, sedatives, or analgesics drugs, and having spontaneous respiration were enrolled in this research.

Exclusion criteria: Preterm infants with NEC, pneumothorax, skull fracture, the presence of any major congenital anomalies, suspected or diagnosed metabolic disease, a history of pathological jaundice (jaundice developing in the first 24 hours), hospitalized for less than one month, and having a history of surgery which might affect the residual were not enrolled in this research.

Power analysis was conducted with the G*Power (3.1.9.2) program with the objective of determining the sample size. The effect size (d) was found to be 0.745 by utilizing the mean (28.29 and 20.33) and standard deviations (12.287 and 8.766) acquired from the lengths of hospital stay in the publication titled "A Randomized Controlled Clinical Trial of Olive Oil Added to Human Breast Milk for Weight Gaining in Very Low Birth Weight Infants" (16). Using the formula above, the required minimum sample size was found to be 78 preterm infants, 39 for each group. This study was completed with 96 preterm infants (intervention: 48, control: 48) (Figure 1).

A total of 96 infants forming the study sample were randomly divided in a controlled manner into two groups by their status of receiving or not receiving olive oil supplementation. The urn approach, which is comparable to complete randomization, was used to verify that the groups were randomly assigned (17).

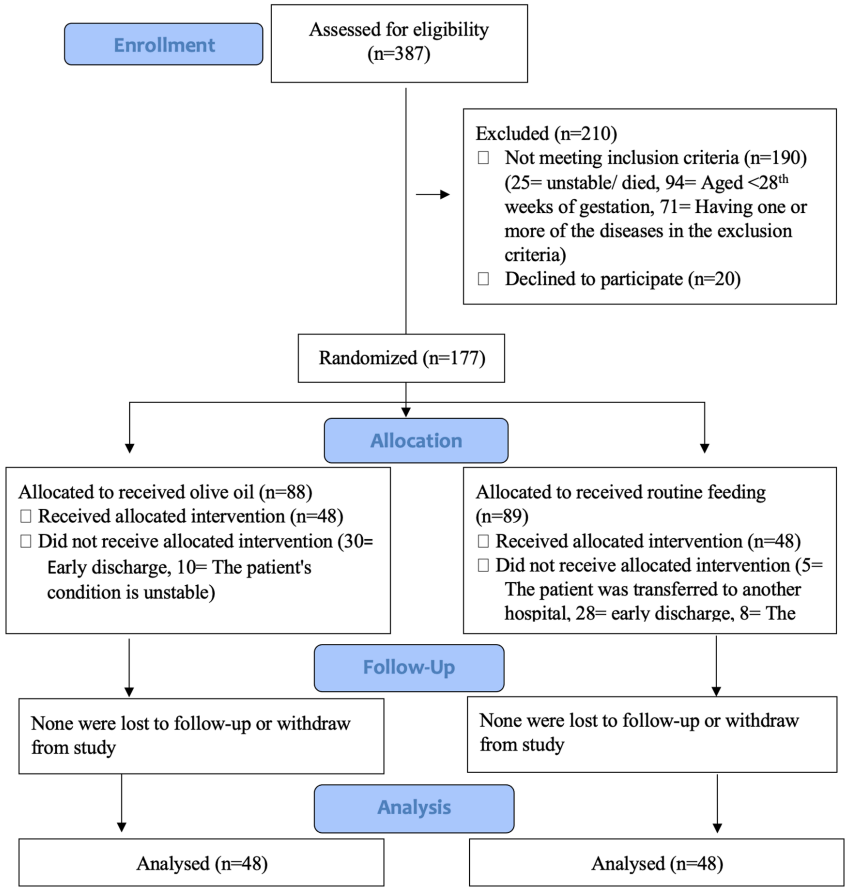

Figure 1. Flow chart of research

Variable Definitions

Demographic and clinical information was obtained from the electronic database of the hospital and patient charts. In sepsis scoring, according to the Turkish Neonatal Society and European Medicines Agency, positivity in at least two of the six clinical categories and at least two of the six laboratory categories was evaluated as clinical sepsis (18). Gastric intolerance (GI) was considered as the inability to digest more than 50% of the enteral nutrition presented as the gastric residual volume, or abdominal distention and vomiting, or both, and accordingly, the patient's nutritional plan being disrupted (6). In the findings of ROP, the ROP Diagnosis and Treatment Guidelines were considered, and it was evaluated as severe ROP in cases of ROP being stage 3 or higher in both eyes or in cases of the infant being treated with laser or anti-vascular endothelial growth factor therapy (19). BPD was graded according to the BPD Prevention and Follow-up Guidelines, and all mild/moderate/severe cases were enrolled in this research. The jaundice levels of the preterm infants were studied by examining direct bilirubin in the blood. Those infants with a history of pathological jaundice were not included in this research (20).

Data Collection

Mother and infant information form: This form includes descriptive information about the mother and the infant (age, weight, birth mode, sex, diagnosis, etc.).

Infant follow-up form: In the said form, information such as the date on which olive oil supplementation was started enterally, vital signs measured before and after the intervention, and the development of complications in the infant were recorded.

Procedure

- Those families who wished to participate in this study received an informed consent form, which they reviewed and accepted. The data gathering forms were filled out.

- The same feeding protocol (breast milk and/or Low Birth-Weight Infant Milk Formula) was applied to the infants in both groups, and minimal enteral nutrition was started at 15-20 mL/kg/day (21). Prior to the procedure, the breast milk of the preterm babies in both groups was warmed in a regular food warmer and supplemented with eoprotein (one measure of eoprotein to 25 cc of breast milk).
- Approximately from the seventh day after starting to take 25-30 mL/kg/day orally, 0.5 cc/30 mL of olive oil (a brand easily available in the markets) was added to the milk at each feeding of the infants in the intervention group (9,16,22). The preterm infants were fed in this way for at least one week until discharge. The oil used for this purpose was stored in a special glass bottle with a label attached and protected from heat/light to prevent contamination. The olive oil and breast milk mixture was carefully shaken by a staff member previously trained for this project until the mixture turned into microlipids. The target was determined as being 150-160 mL/kg, and if growth was insufficient, it was determined as being 180-200 mL/kg (if tolerated).
- Prior to feeding, the babies in both groups had their vital signs measured (heart rate 100-160 beats per minute, axillary temperature 36.5-37.4 °C, respiration rate <60/min) (23).
- Following the placement of an OG or NG tube for position control, the neonates in both groups were fed while lying supine and had their heads elevated between 30 and 45 degrees.
- The infants' daily weight gain, GI, BPD, and ROP findings were monitored daily until discharge, and jaundice findings were evaluated by considering the bilirubin levels on the 10th and 15th days and recorded in the infant follow-up form together with the other findings.

Ethical Considerations

The Clinical Studies Ethics Committee of Kahramanmaraş Sütçü İmam University in Turkey approved this study (date: 25.12.2019, approval no.: 15). All subjects provided written informed permission prior to inclusion in this research.

Statistical Analysis

In the study, 98 participants' data were analyzed and input into the IBM SPSS Statistics 23 application. The analysis of the participants' descriptive characteristics was carried out by utilizing frequency (n, %) for categorical variables and mean and standard deviation for continuous variables. A comparison of qualitative data was carried out using Pearson's chi-square test. The difference between two-group discontinuous variables was examined using the independent samples t-test. Statistical significance was considered at a p-value <0.05.

Results

When the individuals in the intervention and control groups were compared according to their birth weight, length, sex, week of gestation, and average head circumference at birth, the groups did not show statistically significant differences (p>0.05) (Table 1).

In terms of their length of hospital stay, weight growth, and feeding, there was a statistically significant difference found between the groups. Accordingly, in comparison with the control group, the participants in the intervention group had a higher daily weight gain rate in the first month and a higher weight on the tenth day, a shorter transition time to full enteral nutrition, a higher amount of calories on the day

Table I. Comparison of some descriptive and clinical characteristics by the groups

	Intervention group (n=48) Mean±SD	**Control group (n=48) Mean±SD**	**Test value**	**p value**
Week of gestation week	30.1875±2.98	29.10±2.43	t=1.948	0.054
Birth weight (gr)	1214.89±223.07	1128.54±219.48	t=1.912	0.059
Birth length (cm)	37.83±2.67	37.16±2.81	t=1.189	0.237
Head circumference at birth (cm)	27.72±2.18	27.6042±2.34	t=.270	0.788
Sex	**n (%)**	**n (%)**		
Female	30 (62.5)	22 (45.8)	χ^2: 2.685	0.101
Male	18 (37.5)	26 (54.2)		

χ^2: Pearson's chi-square test, t: Student's t-test, *p<0.05, **p<0.001
SD: Standard deviation

of transition to full enteral nutrition, and a shorter length of hospital stay (Table 2).

When the participants were compared according to sepsis, rectal enema, BPD, GI intolerance, and bilirubin value, a statistically significant difference emerged between the groups. Accordingly, the participants in the intervention group had a significantly lower probability of receiving rectal enemas in comparison with the control group. Moreover, the participants in the intervention group had significantly less sepsis, GIS intolerance, and BPD than those in the control group. Additionally, the bilirubin levels on the 15th day were found to be statistically significantly lower in the control group in comparison with the intervention group ($p<0.05$). There was no statistically significant difference between the intervention and control groups in terms of the development of ROP and bilirubin levels on the 10th day ($p>0.05$) (Table 3).

Upon comparing the participants in the intervention and control groups according to the time of discontinuation of respiratory support, a statistically significant difference was identified between the groups. Accordingly, the participants in the intervention group had a statistically significantly shorter time to the discontinuation of respiratory support in comparison with the control group ($p<0.05$) (Table 4).

Table II. Comparison of weight gain, nutrition, and length of hospital stay in preterm neonates by the groups

Variables	Intervention group (n=48)	Control group (n=48)	Test value	p value
	Mean±SD	Mean±SD		
Daily weight gain rate in the first month (gr)	13.45±2.17	11.77±2.39	t=3.619	**0.000****
Weight on the tenth day (gr)	1264.27±260.35	1125.10±222.00	t=2.818	**0.006****
Weight at discharge (gr)	1924.16±124.00	1949.68±163.16	t=-0.863	0.390
Transition time to full enteral nutrition (days)	12.12±6.26	20.18±13.54	t=-3.743	**0.000****
Calorie amount on the day of transition to full enteral nutrition	137.37±22.35	127.02±18.45	t=2.474	**0.015***
Length of hospital stay (days)	52.00±21.32	73.22±22.79	t=-4.712	**0.000****

t: Student's t-test, *p<0.05, **p<0.001
SD: Standard deviation

Table III. Comparison of some complications in preterm neonates by the groups

Variables		Intervention group (n=48)	Control group (n=48)	Test value	p value
		n (%)	n (%)		
Status of receiving rectal enema	Yes	6 (12.5)	34 (70.8)	33.600	**0.000****
	No	42 (87.5)	14 (29.2)		
Sepsis	Yes	8 (16.7)	23 (47.9)	10.720	**0.001****
	No	40 (83.3)	25 (52.1)		
GI	Yes	7 (14.6)	26 (54.2)	16.670	**0.000****
	No	41 (85.4)	22 (45.8)		
ROP	Yes	23 (47.9)	17 (35.4)	1.543	0.214
	No	25 (52.1)	31 (35.4)		
BPD	Yes	5 (10.4%)	16 (33.3)	7.375	**0.007****
	No	43 (89.6%)	32 (66.7%)		
		Mean±SD	Mean±SD	Test value	p value
Bilirubin value on the 10th day		4.88±2.58	4.24±2.30	1.283	0.203
Bilirubin value on the 15th day		3.65±2.50	2.57±1.71	2.462	**0.016**

$\chi 2$: Pearson's chi-square test, t: Student's t-test, *p<0.05, **p<0.001
GI: Gastric intolerance, ROP: Retinopathy of prematurity, BPD: Bronchopulmonary dysplasia, SD: Standard deviation

Table IV. Comparison of the time of discontinuation of respiratory support in preterm neonates by the groups

Variables	Intervention group (n=27)	Control group (n=34)	Test value	p value
	Mean±SD	Mean±SD		
Time of discontinuation of respiratory support (days)[a]	22.63±15.87	36.20±22.02	t=-2.694	0.009**

$\chi 2$: Pearson's chi-square test, t: Student's t-test, *p<0.05, **p<0.001
[a]: The participants who did not receive respiratory support at all or whose respiratory support was discontinued within the first 7 days were excluded from the statistical analysis.

Discussion

Preterm infants are vulnerable to postnatal nutritional deficiencies, e.g., fat, since they do not receive the nutrients they need to store in the third trimester of pregnancy and cannot experience the rapid growth phase (4,24,25). Despite the numerous benefits of human milk for the said population, its fat content varies considerably and may be insufficient for optimum growth and development. Hence, the additional fat intake of the infant can be ensured by adding commercially prepared fat mixtures to a small amount of expressed breast milk. Fat taken with the diet is crucial to maintaining energy, growth, and long-term health in preterm infants. Nevertheless, in a systematic review published in the Cochrane Database by Amissah et al. (9), the researchers revealed no clear pieces of evidence for the benefits or harms of adding fat to breast milk in preterm infants (10). Therefore, there is a need for studies assessing these effects, including LC-PUFA supplementation.

The most suitable oils to add to neonatal nutrition nowadays are a combination of medium-chain triglycerides (MCTs) and long-chain triglycerides (LCTs). Although LCTs contain a high amount of essential fatty acids which can be found in almost all vegetable oils, such as olive oil, MCTs cannot provide essential fatty acids. Nevertheless, the impacts of essential fatty acids (unsaturated) on the neurodevelopmental growth of infants have been proven (5,26,27). It is thought that the natural antioxidant effect of phenolic compounds and oleic acid, which is rich in unsaturated fats, in olive oil strengthens the infant's immune system and may also positively affect some physiological parameters in bone development (28). This study demonstrated that adding vegetable oil (olive oil) to the diet of preterm infants is highly effective in terms of weight gain and shortening the length of hospital stay due to improving the immune system. Along with the significant anti-inflammatory effects of olive oil and its bioactive compounds, the evidence of its effects on inflammatory bowel disease has also been emphasized in the recent literature (29,30). The lower level of BPD and the lower incidence of GI in those individuals given olive oil in our study were thought to be associated with these properties of olive oil. Similar to our study, the study by Amini et al. (16) also determined that olive oil positively impacted the weight gain and length of hospital stay of infants. Furthermore, although the table did not display a statistically significant difference in complications related to GI and respiration, it was observed that they were more common in the control group in comparison to the intervention group. However, in another pilot study with a limited number of infants, Ecevit et al. (22) found no significant difference between the two groups according to weight gain, length of hospital stay, and oxidative stress-related diseases. In another study also conducted with a limited number of infants (n=14), Polberger et al. (32) stated that adding extra fat to breast milk for preterm infants did not have a clear benefit in terms of short-term weight gain, length growth, and head growth rates. However, they also found no pieces of evidence that adding additional fat increased the risk of feeding intolerance. In this respect, our study will contribute to the field due to it being an evidence-based randomized controlled trial which included a larger sample group compared to other studies on this subject.

Study Limitations

There were some limitations in this study. Firstly, follow-ups after long-term use of olive oil could not be performed due to the infants being discharged. Secondly, the amount of olive oil administered orally may vary depending on each infant' weight and health status. Further studies are needed on standardized usage amounts.

Conclusion

Olive oil is a very important natural antioxidant and anti-inflammatory nutrient, especially for premature infants. In conclusion, considering the current research results, the use of vegetable oils to increase caloric intake in preterm neonates appears to deserve further and more comprehensive investigation. As adding additional fat to breast milk is already carried out as part of multi-nutrient

supplementation, we urge that future studies look at the effects of the fat component on short- and long-term growth, body fat ratio, blood sugar, and brain development. Considering the differences depending on the season and agricultural areas where olive oil is produced, it is extremely important to present standard content in clinical routine use. Bioavailability studies of the bioactive components contained in olive oil need to be carried out, especially in this high-risk patient group. Furthermore, the correct amount and composition of the extra fat required for preterm infants, its potential side effects, and its impact on delivery practices are among the other issues which should be researched.

Ethics

Ethics Committee Approval: The Clinical Studies Ethics Committee of Kahramanmaraş Sütçü İmam University in Turkey approved this study (date: 25.12.2019, approval no.: 15).

Informed Consent: All subjects provided written informed permission prior to inclusion in this research.

Authorship Contributions

Surgical and Medical Practices: S.A., M.S., S.Y., Y.Ç., S.T., S.Y.Ç., Y.Ö., M.N.Y.İ., Concept: S.A., M.S., S.Y., Y.Ç., S.T., S.Y.Ç., Y.Ö., M.N.Y.İ., Design: S.A., M.S., Y.Ç., Y.Ö., M.N.Y.İ., Data Collection and/or Processing: S.T., S.Y.Ç., Y.Ö., M.N.Y.İ., Analysis and/or Interpretation: S.A., M.S., S.Y., S.T., S.Y.Ç., Literature Search: S.A., M.S., S.Y., S.T., S.Y.Ç., Y.Ö., Writing: S.A., S.Y., S.T., S.Y.Ç., Y.Ö., M.N.Y.İ.

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References

1. Uauy R, Hoffman DR, Peirano P, Birch DG, Birch EE. Essential fatty acids in visual and brain development. Lipids 2001; 36:885-95.
2. Sharda B. Free radicals: emerging challenge in environmental health research in childhood and neonatal disorders. Int J Environ Res Public Health 2006; 3:286-91.
3. Lembo C, Buonocore G, Perrone S. Oxidative Stress in Preterm Newborns. Antioxidants (Basel) 2021; 10:1672.
4. Robinson DT, Martin CR. Fatty acid requirements for the preterm infant. Semin Fetal Neonatal Med 2017; 22:8-14.
5. Grimm H, Mertes N, Goeters C, et al. Improved fatty acid and leukotriene pattern with a novel lipid emulsion in surgical patients. Eur J Nutr 2006; 45:55-60.
6. Moore TA, Wilson ME. Feeding intolerance: a concept analysis. Adv Neonatal Care 2011; 11:149-54.
7. Su BH. Optimizing nutrition in preterm infants. Pediatr Neonatol 2014; 55:5-13.
8. Bhatia J. Human Milk for Preterm Infants and Fortification. Nestle Nutr Inst Workshop Ser 2016; 86:109-19.
9. Amissah EA, Brown J, Harding JE. Fat supplementation of human milk for promoting growth in preterm infants. Cochrane Database Syst Rev 2020; 8:CD000341.
10. Boquien CY. Human Milk: An Ideal Food for Nutrition of Preterm Newborn. Front Pediatr 2018; 6:295.
11. Underwood MA. Human milk for the premature infant. Pediatr Clin North Am 2013;60: 189-207.
12. Lucas L, Russell A, Keast R. Molecular mechanisms of inflammation. Anti-inflammatory benefits of virgin olive oil and the phenolic compound oleocanthal. Curr Pharm Des 2011; 17:754-68.
13. Thureen PJ. Early aggressive nutrition in very preterm infants. Nestle Nutr Workshop Ser Pediatr Program 2007; 59:193-204.
14. Drenckpohl D, McConnell C, Gaffney S, Niehaus M, Macwan KS. Randomized trial of very low birth weight infants receiving higher rates of infusion of intravenous fat emulsions during the first week of life. Pediatrics 2008; 122:743-51.
15. Salama GS, Kaabneh MA, Almasaeed MN, Alquran MIa. Intravenous lipids for preterm infants: a review. Clin Med Insights Pediatr 2015; 9:25-36.
16. Amini E, Shariat M, Nayeri F, Nili F, Ebrahim B, Dalili H. A Randomized Controlled Clinical Trial of Olive Oil Added to Human Breast Milk for Weight Gaining in Very Low Birth Weight Infants. J Family Reprod Health 2011; 5:73-8.
17. Kanik EA, Taşdelen B, Erdoğan S. Klinik Denemelerde Randomizasyon. Marmara Medical Journal 2011;24:149-55.
18. Satar M, Arısoy AE, Çelik İH. Türk Neonatoloji Derneği yenidoğan enfeksiyonları tanı ve tedavi rehberi. Türk Pediatri Arşivi 2018; 53:88-100.
19. Derneği TN, Derneği TO. Türkiye Prematüre Retinopatisi Rehberi 2021 Güncellemesi. [cited 2022 Aug 27]. Available from: http://www.neonatology.org.tr/wp-content/uploads/2021/08/Turkiye-Premature-Retinopa-tisi-2021-Guncellemesi-1.pdf
20. Derneği TN. Yenidoğan Sarılıklarında Yaklaşım, İzlem ve Tedavi Rehberi 2022 Güncellemesi. [cited 2022 Aug 27]. Available from: http://www.neonatology.org.tr/wp-content/uploads/2022/09/Turk-Neonatoloji-Dernegi-Sarilik-Rehberi-2022-Guncellemesi.pdf
21. Kültürsay N, Bilgen H, Türkyılmaz C. Türk Neonatoloji Derneği Prematüre ve Hasta Term Bebeğin Beslenmesi Rehberi. Turk Pediatri Ars 2018; 53:S109-18.
22. Ecevit A, Abbasoglu A, Tugcu U, et al. PO-0588 Neonatal Outcomes Of Very Low Birth Weight Infants Who Received Enteral Nutrition With And Without Olive Oil Support: Randomised Controlled Pilot Study. Arch Dis Child 2014; 99:A444.
23. Acunaş B, Uslu S, Baş A. Y.Türk Neonatoloji Derneği yüksek riskli bebek izlem rehberi. Türk Pediatri Arşivi 2018; 3:180-95.
24. Delplanque B, Gibson R, Koletzko B, Lapillonne A, Strandvik B. Lipid Quality in Infant Nutrition: Current Knowledge and Future Opportunities. J Pediatr Gastroenterol Nutr 2015; 61:8-17.

25. Hadley KB, Ryan AS, Forsyth S, Gautier S, Salem N Jr. The Essentiality of Arachidonic Acid in Infant Development. Nutrients 2016; 8:216.

26. Makrides M, Gibson RA, McPhee AJ, et al. Neurodevelopmental outcomes of preterm infants fed high-dose docosahexaenoic acid: a randomized controlled trial. JAMA 2009; 301:175-82.

27. Nova K EM, Dyer RA, Innis SM. High dietary omega -6 fatty acids contribute to reduced docosahexaenoic acid inhibits secondary neuritis growth. Brain Res. 2008; 1237: 136-4.

28. Bayaz M. Natürel Zeytinyağlarındaki Fenolik Bileşiklerin Biyolojik Aktivitesi. Akademik Gıda 2016; 14:441-50.

29. Cicerale S, Lucas LJ, Keast RS. Antimicrobial, antioxidant and anti-inflammatory phenolic activities in extra virgin olive oil. Curr Opin Biotechnol 2012; 23:129-35.

30. Vrdoljak J, Kumric M, Vilovic M, et al. Effects of Olive Oil and Its Components on Intestinal Inflammation and Inflammatory Bowel Disease. Nutrients 2022; 14:757.

31. Polberger SK, Axelsson IA, Räihä NC. Growth of very low birth weight infants on varying amounts of human milk protein. Pediatr Res 1989; 25:414-9.

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Original Article

Pediatric Diabetic Ketoacidosis: A Retrospective Study on Triggering Factors and Complications in a Turkish Intensive Care Unit

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ABSTRACT

Aim: Diabetic ketoacidosis (DKA) is a critical, potentially life-threatening complication of diabetes mellitus (DM) in children, characterized by hyperglycemia, acidosis, and ketonemia/ketonuria. Despite its known risk factors and mortality rates, few studies have focused on the pediatric population, especially in specific regions under standard treatment protocols. This study investigated the demographic, clinical, and laboratory characteristics of children with DKA, identified its triggering factors, the factors affecting DKA severity and its complications, as well as evaluating the outcomes of a standardized treatment protocol in a Turkish pediatric intensive care unit (PICU).

Materials and Methods: In this single-center retrospective study at Göztepe Prof. Dr. Süleyman Yalçın City Hospital's PICU, we included 115 children diagnosed with DKA between 2015 and 2022, following the DKA Treatment Protocol of the Turkish Society of Pediatric Emergency and Intensive Care Medicine and the International Society for Pediatric and Adolescent Diabetes guidelines. We analyzed the patients' demographic, clinical, and laboratory characteristics, treatment outcomes, and their complications using SPSS 25.0.

Results: The sample primarily consisted of female patients and those newly diagnosed with DM, with a median age of 110 months. The mortality rate was low at 0.87%, with one death due to sepsis-induced multiple organ failure. DKA severity (lower GCS, younger age, electrolyte imbalance, acidosis, complications) correlated with longer PICU stays and recoveries in the children. The findings also highlighted the standardized treatment protocol's effectiveness in managing DKA and reducing complications.

Conclusion: This study underscores the importance of early diagnosis, standardized treatment protocols, and comprehensive care in pediatric DKA management. It emphasizes the need for ongoing education and awareness among healthcare providers and caregivers in order to prevent DKA and its severe outcomes. Further multicenter studies are necessary to extend these findings to the broader pediatric population and refine DKA management strategies.

Keywords: Diabetic ketoacidosis, pediatrics, intensive care, treatment outcomes, Turkey

Introduction

Diabetic ketoacidosis (DKA), characterized by hyperglycemia, acidosis, and ketonemia/ketonuria, is a severe acute complication of diabetes mellitus (DM) and a significant cause of morbidity and mortality. DKA occurs in 15-70% of children with DM at disease onset and in 1-10% of those children previously diagnosed with DM (1).

DKA is associated with various clinical signs and symptoms, including dehydration, tachypnea, nausea, vomiting, abdominal pain, and impaired consciousness.

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DKA primarily affects children with type 1 DM (T1DM), but children with type 2 DM may also develop DKA (2). In our pediatric intensive care unit (PICU), the DKA Treatment Protocol of the Turkish Society of Pediatric Emergency and Intensive Care Medicine and the International Society for Pediatric and Adolescent Diabetes (ISPAD) guidelines are followed in the diagnosis, follow-up, and treatment of patients with DKA.

The most well-known conditions which increase the risk of DKA development are reportedly low socio-economic level, discontinuation of insulin use, insulin pump malfunctions, poor metabolic control, infections, adolescence, and difficulty in accessing healthcare services (3).

The mortality rate in children with DKA has been reported as 0.4-3.4% in the literature (4,5). The most common cause of death from acute complications in children with DKA is cerebral edema, followed by electrolyte abnormalities, acute kidney injury (AKI), and pancreatitis (6).

In light of this information, this study was carried out to investigate the demographic, clinical, and laboratory characteristics of those children hospitalized with a diagnosis of DKA in a PICU, the triggering factors of DKA, the factors affecting the severity of DKA and DKA-related complications in order to determine changes which may improve disease outcomes when implemented within the scope of the general management of DKA.

Materials and Methods

This study was designed as a single-center retrospective study. The study protocol was approved by the İstanbul Medeniyet University Göztepe Süleyman Yalçın City Hospital Clinical Research Ethics Committee (approval no: 2022/0666, date: 23.11.2022). This study was conducted in accordance with the Declaration of Helsinki, good clinical practice, and all applicable laws and regulations.

The study population consisted of 2,460 children monitored in the PICU of Göztepe Süleyman Yalçın City Hospital, which has a capacity of 9 beds and serves approximately 250 teritary pediatric (6 months to 18 year old) intensive care patients per year. It was carried out between January 2015 and December 2022. The patients' data were obtained from the archive files available in the hospital information management system. Children with incomplete information in their files, those with only hyperglycemia, and those who did not meet the diagnostic criteria for DKA were excluded from this study. In total, our study included 115 children hospitalized with DKA in our PICU, representing 4.7% of all patients admitted during the study period.

DKA Treatment Protocol of the Turkish Society of Pediatric Emergency and Intensive Care Medicine and ISPAD guidelines were used in diagnosing DKA, determining its severity, and classifying the patients (1,3).

The anamneses of the patients were evaluated in order to determine whether they were newly diagnosed with DM, whether they had previously had DKA, and how many DKAs they had had. In this context, the patients' ages, genders, body mass index values, admission dates to the PICU, admission symptoms, Glasgow Coma Scale scores, mechanical ventilation needs, duration of crystallized insulin intakes, lengths of PICU and hospital stays and mortality status were recorded. Serum biochemistry measurements included the measurement of serum glucose, sodium, blood urea nitrogen, creatinine, blood and urine ketone and hemoglobin A1c (HbA1c) values, as well as pH, bicarbonate (HCO_3^-), carbon dioxide (CO_2) and lactate levels within the scope of venous blood gas analysis. The data recorded on a data form also included whether the patients had experienced cerebral edema, AKI, infection, etc., during their treatment. AKI cases were defined in accordance with the Kidney Disease: Improving Global Outcomes clinical practice guidelines. Accordingly, those children with an increase in serum creatinine levels of at least 1.5 times compared to their estimated baseline values were considered to have AKI (7).

Statistical Analysis

The collected data were analyzed using the SPSS 25.0 (Statistical Product and Service Solutions for Windows, Version 25.0, IBM Corp., Armonk, NY, US, 2017) software package. The study's findings are presented as means and standard deviations (SD) or frequencies and percentages. The Kolmogorov-Smirnov test was used to determine whether numerical variables conformed to the normal distribution. One-way analysis of variance (ANOVA), an extension of the independent samples t-test, was used to compare data meeting parametric assumptions. The Mann-Whitney U test was used to analyze relationships between skewed variables, and Pearson's correlation analysis was used to analyze the relationships between numerical variables. Values of $p \leq 0.05$ were deemed to indicate statistical significance.

Results

The incidence of DKA among all patients followed up at our institution has ranged from 1.9% to 11% over the

years. Our study of DKA patients found a predominance of newly diagnosed and females with a median age of 110 [interquartile range (IQR), 56-165] months. Almost all of the patients had T1DM (96.5%). On the other hand, most of the patients did not have any comorbidity or a history of DKA attacks (62.6%). Analysis of our cohort revealed a recurrent DKA rate of 18.2%.

There was no mortality except for one patient who presented with DKA and sepsis and died due to sepsis-driven multiple organ failure and brain edema. The median duration of DM in those patients with a diagnosis of DM was 5 (IQR, 3.5-8) years, and the median HbA1c level was 12.3% (IQR, 11%-14.4%). The patients' demographic characteristics and admission findings are given in Table I.

Accordingly, among those patients hospitalized with a diagnosis of DKA, patients newly diagnosed with DM were significantly younger than those previously diagnosed with DM (p=0.00, <0.001). The pediatric risk of mortality III scores and HbA1c values were found to be significantly higher in those patients with newly diagnosed DM than in those with previously diagnosed DM (p≤0.05). On the other hand, there was no significant difference in the time to recovery from DKA between those patients with newly diagnosed DM and those with previously diagnosed DM.

Table I. Patients' demographic characteristics and admission findings

Variables	Overall study group (n=115)	Patients newly diagnosed with DM (n=36)	Patients previously diagnosed with DM (n=79)	p values
Age (months), median (IQR)	110 (56-165)	164 (122-184)	88 (39-132)	**<0.001**
Male gender, n (%)	43 (37.4)	8 (22)	35 (44.3)	0.037
PRISM III score, median (IQR)	4 (2.5-5.8)	3.6 (1.5-4.9)	4.6 (2.8-6.9)	**0.010**
GCS score, median (IQR)	14 (13-15)	14 (13-14)	14 (12-15)	**0.046**
DM diagnosis, n (%)				
Previously diagnosed	36 (31.3)	-	-	-
Newly diagnosed	79 (68.7)	-	-	-
Type of DM, n (%)				
Type 1	109 (96.5)	32 (88)	79 (100)	0.014
Type 2	2 (1.8)	2 (6)	0	0.014
MODY	2 (1.8)	2 (6)	0	0.014
Patients with a comorbidity, n (%)	17 (14.8)	4 (11.1)	13 (16.5)	0.570
DKA severity, n (%)				
Severe DKA	102 (88.7)	29 (81)	73 (92.4)	0.064
Moderate DKA	13 (11.3)	7 (19)	6 (7.6)	
DKA history, n (%)				
None	94 (81.7)	15 (41.6)	79 (100)	<0.001
Second DKA	12 (10.4)	12 (33.3)	0 (0)	
Third DKA or more	9 (7.8)	9 (25)	0 (0)	
Mortality rate, n (%)	1 (0.9)	0	1 (1.26)	
Duration of DM in previously diagnosed patients, median (IQR)		5 (3.5-8)		
Admission findings, n (%)				
Polydipsia	56 (48.7)	3 (8.3)	53 (67.1)	**<0.001**
Polyuria	59 (51.3)	4 (11.1)	55 (69.6)	**<0.001**
Tachypnea	39 (33.9)	17 (47.2)	22 (27.8)	**0.042**
Weight loss	26 (22.6)	0 (0)	26 (32.9)	**<0.001**

Table I. Continued

Variables	Overall study group (n=115)	Patients newly diagnosed with DM (n=36)	Patients previously diagnosed with DM (n=79)	p values
Enuresis	6 (5.2)	0 (0)	6 (7.6)	0.090
Nausea and vomiting	70 (60.9)	25 (69.4)	45 (56.9)	0.220
Stomach ache	30 (26.1)	9 (25)	21 (26.5)	0.850
Fatigue	72 (62.6)	17 (19.4)	55 (69.6)	**0.020**
Laboratory parameters, median (IQR)				
HbA1c level (%)	12.3 (11-14.4)	10.8 (8.8-11.2)	13.6 (12.7-14.8)	**<0.001**
pH value	6.9 (6.9-7)	6.8 (6.8-6.9)	6.96 (6.9-7.0)	**<0.001**
Bicarbonate level (mmol/L)	7.8 (5.8-6.9)	4.75 (3.6-5.8)	6 (5.1-7.2)	**<0.001**
Lactate level (mmol/L)	1.55 (1.1-2.4)	1.4 (0.9-2)	1.6 (1.1-2.7)	0.120
Glucose level (mg/dL)	474 (405-597)	490.5 (442-574)	468 (379-599)	0.340
Blood ketone level (mmol/L)	5.8 (4.1-7.7)	6.7 (4.7-8)	5.3 (4-7)	0.150
Urea level (mg/dL)	26 (19-36)	28 (20-41)	26 (17.8-36)	0.270
Sodium level (mmol/L)	134 (131-139)	135 (132-141)	134 (131-138)	0.530
Triggering factors, n (%)				
Insulin disruption	20 (17.4)	20 (55.5)	0 (0)	<0.001
Insulin pump malfunctions	2 (1.7)	2 (5.5)	0 (0)	NaN
Pneumonia	10 (8.7)	2 (5.5)	8 (10.1)	0.870
Gastroenteritis	9 (7.8)	5 (13.9)	4 (5)	0.140
Urinary tract infection	7 (6.1)	2 (5.5)	5 (6.3)	0.420

DM: Diabetes mellitus, IQR: Interquartile range, PRISM III: Pediatric risk of mortality III, GCS: Glasgow coma scale, MODY: Maturity-onset diabetes of the young, DKA: Diabetic ketoacidosis, HbA1c: Hemoglobin A1c

The median time to recovery from DKA was 19 (IQR, 13-26.2) hours, the median length of PICU stay was 2 (IQR, 2-3) days, and the median length of hospital stay was 8 (IQR, 5-12) days. The most prevalent complications encountered in our study were cerebral edema (6.9%) and AKI (6%).

In our cohort, DKA discharge time was significantly shorter with increasing GKS, pH, and bicarbonate levels (p=0.000, p=0.003, p=0.012, respectively). Hypernatremia, the need for invasive mechanical ventilation, the need for dialysis, cerebral edema, AKI, and older age were significantly associated with longer DKA discharge times (p=0.022, p=0.000, p=0.001, p=0.017, p=0.000, p=0.015, respectively). PICU length of stay was significantly shorter with increasing GKS and pH levels (p=0.000, p=0.007, respectively). Age, urea levels, sodium levels, the need for invasive mechanical ventilation, the need for dialysis, AKI, cerebral edema, and MODS were positively correlated with PICU lengths of stay (p=0.009, p=0.021, p=0.001, p=0.000, p=0.000, p=0.000, p=0.006, p=0.011, respectively) (Table II).

Table II. Factors affecting time to recovery from DKA and length of PICU stay

	Time to recovery from DKA		Length of PICU stay	
	r	p values	r	p values
GCS score	-0.374	<0.001	-0.356	**<0.001**
PRISM III score	0.033	0.725	-0.184	0.050
Age	0.230	**0.015**	0.243	0.009
HbA1c level	-0.024	0.830	0.074	0.525
Glucose level	-0.074	0.430	0.030	0.752
Urea level	0.163	0.085	0.220	**0.021**
Sodium level	0.215	**0.022**	0.290	**0.001**
DM diagnosis	-0.112	0.237	0.061	0.521
DKA severity	0.151	0.108	0.184	**0.049**
Duration of DM	0.131	0.164	-0.023	0.805
DKA history	0.120	0.205	-0.143	0.128
pH value	-0.280	**0.003**	-0.251	**0.007**
Bicarbonate level	-0.238	**0.012**	-0.128	0.181
Lactate level	-0.087	0.417	-0.199	0.060

Table II. Continued

	Time to recovery from DKA		Length of PICU stay	
	r	**p values**	**r**	**p values**
Base deficit	-0.151	0.108	-0.042	0.654
Blood ketone level	-0.120	0.418	0.028	0.848
IMV need	0.404	**<0.001**	0.429	**<0.001**
Dialysis need	0.307	**0.001**	0.328	**<0.001**
Complications	0.399	**<0.001**	0.398	**<0.001**
Brain edema	0.223	**0.017**	0.256	**0.006**
Hypokalemia	0.297	**0.001**	0.322	**<0.001**
AKI	0.357	**<0.001**	0.257	**0.006**
MODS	0.177	0.060	0.237	**0.011**

DKA: Diabetic ketoacidosis, PICU: Pediatric intensive care unit, GCS: Glasgow coma scale, PRISM III: Pediatric risk of mortality III, HbA1c: Hemoglobin A1c, IMV: Invasive mechanical ventilation, AKI: Acute kidney injury, MODS: Multiple organ dysfunction syndrome

Discussion

Our study aimed to contribute to the literature by characterizing the demographic, clinical, laboratory, precipitating factors, and complication profiles of cases followed up in a PICU due to DKA. However, it was not possible to reach significant findings on those factors which determine the severity of DKA. Therefore, there is a need for prospective studies with larger sample sizes.

The proportion of DKA cases among patients admitted during the study period was consistent with the range of 0.8% to 5.6% reported in a meta-analysis of 19 studies involving adult patients from North America, Europe, and Israel (8). Similarly, a study by Albuali and Al-Qahtani (9) reported a DKA incidence of 3.93% in critically ill pediatric patients.

In a review of our PICU population, the incidence of DKA did not demonstrate a statistically significant trend over time. In contrast, a significant increase in the incidence of DKA in 2020 and 2021 due to the coronavirus disease-2019 (COVID-19) pandemic was reported in the literature (10). The discrepancy between the said finding of our study and the relevant findings of studies in the literature can be attributed to the fact that the emergency and outpatient services in our country continued to operate effectively during the COVID-19 pandemic.

In our study, 68.7% of the patients hospitalized in the PICU due to DKA did not have a history of T1DM. This finding is consistent with other relevant studies conducted in other developing countries (11). The overall mortality rate in children with DKA reportedly varies between 3.4-13.4% in developing countries (12). In comparison, only one of the 115 DKA patients in our sample died. The cause of death of the said patient was sepsis-related multiple organ failure. In addition, the patient also developed cerebral edema during the follow-up period, but this was treated successfully. In undiagnosed diabetic patients, stress factors such as intercurrent infections pave the way for a worsening of the clinical condition and a deterioration of the response to treatment.

The use of bicarbonate to eliminate acidosis in children with DKA is not recommended unless there is life-threatening hyperkalemia associated with an increased risk of complications such as cerebral edema and hypokalemia (1). In accordance with this, bicarbonate was not used in any patient in our cohort. Insulin treatment generally causes a decrease in the serum potassium level due to an increase in potassium uptake into the cell and potentially causes insulin to have an aldosterone-like effect on the renal tubule, further increasing potassium losses through urine (13).

However, severe hypokalemia (<2 mEq/L) has been reported very rarely in children with DKA in the literature (14). In parallel, severe hypokalemia was not observed in any patient in our cohort. Additionally, T-wave flattening, QT prolongation, short P-R interval, U wave, ventricular dysrhythmia, and electrocardiographic changes were not observed in patients with hypokalemia (15).

AKI is one of the most common complications of severe DKA and it is often associated with reduced renal perfusion caused by intravascular volume depletion (16). Consistent with the relevant data in the literature, AKI was detected in 6% of those patients with DKA admitted to the PICU in our cohort (17,18). The mechanism of brain edema, which is the leading cause of mortality among DKA complications, is thought to be cerebral hypoperfusion and reperfusion injury associated with neuroinflammation (19). In our investigation, cerebral edema exhibited a higher incidence than the previously reported rate of 1% in the medical literature (20). Our finding might be attributed to the fact that 88.7% of DKA patients admitted to our clinic had severe DKA. In the literature, the overall mortality rate in pediatric patients with DKA who develop brain edema has been reported to be around 20% (21,22). In comparison, in our study, one (16.7%) of the six patients with brain edema died due to sepsis-related multi-organ failure. None of the patients with DKA who we followed up with brain edema had neurological sequelae at the time of discharge, and all of them were successfully treated with mannitol, a hypertonic agent. Consistent with the published literature,

our study identified the most frequent causes of recurrent DKA to be accidental/deliberate insulin discontinuation, intercurrent infections, and undetected malfunctions of insulin pumps (1). The involvement of clinical psychologists and appropriate therapeutic interventions are paramount in mitigating these detrimental factors.

The primary strength of our study was that the data of the patients who were followed up and treated under the same protocol and hospitalized for a certain period were analyzed without exceptions which could have created bias, thus making our findings fully comparable. Additionally, our sample size was large enough to include patients of all pediatric ages who were hospitalized for a relatively common reason. We could not find any other comparable study conducted in Turkey recently.

Study Limitations

The primary limitations of our study were its retrospective nature, reliance on archival records, and difficulty establishing causality. The lack of sociodemographic data, such as parental education and the parents' roles in their children's compliance with glycemic control, which may be influential in developing DKA in the pediatric population, may be considered another limitation. Future large-scale, multicenter, and well-designed prospective studies may shed more light on DKA's prevalence, complications, and importance in this special patient population.

Conclusion

This study, conducted in a PICU in Turkey, provides valuable insights into the demographic, clinical, and laboratory characteristics of those children diagnosed with DKA, alongside evaluating the triggering factors, severity, complications, and outcomes under a standardized treatment protocol. Our findings highlight the predominance of female patients and a significant portion of newly diagnosed DM cases within the study population. The low mortality rate observed, at 0.87%, underscores the effectiveness of the standardized treatment protocols in place, which align with the guidelines of the Turkish Society of Pediatric Emergency and Intensive Care Medicine and the ISPAD.

The association between the severity of DKA at admission, the presence of complications such as cerebral edema or AKI, and the length of PICU stay with recovery time emphasizes the critical need for early diagnosis and the initiation of standardized treatment protocols. These findings advocate for the importance of continuous education and awareness among healthcare providers and caregivers in order to prevent the occurrence of DKA and its severe outcomes.

Moreover, this study sheds light on the necessity for further multicenter studies in order to generalize these findings to the broader pediatric population across different regions. Such research could lead to the development of more refined DKA management strategies, potentially reducing the incidence and severity of DKA in children. Our research underscores the critical role of standardized care protocols in managing pediatric DKA effectively and highlights areas for future investigation in order to further enhance patient outcomes.

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Ethics

Ethics Committee Approval: The study protocol was approved by the İstanbul Medeniyet University Göztepe Süleyman Yalçın City Hospital Clinical Research Ethics Committee (approval no: 2022/0666, date: 23.11.2022).

Informed Consent: This study was designed as a single-center retrospective study.

Authorship Contributions

Surgical and Medical Practices: A.Ş.D., M.E., M.D., Concept: A.A., Design: A.A., Data Collection and/or Processing: A.A., A.Ş.D., M.E., Analysis and/or Interpretation: A.A., A.Ş.D., M.D., Literature Search: A.A., Writing: A.A.

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References

1. Glaser N, Fritsch M, Priyambada L, et al. ISPAD clinical practice consensus guidelines 2022: Diabetic ketoacidosis and hyperglycemic hyperosmolar state. Pediatr Diabetes 2022; 23:835-56.
2. Dabelea D, Rewers A, Stafford J, et al. Trends in the prevalence of ketoacidosis at diabetes diagnosis: the SEARCH for diabetes in youth study. Pediatrics 2014; 133:e938-45.
3. Besli GE, Akyıldız BN, Ağın H, et al. Diyabetik Ketoasidoz Tedavi Protokolü. J Pediatr Emerg Intensive Care Med 2020; 7(Suppl-1):74-90.
4. Benoit SR, Zhang Y, Geiss LS, Gregg EW, Albright A. Trends in diabetic ketoacidosis hospitalizations and in-hospital mortality

- United States, 2000-2014. MMWR Morb Mortal Wkly Rep 2018; 67:362-5.

5. Syed M, Khawaja FB, Saleem T, Khalid U, Rashid A, Humayun KN. Clinical profile and outcomes of paediatric patients with diabetic ketoacidosis at a tertiary care hospital in Pakistan. J Pak Med Assoc 2011; 61:1082-7.
6. Rewers A, Chase HP, Mackenzie T, et al. Predictors of acute complications in children with type 1 diabetes. JAMA 2002; 287:2511-8.
7. Khwaja A. KDIGO clinical practice guidelines for acute kidney injury. Nephron Clin Pract 2012; 120:c179-84.
8. Fazeli Farsani S, Brodovicz K, Soleymanlou N, Marquard J, Wissinger E, Maiese BA. Incidence andprevalence of diabetic ketoacidosis (DKA) among adultswith type 1 diabetes mellitus (T1D): a systematic literature review. BMJ Open 2017; 7:e016587.
9. Albuali WH, Al-Qahtani MH. Diabetic ketoacidosis and its severity predictors in type 1 diabetic children; a 10-year experience of a teaching hospital in Saudi Arabia. Rev Diabet Stud 2022; 18:146-51.
10. Qeadan F, Tingey B, Egbert J, et al. The associations between COVID-19 diagnosis, type 1 diabetes, and the risk of diabetic ketoacidosis: A nationwide cohort from the US using the Cerner Real-World Data. PLoS One 2022; 17:e0266809.
11. Onyiriuka AN, Ifebi E. Ketoacidosis at diagnosis of type 1 diabetes in children and adolescents: frequency and clinical characteristics. J Diabetes Metab Disord 2013; 12:47.
12. Poovazhagi V. Risk factors for mortality in children with diabetic ketoacidosis from developing countries. World J Diabetes 2014; 5:932-8.
13. Carlotti AP, St George-Hyslop C, Bohn D, Halperin ML. Hypokalemia during treatment of diabetic ketoacidosis: clinical evidence for an aldosterone-like action of insulin. J Pediatr 2013; 163:207-12.e1.
14. Davis SM, Maddux AB, Alonso GT, Okada CR, Mourani PM, Maahs DM. Profound hypokalemia associated with severe diabetic ketoacidosis. Pediatr Diabetes 2016; 17:61-5.
15. Sharieff GQ, Rao SO. The pediatric ECG. Emerg Med Clin North Am 2006; 24:195-20.
16. Huang JX, Casper TC, Pitts C, et al. Association of acute kidney injury during diabetic ketoacidosis with risk of microalbuminuria in children with type 1 diabetes. JAMA Pediatr 2022; 176:169-75.
17. Myers SR, Glaser NS, Trainor JL, et al. Frequency and risk factors of acute kidney injury during diabetic ketoacidosis in children and association with neurocognitive outcomes. JAMA Netw Open 2020; 3:e2025481.
18. Passanisi S, Salzano G, Basile P, et al. Prevalence and clinical features of severe diabetic ketoacidosis treated in pediatric intensive care unit: a 5-year monocentric experience. Ital J Pediatr 2023; 49:58.
19. Azova S, Rapaport R, Wolfsdorf J. Brain injury in children with diabetic ketoacidosis: Review of the literature and a proposed pathophysiologic pathway for the development of cerebral edema. Pediatr Diabetes 2021; 22:148-60.
20. Glaser N, Barnett P, McCaslin I, et al., Pediatric Emergency Medicine Collaborative Research Committee of the American Academy of Pediatrics. Risk factors for cerebral edema in children with diabetic ketoacidosis. The Pediatric Emergency Medicine Collaborative Research Committee of the American Academy of Pediatrics. N Engl J Med 2001; 344:264-9.
21. Lawrence SE, Cummings EA, Gaboury I, Daneman D. Population-based study of incidence and risk factors for cerebral edema in pediatric diabetic ketoacidosis. J Pediatr 2005; 146:688-92.
22. Glaser N, Barnett P, McCaslin I, et al. Risk factors for cerebral edema in children with diabetic ketoacidosis. N Engl J Med. 2001; 344:264-9.

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Original Article

Investigation of Phagocyte Functions in Pseudomonas-Colonized Cystic Fibrosis Patients

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ABSTRACT

Aim: Cystic fibrosis (CF) is an autosomal recessive disorder. Although it is considered as an epithelial disease due to impaired chloride transport, its pathogenesis remains unclear. CF is classified as a syndrome with congenital defects of phagocyte in recent human inborn errors of immunity phenotypic classifications. Neutrophils are the most effective cells in the eradication of bacterial infections such as *Pseudomonas aeruginosa*. The aim of the present study was to investigate the phagocyte functions in pseudomonas colonized CF patients.

Materials and Methods: A total number of 26 pseudomonas colonized CF patients and 21 healthy controls (gender and age matched) were included in this study. Absolute neutrophil counts (ANC), immunoglobulin values (Ig), the Migratest to evaluate chemotaxis in neutrophils and monocytes, CD11A/CD18/CD15 S (β2 integrin) adhesion molecules, and the Phagoburst test for intracellular bacterial killing were analyzed by flow cytometer.

Results: ANC, CD15S expression on neutrophils and IgG, IgA and IgM levels were higher in the CF patients than the control group ($p<0.01$). The neutrophils oxidative burst activity and the chemotactic ability of the CF patients did not differ from those of the controls. Patients with allergic bronchopulmonary aspergillosis and those with a mutation of 2183AA>G had significantly lower chemotaxis indexes than the others ($p=0.01$, $p=0.01$ respectively).

Conclusion: Our results from a small group of patients does not support impaired functions such as migration and phagocytosis of neutrophils in patients with CF. Further studies involving more CF patients are needed to make a definitive interpretation.

Keywords: Cystic fibrosis, chemotaxis, phagocytosis, neutrophil function, adhesion molecule

Introduction

Cystic fibrosis (CF) is an autosomal recessive disorder caused by mutations in the gene encoding the transmembrane regulatory protein (CFTR). As a result of the defect in ion transport, a deterioration of chloride and fluid transport in the respiratory tract epithelium leads to mucus formation with increased viscosity, impaired mucociliary clearance, colonization of the airway epithelium with opportunistic infectious agents such as *Pseudomonas aeruginosa* (*P. aeruginosa*), which is one of the most damaging pathogens, and eventually the development of lung disorders seen in patients with CF (1). The pathogenesis of CF remains unclear. In recent years, abnormal increases in inflammation and defects in the clearance of pathogens are now thought to occur as a result of CFTR mutations

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in immune cells. In 2020, it was classified as a congenital defect of the phagocyte system (2).

Neutrophils and monocytes are phagocytic cells which play a role in innate immunity. First, they migrate to the inflammatory site in response to chemoattractant. The other steps are recognition, adhesion, phagocytosis, and the killing of the target. Neutrophilia of the airway is a common feature seen in CF patients, even in the absence of positive bacterial cultures (3). However, as a result of defective CFTR expression in neutrophils, chloride transport into the phagolysosome is impaired, hypochlorous acid cannot be produced, and bacteria cannot be killed, and impaired phagolysosome activity contributes to airway damage by secreting proteins and enzymes such as elastase, which has been associated with bronchiectasis and severe lung damage (4).

Selectins and integrins are adhesion molecules which have different roles during the process of leucocyte-endothelial adhesion. While selectins mediate margination and slow rolling, integrins play a role in both the rolling and arrest on the vascular endothelium. The integrins CD11a/CD18 (lymphocyte function-associated antigen-1/LFA-1), which are expressed in leukocytes, help the leukocyte transmigration to the inflammation area, lymphocyte co-stimulatory signaling, and T-lymphocyte alloantigen-induced proliferation. De Rose et al. (5) reported that the high p-selectin and e-selectin levels they detected were caused by the defective process in neutrophil adhesion in patients with CF. Sorio et al. (6) reported that β1 and β2 integrin-related activation of chemotaxis are defective in monocytes in patients with CF, and CF is a monocyte-selective adhesion defect.

The disruption of the normal phagocytic defense of the host has been associated with biofilm formation, which is also the reason why CF patients are chronically infected with strains resistant to antibiotics. The conversion of *P. aeruginosa* from a non-mucoid phenotype to a mucoid phenotype usually causes a deterioration in respiratory functions and a worsening of the prognosis (7).

In conclusion, it is not clear whether the primary defect is neutrophil and monocyte function defect or an excessive response to over pro-inflammatory stimuli. The aim of the present study was to investigate phagocytic cell functions in pseudomonas colonized CF patients.

Materials and Methods

Patient Selection

A total number of 26 patients with CF who were pseudomonas colonized and 21 healthy controls were included in this study. Informed consent was obtained from all patients and parents. This study was approved by the Ege University Faculty of Medicine Medical Research Ethics Committee (approval no.: 20-12.1T/37, date: 17.12.2020) and sponsored by the University Office of Scientific Research Projects.

Patients aged 2-18 years of age were diagnosed by a positive sweat chloride (>60 mmol/L) test and/or a known genotype with compatible symptoms. *P. aeruginosa* colonization was defined as positivity for *P. aeruginosa* in at least four sputum samples in the prior 12 months and with more than a 50% positive growth of *P. aeruginosa*. The present study included clinically stable CF patients. Demographic information and laboratory data were recorded from the medical files.

The Shwachman-Kulczycki score was used for the evaluation of the clinical status of the patients. The score was calculated via the general activity, physical examination, nutritional status, and chest X-ray findings. Overall, 86 to 100 points were classified as "excellent", 71 to 85 as "good", 56 to 70 as "mild", 41 to 55 as "moderate", and ≤40 as "severe" (8).

Laboratory Evaluation

Whole Blood Count Assay

Whole blood count, leukocyte counts, absolute neutrophil and lymphocyte counts, and relative ratio were performed with a hemocounter Cell-Dyn 3700, Abbott Diagnostics, USA.

Serum Immunoglobulin Assay

Serum immunoglobulins (IgG, IgA, IgM) were analyzed quantitatively via a Dade-Behring BN II Nephelometer, Siemens, Germany, and compared with age-related normal levels (9).

CD11a, CD18 and CD15S Surface Expressions on Neutrophil Granulocytes and Monocytes

Surface expressions of CD11a, CD18, and CD 15s on monocytes and neutrophil granulocytes were analyzed by cell surface staining instructions using anti-CD11a FITC, anti-

CD18 PE moAb and HuCD15SBV510 (Becton-Dickinson, USA) in heparinized blood samples. Leukocytes were also analyzed for their intracellular expression of myeloperoxidase (MPO) by flow cytometry.

Chemotactic Function of Granulocytes (Migratest)

The Migratest (Orpegen Pharma, Heidelberg, Germany) was used in order to evaluate the number of neutrophils which migrated through the cell culture toward chemoattractant fMLP. A percentage of activated granulocytes of more than 95% was accepted as normal. The chemotaxis index was calculated as follows;

Chemotaxis index (10): Д: A-B

A=Neutrophil count which migrated after FMLP/Beat

B=Neutrophil count which migrated before stimulation/ Beat

Chemotaxis ratio (11): Neutrophil count which migrated after FMLP/Neutrophil count which migrated before stimulation. A ratio greater than 1 was accepted as normal.

Oxidative Burst Activity of Granulocytes (Phagoburst Test)

The Phagoburst commercial test kit (Orpegen Pharma, Heidelberg, Germany) was used for oxidative burst activity of monocytes and granulocytes in heparinized whole blood. Stimulants such as *Escherichia coli* (*E. coli*), *n*-formyl-methionine-leucine-phenylalanine (fMLP), and phorbol-12-myristate-13-acetate (PMA) were used to produce reactive oxidants and the percentage of phagocytic activity was determined by flow cytometry (Normal reference ranges; for monocytes by stimulant *E. coli*: 70-100%; for granulocytes by stimulant *E. coli*: 95-100%, fMLP: 1-20%, and PMA: 99-100%).

Statistical Analysis

The data were evaluated using the Statistical Package for Social Sciences 25.0 (SPSS for Windows 25.0, Inc., Chicago, IL, USA) and by analyzing descriptive statistics (means, standard deviation). Student's t-test was used for the comparison of normally distributed variables. The chi-square, Mann-Whitney U test, and Kruskal-Wallis tests were used for non-normally distributed variables. A p-value <0.05 was considered as significant. Recurrent measurements of variant analysis were used for data during chemotaxis (measurements before and after fMLP).

Results

Patient Characteristics

A total of 26 CF patients [Male/Female (M/F): 14/12] with a mean age of 127.85±56.06 months and 21 healthy controls (M/F: 12/9) with a mean age of 122.07±70.2 months were included in the present study. There was no statistical significance for gender or age between the groups. All CF patients had bronchiectasis. All of the demographic and clinical characteristics of the patient group are summarized in Table I. In terms of CTFR mutations: nine (34.6%) patients were homozygous for F508, 10 (38.4%) were homozygous for other mutations and 7 were compound heterozygous.

Laboratory Data

White-blood cell count and absolute neutrophil count were significantly increased in the patient group ($p<0.01$, respectively). Although five of the patients had hypogammaglobulinemia, mean IgG, IgA and IgM levels were significantly higher in the patient group ($p<0.01$). Among those patients with low IgG, the other immunoglobulin levels and lymphocyte subsets were normal and they had normal vaccine responses. There was no finding suggesting primary antibody deficiency accompanying CF. There were no differences for C-reactive protein (CRP) level or absolute lymphocyte count between the groups (Table II).

Table I. Demographic and clinical characteristics of cystic fibrosis patients

	n=26 (%)
Age (months) (median) (IQR) (min.-max.)	105.0 (143.2) (24-216)
Age at diagnosis (months) (median) (IQR) (min.-max.)	3 (9.0) (1-192)
Age at symptom onset (months) (median) (IQR) (min.-max.)	4 (1.0) (1-12)
Follow-up period (months) (median) (IQR) (min.-max.)	86 (75.7) (17-216)
IRT Level (ng/ml) (mean ± SD)	185.33±73.36
Sweat test level (Cl) (mmol/L) (mean ± SD)	77.66±17.21
History of consanguinity	15 (57.7%)
Number of hospitalizations per year (median) (min.-max.)	7 (13) (0-48)
Failure to thrive	24 (92.3%)
Concomitant colonization of *S. aureus*	11 (42.3%)
Presence of ABPA	4 (15.3%)

IQR: Interquartile range, min.-max.: Minimum-maximum, ABPA: Allergic bronchopulmonary aspergillosis

The adhesion molecule percentages (CD11A, CD18, CD15s on neutrophils and monocytes) were similar for the patient and control groups except for CD15s levels on neutrophils, which were higher in the patient group (p=0.01) (Table III).

The chemotaxis ratio was <1% in seven (26.9%) cases in the patient group and one (4.8%) in the control group. There was no difference between the patient and control groups in terms of low chemotaxis ratios. The case with the low chemotaxis ratio in the control group had no addition pathology and no history of acute infection or recent drug usage. Three of the four patients (75%) with allergic bronchopulmonary aspergillosis (ABPA) had a ratio <1, which was significantly higher than in those patients without ABPA (p=0.04). Among the three patients with ABPA, two of them had received azithromycin treatment. There was no difference for body-mass index in those patients with and those without ABPA. Although hospitalization rates per year were higher (5.6 per year) in those patients with low chemotaxis ratios, there was no significant difference when compared to those patients with normal ratios. The comparison of the patients with decreased/normal chemotaxis ratios is given in Table IV.

The chemotaxis index was significantly lower in those patients with a CTFR mutation homozygous for 2183AA>G than for the others (p=0.01).

According to recurrent measurement variant analysis during chemotaxis, there was a significant increment in chemotactic cells between before and after stimulation in both groups (p<0.01), but there was no difference between the groups (p>0.05).

There was no significant relationship between the frequency of hospitalization and the annual number of active infections, and the IgG level and chemotaxis index (p>0.05).

While phagocytic activity in response to PMA was lower in one CF patient and in one control, there was no significant difference between the groups. Also, no difference was found for phagocytic activity in response to *E. coli*. Myeloperoxidase levels were also similar (p>0.05) (Table III).

Discussion

Patients with CF are susceptible to recurrent infections from bacteria, viruses, and fungi due to abnormally thick, sticky mucus trapping these germs in the airways. Additionally, immunologic abnormalities, such as decreased lymphocyte responsiveness, defects in opsonic activity and increased circulating immune complexes, predispose these individuals to recurrent and invasive infections and ongoing inflammations. In the present study, phagocytosis and chemotaxis abilities, which are important functions of neutrophils, were investigated by flow cytometry. No evidence for defective phagocytosis or chemotaxis were detected in those patients with CF. The present study only found an increased number of neutrophils and immunoglobulin levels in the CF patients. The increase in neutrophil and Ig counts was evaluated as a compensatory response to chronic infection and inflammation. Those patients with ABPA had significantly decreased neutrophil chemotaxis ratios, and the patients

Table II. Hemogram and immunoglobulin levels of the study groups

	Patients, n (26)	**Control, n (21)**	**p value**
White-blood cell/mm3 (mean ± SD)	11170±3938	7422.8±1677.7	<0.01
Absolute neutrophil count (mean ± SD) Low ANC Normal ANC	6184.3±3119.2 0 26 (100%)	3668.5±1151.9 1 (4.7%) 20 (95.2%)	<0.01
Absolute lymphocyte count (median) (IQR) (min.-max.)	2920 (2690) (1790-9540)	2800 (1200) (1000-4100)	0.27
IgG (mean ± SD) (mg/dL)	1173.4±481.6	759.4±143.8	<0.01
IgG values of patients Low IgG Normal IgG High IgG	 (19.2%) 13 (50%) 8 (30.7%)	 1 (4.7%) 20 (95.2%) -	<0.01
IgM (mg/dL)	120 (93) (59.7-852.0)	80.0 (45.1) (50.2-130.9)	<0.01
IgA (mg/dL)	198.4±118.7	89.8±27.0	<0.01
IgE kU/L	46.9 (167.8) (17-1410)	41.3 (108.0) (17-810)	0.64
CRP (mg/L)	3.3 (14) (0.3-31.5)	3.0 (1) (1-15)	0.45

IQR: Interquartile range, min.-max.: Minimum-maximum, SD: Standard deviation, ANC: Absolute neutrophil counts, Ig: Immunoglobulin, CRP: C-reactive protein

Table III. Comparison of adhesion molecule/burst test/MPO/L-Selectin levels of study groups

	Patients (n=26)	Control (n=21)	p value
CD11A (monocyte) %	99 (76-100)	99 (90-100)	0.31
CD11A (neutrophil) %	100	99.7	0.26
CD15S (monocyte) %	83 (23-98)	85 (75-90)	0.22
CD15S (neutrophil) %	100 (74-100)	99 (82-100)	0.01
CD18 (monocyte) %	99 (83-100)	99 (95-100)	0.23
CD18 (neutrophil) %	100 (100-100)	100 (95-100)	0.11
Burst test - PMA - *E. coli*	 99.8±0.47 97.6±2.81	 99.6±0.74 97.6±1.71	 0.15 0.28
MPO (monocyte)	83.0±8.33	83.33±7.33	0.10
MPO (neutrophil)	98.6 ±1.43	99.0±1.94	0.06
L-Selectin-Migratest	95.2±5.4	92.5±6.3	0.06

PMA: Phorbol-12-myristate-13-acetate, MPO: Myeloperoxidase

Table IV. The comparison of the patients with decreased/normal chemotaxis ratio

	Decreased CR, n (7)	Normal CR, n (19)	p value
Gender (F/M)	4/3	8/11	0.66
F508 Homozygous	2	7	0.99
ABPA	3 (42.9)	1 (5.3)	0.04
Age at pseud. colonization	89.0±70.7	73.1±57.2	0.58
Age at first pulmonary attack	80.0±67.3	79.3±53.5	0.98
Azithromycin	3 (42.9)	3 (15.7)	0.29
Hospitalization number/age (per year)	5.6 (9.4) (0-12)	0.6 (1.2) (0-8)	0.18
Sweat Test Cl (mmol/L)	85.6±16.9	98.5±26.9	0.32
Shwachman-Kulczycki score	10 (9) (6-19)	11 (2) (5-20)	0.69
BMI	13.6 (5.8) (8.0-18.9)	15 (3.1) (1.0-17.8)	0.49
White blood cell (mean ± SD)	12,965.0±5,041.7	10,508.4±3,368.9	0.16
Absolute neutrophil count (median) (IQR) (min.-max.)	5,040 (7,590) (3,870-12,630)	4,980 (4,840) (2,780-114,940)	0.33
Absolute lymphocyte count (median) (IQR) (min.-max.)	2,950 (3,550) (1,790-9,540)	2,865 (2,760) (2,760-8,630)	0.91
IgG (mean ± SD)	1,135.4±391.4	1,190.1±527.2	0.80
IgM	117.0 (106.0) (104.0-229.0)	119.0 (84.5) (59.7-288)	0.53

F/M: Female/Male, IQR: Interquartile range, min.-max.: Minimum-maximum, SD: Standard deviation, Ig: Immunoglobulin, BMI: Body mass index, ABPA: Allergic bronchopulmonary aspergillosis

with a mutation of 2183AA>G had significantly lower chemotaxis indexes.

CF is a well-known neutrophil dominant inflammatory disease. There are numerous studies showing neutrophilia in the airways of these patients even in the absence of infection (3,12). Spontaneous apoptosis of neutrophils is a necessary event which protects the host from the harmful effects of inflammation. It has been shown in CF patients that they have defective neutrophil apoptosis leading to long survival of neutrophils. Gray et al. (13) studied blood neutrophil apoptosis by flow cytometry in CF piglets and CF patients. They showed that, because of decreased apoptosis, the life of CF neutrophils was longer. They also showed neutrophil extracellular trap (NET) formation (NETosis), which helps bacterial killing, was excessive in CF patients causing more inflammation in the airways. The present study found an increased number of neutrophils in the absence of active

infection (CRP levels were similar in the CF and control groups), which is consistent with the literature.

Serum immunoglobulin levels were high among the patients, especially those with the mutation F508. One study reported hypergammaglobulinemia among CF patients and commented that this was a result of chronic infections and inflammation, and that it was related with poor prognosis (14). A recent study found Hypo-IgG levels among 66 CF children with a low number of pulmonary exacerbations and duration of antibiotic therapy (15). Five of our patients with higher rates of hospitalization had hypogammaglobulinemia, but this difference was not significant. The total protein/albumin levels were normal, and there was no malabsorption or protein lose that could explain hypogammaglobulinemia in these patients. Their body-mass index was low but there was no difference from those patients with normal globulin levels. There was no other finding such as inadequate vaccine responses which would suggest primary antibody deficiency. In addition, two patients with initial hypogamaglobulinemia showed gradual IgG elevation during follow-up. It was suspected that the chronic inflammation might be the cause of transient hypogammaglobulinemia. In recent years, studies have reported on the upregulation of mucosal IgA and its receptor, pIgR, as well as serum IgA (16). Also, a strong upregulation of IgA and pIgR was achieved after pseudomonas infection (17).

Similar to the present study, Leuer et al. (18) found no inability of phagocytosis both in neutrophils and monocytes in 35 CF adult patients in comparison to 12 healthy controls. In contrast to our findings, Aslanhan et al. (11), who compared the neutrophil functions of CF patients with pseudomonas colonized (n=8), non-colonized (n=8) and a control group (n=8), found increased phagocytosis, oxidative burst and chemotaxis in the CF patients. They reported that, although the phagocytosis capacity in peripheral blood is more prominent, there was a failed elimination of the colonized bacteria in the lung bronchi.

Three of the four patients (75%) with ABPA in our study had a chemotactic ratio below <%1, which was significantly higher than those patients with normal ratios. ABPA, which is described as hypersensitivity to aspergillus fumigatus, is seen in nearly 10% of patients with CF. Low body-mass index, long-term medication with macrolides, and prophylactic antibiotics for colonization with *Staphylococcus aureus* and *P. aeruginosa* have been reported as risk factors for ABPA (19,20). It has been reported that macrolide treatment has immunomodulatory effects in CF patients; the inhibition of neutrophil chemotaxis to the airways and the production of chemotactic factors (IL8 and LTB4) (21,22). Among our three patients with low chemotactic ratios (<1) with ABPA, two of them used macrolide treatment. As the patient number was very low in the present study, it was not possible to propose that macrolides play a role in neutrophil chemotaxis. *In vitro* functional studies are needed to prove the inhibitory role of macrolides on neutrophil chemotaxis.

Hardisty et al. (23) reported that ivacaftor treatment improved neutrophil markers of adhesion and activation in patients with R117H residual function CFTR mutations. Bratcher et al. (24) showed that ivacaftor normalized CD11b on neutrophils and CD63 on monocytes in patients with mutations of G551D. Therefore, it can be said that modulator therapies achieve mutation specific leucocyte modulation. We found that the chemotaxis index was significantly lower in those patients with a mutation of 2183AA>G than in the others. There might be differences of neutrophil functions depending on the mutations and manifestations of the disease.

Integrins and selectins play a role in leukocyte transmigration to the inflammation area. Sorio et al. (6) reported diminished activation of β1 and β2 integrins and chemotaxis in the mononuclear cells of CF patients. They reported that neutrophil adhesion and chemotaxis was normal, and defined CF as a monocyte-selective adhesion deficiency. The present study found no differences for adhesion molecules, CD11B and CD18, between the CF and control groups on either neutrophils or monocyte. We only found increased levels of CD15s on neutrophils. CD15s is a ligand for selectins playing a role in neutrophil aggregation and slow rolling (25). Markic et al. (26) reported CRP, procalcitonin and CD15s percentages to be predictors of severe bacterial infection. Although our patients did not have acute infection at the time of the study, chronic inflammation in CF patients could be the reason.

Study Limitations

The inclusion of more patients, both pseudomonas colonized and non-colonized, the analysis of the same patient in both their stable and active infectious periods, and performing Netosis assays and functional analyses on the phagocytic cells obtained from bronchoalveolar lavage material could provide more meaningful results. Unfortunately, only basic tests could be carried out with the available project budget on a limited number of patients.

Conclusion

In conclusion, although our data showed no evidence for defective phagocytosis or chemotaxis in those patients with CF, we found decreased chemotactic ability in those patients with ABPA and mutations of 2183AA>G. Our results might provide an insight into the neutrophil function differentiation of different mutations. Since our study had a small sample size, further studies involving more CF patients are needed to make a definitive interpretation.

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Ethics

Ethics Committee Approval: This study was approved by the Ege University Faculty of Medicine Medical Research Ethics Committee (approval no.: 20-12.1T/37, date: 17.12.2020).

Informed Consent: Informed consent was obtained from all patients and parents.

Authorship Contributions

Surgical and Medical Practices: M.B., F.G., E.D., N.E.K., Concept: H.D.Ş., M.B., G.A., N.K., E.D., N.E.K., Design: H.D.Ş., E.T., F.G., N.K., E.D., N.E.K., Data Collection and/or Processing: H.D.Ş., M.B., E.T., N.K., Analysis and/or Interpretation: H.D.Ş., E.T., G.A., E.D., N.E.K., Literature Search: E.T., F.G., G.A., Writing: H.D.Ş., F.G., G.A., N.K., E.D.**Conflict of Interest:** The authors declare that there is no conflict of interest regarding the publication of this article.

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References

1. Riordan JR, Rommens JM, Kerem B et al. Identification of the cystic fibrosis gene: cloning and characterization of complementary DNA. Science 1989; 245:1066-73.
2. Tangye SG, Al-Herz W, Bousfiha A, et al. Human Inborn Errors of Immunity: 2019 Update on the Classification from the International Union of Immunological Societies Expert Committee. J Clin Immunol 2020;40:24-64.
3. Balough K, McCubbin M, Weinberger M, Smits W, Ahrens R, Fick R. The relationship between infection and inflammation in the early stages of lung disease from cystic fibrosis. Pediatr Pulmonol 1995;20:63-70.
4. Kelly E, Greene CM, McElvaney NG. Targeting neutrophil elastase in cystic fibrosis. Expert Opin Ther Targets 2008; 12:145-57.
5. De Rose V, Oliva A, Messore B, Grosso B, Mollar C, Pozzi E. Circulating adhesion molecules in cystic fibrosis. Am J Respir Crit Care Med 1998; 157:1234-9.
6. Sorio C, Montresor A, Bolomini-Vittori M, et al. Mutations of Cystic Fibrosis Transmembrane Conductance Regulator Gene Cause a Monocyte-Selective Adhesion Deficiency. Am J Respir Crit Care Med 2016; 193:1123-33.
7. Govan JR, Brown AR, Jones AM. Evolving epidemiology of Pseudomonas aeruginosa and the Burkholderia cepacia complex in cystic fibrosis lung infection. Future Microbiol 2007; 2:153-64.
8. SHWACHMAN H, KULCZYCKI LL. Long-term study of one hundred five patients with cystic fibrosis; studies made over a five- to fourteen-year period. AMA J Dis Child 1958; 96:6-15.
9. Aksu G, Genel F, Koturoğlu G, Kurugöl Z, Kütükçüler N. Serum immunoglobulin (IgG, IgM, IgA) and IgG subclass concentrations in healthy children: a study using nephelometric technique. Turk J Pediatr 2006; 48:19-24.
10. Hertwig L. The implication of natural killer cells and neutrophils in autoimmune disorders of the central nervous system. 2016.
11. Aslanhan U, Cakir E, Pur Ozyigit L, et al. Pseudomonas aeruginosa colonization in cystic fibrosis: Impact on neutrophil functions and cytokine secretion capacity. Pediatr Pulmonol 2021; 56:1504-13.
12. Khan TZ, Wagener JS, Bost T, Martinez J, Accurso FJ, Riches DW. Early pulmonary inflammation in infants with cystic fibrosis. Am J Respir Crit Care Med 1995; 151:1075-82.
13. Gray RD, Hardisty G, Regan KH, et al. Delayed neutrophil apoptosis enhances NET formation in cystic fibrosis. Thorax 2018; 73:134-44.
14. Garside JP, Kerrin DP, Brownlee KG, Gooi HC, Taylor JM, Conway SP. Immunoglobulin and IgG subclass levels in a regional pediatric cystic fibrosis clinic. Pediatr Pulmonol 2005; 39:135-40.
15. Hanssens LS, Cellauro S, Duchateau J, Casimir GJ. Immunoglobulin G: A useful outcome marker in the follow-up of cystic fibrosis patients? Immun Inflamm Dis 2021; 9:608-14.
16. Gohy S, Moeremans A, Pilette C, Collin A. Immunoglobulin A Mucosal Immunity and Altered Respiratory Epithelium in Cystic Fibrosis. Cells 2021; 10:3603.
17. Collin AM, Lecocq M, Noel S, et al. Lung immunoglobulin A immunity dysregulation in cystic fibrosis. EBioMedicine 2020; 60:102974.
18. Leuer L, Krill A, Wilkens H, et al. The Phagocytosis of Blood Leukocytes from Cystic Fibrosis Patients is not Impaired in General. Lung 2020; 198:235-9.
19. Jubin V, Ranque S, Stremler Le Bel N, Sarles J, Dubus JC. Risk factors for Aspergillus colonization and allergic bronchopulmonary aspergillosis in children with cystic fibrosis. Pediatr Pulmonol 2010; 45:764-71.
20. Bargon J, Dauletbaev N, Köhler B, Wolf M, Posselt HG, Wagner TO. Prophylactic antibiotic therapy is associated with an increased prevalence of Aspergillus colonization in adult cystic fibrosis patients. Respir Med 1999; 93:835-8.
21. Tamaoki J, Kadota J, Takizawa H. Clinical implications of the immunomodulatory effects of macrolides. Am J Med 2004; 117 Suppl 9A:5S-11S.

22. Tsai WC, Rodriguez ML, Young KS, et al. Azithromycin blocks neutrophil recruitment in Pseudomonas endobronchial infection. Am J Respir Crit Care Med 2004; 170:1331-9.

23. Hardisty GR, Law SM, Carter S, et al. Ivacaftor modifies cystic fibrosis neutrophil phenotype in subjects with R117H residual function CFTR mutations. Eur Respir J 2021; 57:2002161.

24. Bratcher PE, Rowe SM, Reeves G, et al. Alterations in blood leukocytes of G551D-bearing cystic fibrosis patients undergoing treatment with ivacaftor. J Cyst Fibros 2016; 15:67-73.

25. Mulligan MS, Warner RL, Rittershaus CW, et al. Endothelial targeting and enhanced antiinflammatory effects of complement inhibitors possessing sialyl Lewisx moieties. J Immunol 1999; 162:4952-9.

26. Markic J, Jeroncic A, Polancec D, et al. CD15s is a potential biomarker of serious bacterial infection in infants admitted to hospital. Eur J Pediatr 2013; 172:1363-9.

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Original Article

Management of Ewing Family of Tumors Arising from Chest Wall in Children

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ABSTRACT

Aim: Malignant chest wall tumors are rare in children. We aimed to study the management of the Ewing family of tumors (EFT) in the chest walls of children.

Materials and Methods: The files of patients diagnosed with primitive neuro-ectodermal tumor and Ewing sarcoma of the chest wall in the prior 2 decades were retrospectively reviewed.

Results: A total of 15 children with a median age of 10 (8-14) years were included. Their symptoms were palpable mass (n=6), chest pain (n=4), B symptoms (n=2), cough (n=2) and abdominal pain. Their history revealed delayed diagnoses in 1/3 of the patients due to a misinterpretation of the symptoms. All children except for one had tru-cut biopsy and upfront chemotherapy. The median greatest diameter of mass at diagnosis was 100 (67-148) mm and 51 (39-100) mm at preoperative imaging after chemotherapy. Pleural effusion (n=8), costal destruction (n=9), extension to neural foramina (n=3), pulmonary or diaphragmatic nodule (n=5) and distant bone metastasis (n=4) were present at diagnosis in some cases. More than one surgery was performed in 9 (60%) of the children in order to remove the primary tumor and metastases. Costal excision (n=11), diaphragmatic resection (n=6) and chest wall reconstruction with graft material were performed on 8 patients. All cases had radiotherapy postoperatively. Mortality occurred in 9 (60%) patients due to local recurrences or metastases. Only 3 children reached 5-year survival and were considered to be cured. The remaining 3 patients completed their first line oncologic treatments and were under surveillance without treatment.

Conclusion: The EFT in the chest wall are aggressive tumors with poor prognosis despite multimodality treatment. Surgeons dealing with these patients should be familiar with complex chest wall reconstruction techniques.

Keywords: PNET, Ewing sarcoma, chest wall, child

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Introduction

Chest wall tumors are rare in childhood, comprising only 2% of all childhood malignancies. Most of them are soft tissue sarcomas or tumors arising from bony or cartilaginous tissues of the chest wall (1). Peripheral primitive neuro-ectodermal tumor (PNET) and Ewing sarcoma are very similar tumors in terms of their histology, biology and clinical presentation and they are collectively known as the Ewing family of tumors (EFT) (2). Most of the tumors arising from the chest wall in childhood belong to this family of tumors. They show aggressive and infiltrative behavior but are also chemo- and radio-sensitive (3). Treatment is multidisciplinary, which is similar to most of the pediatric solid tumors. Neoadjuvant chemotherapy followed by surgery and radiotherapy is the standard treatment approach. Recurrences and metastasis are common in these patients and they are managed by second line treatments and palliative interventions. The huge diameters and infiltrative growth patterns of these tumors demand extensive resections and reconstructive surgical applications. In this study, we aimed to present our experiences with this rare tumor of childhood from a pediatric surgical oncology standpoint.

Materials and Methods

This study covered patients admitted to a pediatric surgery clinic during the previous 2 decades with a diagnosis of PNET and Ewing sarcoma. The patient list was derived from the institutional tumor registry of the hospital electronic database. The selected patient files were reviewed retrospectively for age, gender, presenting symptoms, computed tomography reports, surgical details and pathology results. All cases in this cohort were evaluated, treated and followed by a team of physicians from the pediatric surgery, radiology, pathology and oncology departments which are dedicated to the care of pediatric oncology and oncologic surgery patients. The management algorithm is as follows: tru-cut biopsy followed by 6 cycles of induction chemotherapy, surgical excision and adjuvant chemotherapy and radiotherapy. The patients in this study were treated according to the European Ewing regimen, specifically Arm A, which consists of VIDE (vincristine, ifosfamide, doxorubicin, and etoposide) induction therapy followed by VAC (vincristine, actinomycin D, and cyclophosphamide) consolidation therapy.

This study was approved by the Hacettepe University Non-interventional Clinical Research Ethics Board (approval no.: 2022/22-10, date: 27.12.2022).

Statistical Analysis

The statistical analyses were performed with SPSS software (version 22.0). Two-tailed p-values <0.05 were considered statistically significant. Continuous variables are presented as median (interquartile range-IQR) because of their non-normal distribution.

Results

A total of 15 children (10 female, 5 male) were diagnosed with PNET/Ewing sarcoma of the chest wall during the study period. The median age of the patients was 10 (8-14) years. The presenting symptoms were palpable mass in 6 cases, chest pain in 4, B symptoms in 2, cough in 2 and abdominal pain in one patient. History revealed delayed diagnosis in 1/3 of the patients due to misinterpretations of the symptoms. At the time of diagnosis, 12 patients had positron emission tomography computed tomography which showed long bone metastasis in 3, lymph node metastasis in one and bone marrow infiltration in another patient. Fluorodeoxyglucose uptake limited to the primary tumor was the case in 7 patients. Median greatest diameter of mass at diagnosis was 100 (67-148) mm and 51 (39-100) mm at preoperative imaging after chemotherapy. The mass was located posteriorly in 8, laterally in 6 and anteriorly in one patient (Table I). Imaging studies revealed costal destruction in 9 cases (Figure 1), pleural effusion in 8, pulmonary or diaphragmatic involvement in 5 and extension to neural foramina in 3 patients. All of the children except one had tru-cut biopsy at the time of diagnosis and received upfront chemotherapy. More than one surgery was performed on 9 (60%) children in order to remove the primary tumor, its metastases or recurrences. Costal resection was carried out in 11 patients and the median number of resected ribs was 3 (1-3). The chest wall was reconstructed with ePTFE graft material in 8 out of the 11 patients who had more than one costal excision. To ensure en bloc resection, pulmonary tissue adjacent to the tumor was resected in 8 cases and diaphragmatic resection was performed in 6 cases. In all cases, gross total resection without macroscopic residue was achieved. The postoperative period was uneventful and the median hospital stay was 7 (6-10) days. Pathologic examination of the resected tumor showed diffuse histologic changes secondary to treatment in 11 patients and >95% necrosis in 5 individuals. Surgical margins were positive in 10 patients. FISH study was positive for Ewing sarcoma breakpoint region 1 (EWSR1) translocation in 10 out of 11 patients.

Table I. Clinical characteristics of patients at the time of diagnosis	
Age (median, years)	**10 (8-14)**
Sex	
Male	5
Female	10
Side	
Right	7
Left	8
Location	
Anterior	1
Posterior	8
Lateral	6
Presenting symptom	
Palpable mass	6
Chest pain	4
B symptoms	2
Cough	2
Abdominal pain	1
Metastasis	
Pulmonary nodule	4
Bone	3
Bone marrow	1
Lymph node	1

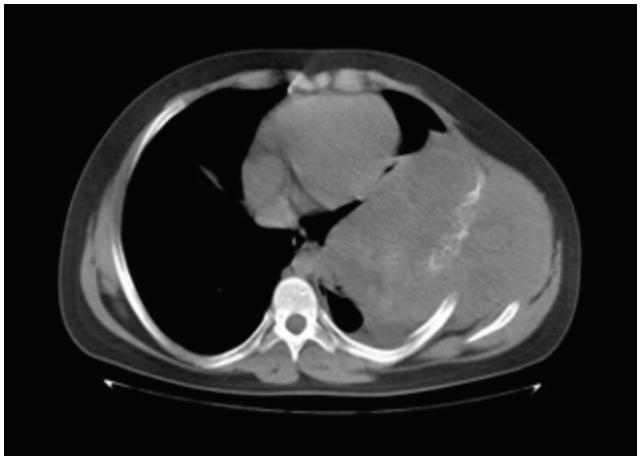

Figure 1. 14-year-old boy with PNET. Axial non-contrast computed tomography scan demonstrates an extra-pleural giant heterogeneous solid mass causing osseous destruction of the left lateral 6th rib. The mass extends into the left chest wall and hemithorax

All cases received radiotherapy and chemotherapy in the postoperative period. The mortality rate was 60%. The cause of death was distant metastasis in 5, local recurrences

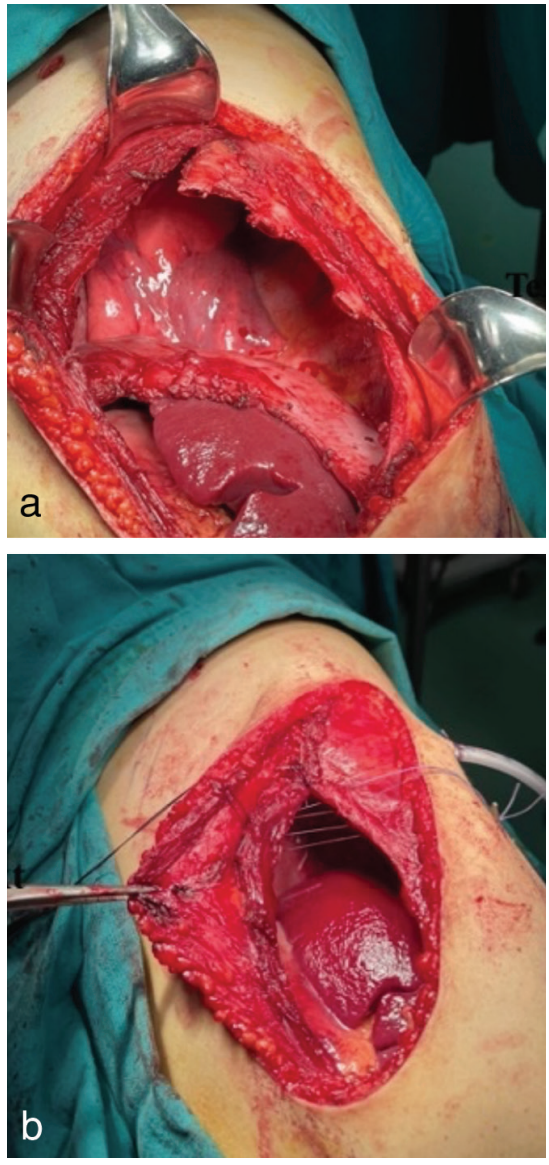

Figure 2. Intraoperative image of the chest wall tumor resection which infiltrates the lower ribs, intercostal muscles and diaphragm **(a)** and primary reconstruction **(b)**

in 3 and secondary leukemia in one patient. Only 3 children reached 5-year survival and were considered to be cured. The remaining 3 patients completed their treatment and were under close surveillance. There were no long-term complications such as scoliosis or posture abnormality in those patients who survived the disease.

Discussion

Malignant tumors arising from the chest wall are very rare in childhood and Ewing sarcoma/PNET is the most common diagnosis in these patients. They arise from bony structures, muscles or connective tissue but tend to grow

in an infiltrative pattern affecting various tissues. The main surgical aim should be total resection without macroscopic residue as in all cases of sarcomas (1). Due to these features, surgery involves the resection of different tissues and the reconstruction of a functioning chest wall. Experience is limited in this patient group since they constitute only 2% of childhood malignancies (4). We reviewed our experiences in the management of EFT arising from the chest wall in children.

EFT at the time of diagnosis are usually huge in diameter with costal, pleural, muscular, adjacent pulmonary or diaphragmatic invasion and extension to spinal foramen. Surgical resection should be deferred until after the induction chemotherapy (5). Treatment-induced size reduction may allow for less extensive resection and a higher rate of negative margins. A fifty percent reduction in the greatest diameter was achieved with neoadjuvant chemotherapy in our patients. This finding was compatible with previous data (2,4). In addition, chemotherapy should be employed upfront to treat occult metastatic foci and circulating tumor cells (3).

Surgical resection and the reconstruction of the chest wall is a formidable challenge in children. Their growing body size and long life expectancy distinguish children from adults. There are various materials which can be employed to cover defects in the chest wall when we resect components of the chest wall neighboring a tumor. There are synthetic, biologic and metallic prosthetic materials which are used to cover defects in the chest wall and each have different properties (4-6). In cases of malignant tumor resection, the graft material should be compatible with radiotherapy, suitable for re-entrance for surgery since these tumors tend to recur and possess some amount of flexibility in order to adapt to a growing thoracic wall (5-7). We prefer ePTFE mesh in our practice. It integrates well into the thoracic wall showing minimal tissue reaction. Also, all patients had radiotherapy on the surgical field in the postoperative period. Scoliosis is a well-known long-term complication in patients with chest wall resection (8). Due to the low life expectancy and complete cure rates in our group, we could not deduce any statistically significant information regarding this issue.

The risk of local recurrence and distant metastasis remains high in the EFT despite advances in all aspects of oncologic treatments. Histologic response to neoadjuvant chemotherapy is an indicator of prognosis in these patients. Good histologic response is defined by the degree of necrosis and >95% is generally acceptable (3). We observed this amount of necrosis in only 1/3 of our patients and assumed that it was one of the major contributors to the high mortality rate in our cohort. Another issue is to achieve negative surgical margins in sarcoma patients (3). Even though aggressive surgical resection involving the full thickness of the chest wall, pulmonary and diaphragmatic resections, the surgical margins were positive microscopically in the majority of our patients which implies a high possibility of local recurrence. The posteriorly location of the tumor and a tumor diameter >10 cm at diagnosis are associated with poor prognosis (1,9). More than half of our patients had posteriorly located tumors and >10 cm diameter at the time of diagnosis. The largest study regarding the outcome in Ewing sarcoma and PNET including 3,575 patients derived from the SEER database revealed a 5-year survival rate of slightly above 50%. However, this cohort included cases of all anatomic locations, not just exclusive to the chest wall (2).

Study Limitations

Main limitation of our study was its small sample size. Multi-centric studies should be designed in order to recruit more patients in such rare tumors of children.

Conclusion

The EFT located in the chest wall are rare tumors in childhood with infiltrative growth patterns. Although they respond well to chemotherapy and radiotherapy, their clinical behavior is usually aggressive. With neoadjuvant chemotherapy, tumors shrink in most cases. Surgical management involves extensive resection of the chest wall components, lung and/or diaphragm and the reconstruction of the defect with graft material. Despite total resection, negative surgical margins are seldom reached. Local recurrences and metastasis are associated with poor prognosis. Multicenter studies should be planned in order to better understand the nature of this rare patient group.

Ethics

Ethics Committee Approval: This study was approved by the Hacettepe University Non-interventional Clinical Research Ethics Board (approval no.: 2022/22-10, date: 27.12.2022).

Informed Consent: Retrospective study.

Authorship Contributions

Surgical and Medical Practices: İ.R.U., B.A., F.Y., D.O., F.Ü., S.E., Concept: A.Ö.Ç., İ.K., Design: T.K., Data Collection and/or Processing: B.A., B.O., M.H., S.E., Analysis and/

or Interpretation: A.Ö.Ç., İ.K., B.O., M.H., D.O., F.Ü., S.E., Literature Search: İ.R.U., B.A., Writing: İ.R.U.

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References

1. Sandler G, Hayes-Jordan A. Chest wall reconstruction after tumor resection. Semin Pediatr Surg 2018; 27:200-6.
2. Campbell K, Shulman D, Janeway KA, DuBois SG. Comparison of epidemiology, clinical features, and outcomes of patients with reported ewing sarcoma and PNET over 40 years justifies current WHO classification and treatment approaches. Sarcoma 2018; 2018:1712964.
3. Stachelek GC, Ligon JA, Vogel J, et al. Predictors of recurrence and patterns of initial failure in localized ewing sarcoma: a contemporary 20-year experience. Sarcoma 2021; 2021:6681741.
4. Lopez C, Correa A, Vaporciyan A, Austin M, Rice D, Hayes-Jordan A. Outcomes of chest wall resections in pediatric sarcoma patients. J Pediatr Surg 2017; 52:109-14.
5. Maistry N, Durell J, Wilson S, Lakhoo K. Primary paediatric chest wall tumours necessitating surgical management. Ann R Coll Surg Engl 2020; 102:335-9.
6. Sanna S, Brandolini J, Pardolesi A, et al. Materials and techniques in chest wall reconstruction: a review. J Vis Surg 2017; 3:95.
7. Theodorou CM, Lawrence YS, Brown EG. Chest wall reconstruction in pediatric patients with chest wall tumors: a systematic review. J Pediatr Surg 2023; 58:1368-74.
8. Saltsman JA, Danzer E, Hammond WJ, et al. Survival and scoliosis following resection of chest wall tumors in children and adolescents: a single-center retrospective analysis. Ann Surg 2021; 274:e167-73.
9. Collaud S, Stork T, Dirksen U, et al. Surgical treatment for primary chest wall sarcoma: a single-institution study. J Surg Res 2021; 260:149-54.

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Original Article

Evaluation of Factors Affecting Surgical Success in Megameatus Intact Prepuce Cases

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ABSTRACT

Aim: Megameatus intact prepuce (MIP) is a rare form of hypospadias. Different meatal/urethral advancement and urethroplasty techniques are used in the treatment of MIP. This study aimed to evaluate the success of various surgical methods for the treatment of MIP.

Materials and Methods: Patients who underwent circumcision, meatoplasty, and urethroplasty techniques due to MIP between 2011 and 2022 were included in this study. Surgical success was accepted as the absence of complications and/or the need for additional treatment. The statistical significance level was accepted as 0.05.

Results: This study included 100 patients with a median age of 33 months. Of these, 94 patients were admitted to our center with untreated MIP, 5 presented after circumcision and 1 after MIP repair in another center. The urethral stent placement rate was significantly lower in those patients with a glanular meatus location as in patients who underwent the meatoplasty technique ($p<0.001$). The complication rate was significantly higher in the Duplay with Posterior Meatal Incision (DPMI) technique when compared to the other techniques ($p=0.033$). There were no significant differences between the meatoplasty, Duplay urethroplasty, Pyramid urethroplasty, and DPMI techniques in terms of the need for additional surgical intervention ($p=0.102$). None of the five previously circumcised patients who underwent Duplay urethroplasty experienced any complications. When the complication rates were compared between the patient group presenting with untreated MIP and those who underwent Duplay urethroplasty, no statistically significant difference was detected ($p=0.534$).

Conclusion: According to the conclusions of this MIP series, prior circumcision or the preference for any specific surgical technique that preserves the urethral plate did not affect the success of MIP treatment.

Keywords: Megameatus intact prepuce, hypospadias, circumcision, meatoplasty, urethroplasty

Introduction

Megameatus intact prepuce (MIP) is a rare concealed form of hypospadias, accounting for 3-5.2% of all cases (1,2). MIP differs from other types of hypospadias due to its anatomy, which includes an intact prepuce, a lack of ventral chordee, and a wide urethral plate. The cause of MIP is believed to be developmental deficiencies in ventral spongiotic tissue in the glanular part of the urethra or possible ischemia secondary to the compression of the glanular urethra (3-5).

The anatomic appearance of MIP has a wide spectrum. Depending on how wide and deep the glanular groove expands, the meatus may extend to the coronal or subcoronal area. While the frenulum may develop normally in cases where the meatus is located in the glans, it may not develop at all when the meatus is located more proximally. Since preputium development is complete, patients are mostly diagnosed during circumcision or after the age of 4-5 years, when preputial retraction is easier.

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MIP surgery is performed for the purpose of a proper and straight projection of urine and a normal cosmetic appearance. The surgical repair technique is determined according to the penile anatomy (meatus location, presence of frenulum, structure of glanular collars) and the surgeon's preference. Meatal/urethral advancement techniques and urethroplasty techniques [Duplay, Glans Approximation Procedure (GAP), Pyramid, and Tubularized Incised Plate (TIP)] are the most commonly used methods in the treatment of MIP (1,6-8).

Although the studies in the literature for the treatment of MIP have a high success rate, to date, there has been no study with a large number of patients investigating the superiority of these methods. This study aimed to evaluate the success of various surgical methods for the treatment of MIP.

Materials and Methods

The hospital records of those patients treated for MIP between 2011 and 2022 were evaluated retrospectively. The treatment median ages of the MIP patients were compared with the median ages of other types of distal hypospadias during the same period. Patients aged 0-17 years with intact preputium and megameatus were included in this study, while those with missing records were excluded.

Demographics, meatus localization, whether it was a primary surgery, circumcision status, curvature, surgical details (the technique used, degloving, urethral stenting), complications, and the need for postoperative interventions were examined. Surgical success was accepted as the absence of complications and/or the need for additional treatment.

Approval for the conduct of this study was obtained from the Medical Research Ethics Committee of Ege University Faculty of Medicine (approval no.: 23-3T/14, date: 09.03.2023). Written informed consent was obtained from the parents.

Definitions Regarding Surgical Management

Circumcision only: For those cases where the glans wings were not widely separated, the frenulum developed normally, and with glanular megameatus, which would not cause voiding disorders or cosmetic problems, only circumcision was performed with the consent of the families.

Meatoplasty: A meatoplasty and glansplasty customized for each patient's specific anatomy were performed for those patients who did not need urethroplasty but required some cosmetic amendment.

Urethroplasty: Duplay and Pyramid techniques were mainly used in those cases requiring urethroplasty. Among the patients who underwent Duplay urethroplasty, a modification of vertically incising the mucosal fold was applied to a subgroup with a mucosal septum extending between the meatus and the mucosal pit to ensure smooth voiding. This technique is referred to as Duplay with Posterior Meatal Incision (DPMI) in this article.

Statistical Analysis

Surgical success and complication rates were evaluated according to the meatus localization and the surgical method applied. The compatibility of the data to the normal distribution was evaluated using the Kolmogorov-Smirnov test. The Mann-Whitney U test was used to compare groups for numeric variables, and the chi-square test was used for categorical variables. SPSS version 23.0 (SPSS, Chicago, IL, USA) was used for statistical analysis. The statistical significance level was accepted as 0.05.

Results

There were 103 patients with MIP who were admitted to our department between 2011 and 2022, and of these, 100 were included in this study. Three patients who had previously undergone circumcision and did not require additional intervention were excluded from this study. The median age was 33 (12.9-88.8) months. The median age of 434 patients who were operated on for other types of distal hypospadias in our department during the same period was 19 (13-42) months. A statistically significant difference was found between the median treatment ages of both groups (p=0.008).

Among all, 26 (27.7%) patients with MIP were diagnosed during circumcision in our clinic, and 68 (72.3%) were referred from other centers with a diagnosis of MIP. Among those patients who were referred from different centers, 8 had undergone circumcision, and one had a failed repair before admission. No additional procedures were applied to 3 of the 8 patients who had been circumcised only, since they did not have any cosmetic or voiding problems. Duplay urethroplasty was applied to the other 5 patients and those with previous MIP repair. The surgical techniques applied to the 100 patients who were managed in our center are shown in Table I.

In our study, out of the 100 patients, 9 (9%) had mild curvature, which improved with degloving alone, and 2

(2%) had severe penile curvature (>30°) requiring additional penile plication. Among the 84 patients who underwent meatoplasty or urethroplasty, 65 received a urethral stent. Table II shows the frequency of urethral stent placement based on meatus location and the surgical technique applied. While the stent placement rates in those patients who underwent MIP repair were 100% in both the coronal and subcoronal meatus groups, this rate was 39.7% (23/58) in the glanular meatus group. The stent placement rates in the meatoplasty, Duplay urethroplasty, DPMI urethroplasty, and Pyramid urethroplasty groups were 21.7%, 97.3%, 100%, and 100%, respectively. Patients with a glanular meatus had significantly lower stent placement rates than those with meatus in other locations (p<0.001), as did those who underwent meatoplasty compared to other techniques (p<0.001).

The median postoperative follow-up time for all 100 patients was 6 (4-8) months. The complication rate in the 84 patients who underwent meatoplasty or urethroplasty was 8.3% (4 had fistula, and 3 had meatal stenosis). The three patients with meatal stenosis were successfully treated with topical betamethasone and dilatation, and no additional surgery was necessary. Five surgical interventions were performed among the four patients with fistulas (Table III). There were no significant differences between the meatoplasty, Duplay urethroplasty, Pyramid urethroplasty, and DPMI techniques in terms of the need for additional surgical intervention (p=0.102). No complications were observed in any patient who underwent meatoplasty or Pyramid urethroplasty. While the complication rate was 10.8% in those patients who underwent Duplay urethroplasty, it was 27.2% in those who underwent DPMI urethroplasty. The complication rate was significantly higher in those patients who underwent DPMI urethroplasty when compared to those who underwent the other procedures (meatoplasty, Duplay urethroplasty, Pyramid urethroplasty) (p=0.033). None of the five previously circumcised patients who underwent Duplay urethroplasty experienced any complications. No statistical difference was observed between the complication rates in those patients who presented with circumcised MIP (none of 5 patients) and those with uncircumcised MIP (4 out of 31 patients) who underwent Duplay urethroplasty (p=0.534).

Discussion

MIP, or megameatus intact prepuce, is a congenital abnormality that may lead to urinary problems and aesthetic dissatisfaction in boys. The severity of clinical symptoms

Table I. Distribution of surgical techniques by meatal localization

	Circumcision (n)	Meatoplasty (n)	Duplay urethroplasty (n)	DPMI urethroplasty (n)	Pyramid urethroplasty (n)
Glanular meatus (n=58)	16	23	11	2	6
Coronal meatus (n=25)	0	0	12	6	7
Subcoronal meatus (n=17)	0	0	14	3	0
Total number of patients (n=100)	16	23	37	11	13

DPMI: Duplay with posterior meatal incision, n: Number of patients

Table II. The frequency of urethral stent placement according to the meatus locations and the surgery techniques

	Meatoplasty n (%)	Duplay urethroplasty n (%)	DPMI urethroplasty n (%)	Pyramid urethroplasty n (%)	Total n (%)
Glanular meatus	5 (21.7)	10 (90.9)	2 (100)	6 (100)	23 (39.7)
Coronal meatus	0	12 (48)	6 (24)	7 (28)	25 (100)
Subcoronal meatus	0	14 (82.4)	3 (17.6)	0	17 (100)
Total number of patients	5 (21.7)	36 (97.3)	11 (100)	13 (100)	65 (65)

DPMI: Duplay with posterior meatal incision, n: Number of patients

Table III. Complications requiring additional treatment in patients operated for megameatus intact prepuce

	Meatal localization	Surgical technique	Complication	Additional treatment-1	Additional treatment-2
Patient 1	Subcoronal	Duplay	Meatal stenosis	Meatal dilatation + topical betamethasone	-
Patient 2	Subcoronal	Duplay	Fistula	Fistula repair	-
Patient 3	Glanular	DPMI	Meatal stenosis	Topical betamethasone	-
Patient 4	Coronal	Duplay	Meatal stenosis	Topical betamethasone	-
Patient 5	Glanular	Duplay	Fistula	Re-do repair	Fistula repair
Patient 6	Subcoronal	DPMI	Fistula	Fistula repair	-
Patient 7	Coronal	DPMI	Fistula	Fistula repair	-

DPMI: Duplay with posterior meatal incision

and the need for surgery depend on the configuration of the meatus and glans. In cases where the meatus is located on the glans and there is a frenulum, no extra intervention may be required other than circumcision, depending on family preference. However, other MIP cases may require meatoplasty or urethroplasty techniques, such as Duplay, Pyramid, GAP, and TIP. Studies have reported successful results for each of these methods, but the optimal technique remains unclear.

The literature suggests a rate of 6.5% for MIP among anterior hypospadias cases (9), but this was higher in our series (18.7%). Also, most of the patients were referred after evaluation for circumcision. We think this difference is probably secondary to being a referral center for hypospadias. This also shows that MIP cases are referred to our center, while surgeons in the surrounding hospitals commonly perform other distal hypospadias. This is reasonable because it is unclear whether corrective repair treatment is necessary for MIP, and no precise data shows the superiority of any technique among the various defined ones.

Zaontz, who defined the GAP technique, reported a urethrocutaneous fistula in one patient in his series of 24 patients (6). There were only two fistulas in three different studies regarding Pyramid urethroplasty, which covered a total of 60 patients (1,5,10). The complication rate of our patient series was consistent with the literature. It was observed that 3 out of 7 (8.3%) patients who developed complications were treated with DPMI, showing a significantly higher complication rate than the other three techniques. However, we do not believe this to be a conclusive result showing the inferiority of this technique as the choice of treatment is made according to the patient's anatomy, which may also determine surgical success.

In our study, the median treatment ages of MIP and other distal hypospadias cases operated on in the same time period were 33 months and 19 months, respectively. The median age of the MIP patients was significantly higher than the median age of the other distal hypospadias repairs (p=0.008). In a study conducted in our country, the median age of circumcision was reported as being 6 years (11). We attribute the significant age difference between the two groups in our study to the fact that infant circumcision is not widely performed in our country, and those patients with MIP are mostly diagnosed during circumcision when the prepuce is retracted.

Studies in the literature state that MIP is not associated with penile curvature or chordee (1,2,5). However, in a series of 118 patients, the overall penile curvature rate was reported as 24% (dorsal curvature was 19%, ventral curvature was 5%. The same study reported that ventral plication was required in 86% of cases with dorsal curvature (12). In our study, the penile curvature rate was 11%, and penile plication was required in 18.2% of those patients with penile curvature. Studies with a large patient series, such as our study, support the evaluation of penile curvature in MIP cases, which is a special form of distal hypospadias.

When MIP is diagnosed during circumcision, the common view is either to postpone the repair by consulting the patient's family or to repair it while the patient is under anesthesia, following family consent (10,13-16). However, it is also argued that the preputium and Dartos tissue are not necessary for MIP repair, and thus, circumcision can be performed (1,17-19). In our study, it was determined that the surgical success of MIP patients was not affected by circumcision. MIP repair was not required in any patient on whom we performed circumcision. No complications were observed in any patient who had been previously

circumcised and on whom we performed MIP repair. It was observed that previous circumcision did not change the repair technique applied in MIP cases (Duplay urethroplasty was performed for all five previously circumcised patients). MIP repair was not required in any patient in whom we performed circumcision. As in uncircumcised MIP patients, the choice of surgical technique was determined based on meatus localization, penile anatomy, and the surgeon's preference.

Study Limitations

The main limitation of our study was that it was based on a retrospective evaluation. Additionally, the surgical procedures were performed by multiple surgeons with varying experience. Surgical procedures were performed by multiple surgeons with varying experience. Another limitation of our study was the relatively short follow-up period. Since MIP patients include a wide anatomical spectrum, prospective studies are needed to record glans and meatus measurements, urethroplasty lengths, and curvature characteristics.

Conclusion

According to the results of our study, the complication rates, in the DPMI technique with mucosal incision were significantly higher than those of the other techniques. However, no difference was detected between the meatoplasty, Duplay urethroplasty, Pyramid urethroplasty and DPMI techniques in terms of the need for additional surgical intervention. In addition, in our series, it was observed that circumcision performed before repair in MIP patients did not change either the surgical technique or the complication rates, in contrast to popular belief. In conclusion, it can be inferred that prior circumcision or the preference for any specific surgical technique that preserves the urethral plate does not affect the success of MIP treatment.

Ethics

Ethics Committee Approval: Approval for the conduct of this study was obtained from the Medical Research Ethics Committee of Ege University Faculty of Medicine (approval no.: 23-3T/14, date: 09.03.2023).

Informed Consent: Written informed consent was obtained from the parents.

Authorship Contributions

Surgical and Medical Practices: U.B., A.T., S.T., İ.U., Concept: U.B., İ.U., Design: U.B., A.T., S.T., İ.U., Data Collection and/or Processing: U.B., A.T., Analysis and/or Interpretation: U.B., A.T., İ.U., Literature Search: U.B., Writing: U.B., A.T., S.T.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the publication of this article.

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References

1. Duckett JW, Keating MA. Technical challenge of the megameatus intact prepuce hypospadias variant: the pyramid procedure. J Urol 1989; 141:1407-9.
2. Cendron M. The Megameatus, Intact Prepuce Variant of Hypospadias: Use of the Inframeatal Vascularized Flap for Surgical Correction. Front Pediatr 2018; 6:55.
3. Hatch DA, Maizels M, Zaontz MR, Firlit CF. Hypospadias hidden by a complete prepuce. Surg Gynecol Obstet 1989; 169:233-4.
4. Borden TA, Rosen RT, Zuber R. Congenital megalourethra associated with hypospadias. Urology 1977; 9:307-9.
5. Nonomura K, Kakizaki H, Shimoda N, Koyama T, Murakumo M, Koyanagi T. Surgical repair of anterior hypospadias with fish-mouth meatus and intact prepuce based on anatomical characteristics. Eur Urol 1998; 34:368-71.
6. Zaontz MR. The GAP (glans approximation procedure) for glanular/coronal hypospadias. J Urol 1989; 141:359-61.
7. Bar-Yosef Y, Binyamini J, Mullerad M, Matzkin H, Ben-Chaim J. Megameatus intact prepuce hypospadias variant: application of tubularized incised plate urethroplasty. Urology 2005; 66:861-864.
8. Duan SX, Li J, Jiang X, et al. Diagnosis and Treatment of Hypospadias With Megameatus Intact Prepuce. Front Pediatr 2020; 8:128.
9. Juskiewenski S, Vaysse P, Guitard J, Moscovici J. Traitement des hypospadias antérieurs. Place de la balanoplastie [Treatment of anterior hypospadias. Place of balanoplasty]. Chir Pediatr 1983; 24:75-9.
10. Hill GA, Wacksman J, Lewis AG, Sheldon CA. The modified pyramid hypospadias procedure: repair of megameatus and deep glanular groove variants. J Urol 1993; 150:1208-11.
11. Sahin F, Beyazova U, Aktürk A. Attitudes and practices regarding circumcision in Turkey. Child Care Health Dev 2003; 29:275-80.
12. Ben-David R, Kupershmidt A, Dekalo S, et al. Dorsal penile curvature and megameatus intact prepuce hypospadias: A common association in a rare variant of hypospadias. J Pediatr Urol 2021; 17:517.e1-517.e4.
13. Docimo SG. Subcutaneous frenulum flap (SCUFF) for iatrogenic or primary megameatus and reoperative hypospadias repair. Urology 2001; 58:271-3.

14. Bhat A, Bhat M, Bhat A, Singh V. Results of tubularized urethral plate urethroplasty in Megameatus Intact Prepuce. Indian J Urol 2017; 33:315-8.

15. Wilcox DT, Mouriquand P. Hypospadias. In: Thomas D, Duffy PG, Rickwood A (eds). Essentials of Pediatric Urology, 1st ed. United Kingdom, Informa Healthcare, 2002; 213-31.

16. American Academy of Pediatrics: Report of the Task Force on Circumcision. Pediatrics 1989; 84:388-91.

17. Snodgrass WT, Khavari R. Prior circumcision does not complicate repair of hypospadias with an intact prepuce. J Urol 2006; 176:296-8.

18. Pieretti RV, Pieretti A, Pieretti-Vanmarcke R. Circumcised hypospadias. Pediatr Surg Int 2009; 25:53-5.

19. Chalmers D, Wiedel CA, Siparsky GL, Campbell JB, Wilcox DT. Discovery of hypospadias during newborn circumcision should not preclude completion of the procedure. J Pediatr 2014; 164:1171-4.