MEDICINE AND HEALTH Science

Modern Analysis, Finding and Researches

Editors
Serkan ARSLAN
Mustafa AZİZOĞLU

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Medicine and Health Sciences Modern Analysis, Finding and Researches

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PREFACE

Dear readers; We are proud to present to you, our valuable readers, our book, **"Medicine and Health Sciences Modern Analysis, Finding and Researches"**, which consists of 12 chapters in different categories, current and covering many health departments in the field of medicine and health in health sciences. This book has been written by academics who are experts in their field. In each section, you will be able to see up-to-date information on its own subject. In each chapter, the authors presented their theoretical knowledge on their subjects by blending a detailed literature knowledge and their own experiences. I believe that this information will be both a guide for scientists and surgeons who have taken a new step, and a foresight in terms of practical knowledge they can acquire on these issues. We would like to thank the coordinators, referees, authors and publishing house for their valuable contributions.

Assoc. Prof. Dr. Serkan ARSLAN
MD. Mustafa AZIZOGLU
Editors

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CHAPTER I

HYGIENE STATUS IN KITCHEN STAFF IN TURKEY: A REVIEW

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1. Introduction

The tourism sector is a sensitive sector that is affected by events such as epidemics, global economic crises and terrorism, and there are significant decreases in the number of tourists in destinations where epidemics occur and spread (1). With the latest COVID-19 epidemic worldwide, travel restrictions have been imposed to all destinations around the world as of March 2020, and international air transport has almost ended with the ban on entry and exit from countries. According to the United Nations World Tourism Organization (UNWTO) data, it was determined that international travels and tourist visits decreased by 87.0% compared to January 2021 and 2020 (2). In Turkey, it was determined that the number of foreign visitors decreased by 71.7% and the tourism income obtained by 65.1% in 2020 compared to 2019 (3). Although this situation negatively affects the tourism sector, it has ensured that businesses comply more carefully with the hygiene rules in order to provide a safe environment for tourists, and it has increased the level of awareness about providing a safe holiday opportunity by reducing the risk of contamination by considering the concept of social distance that has entered our lives with the

epidemic. Parallel to this situation, developments were observed throughout the country. As of 01.01.2021, the "Safe Tourism Certificate" program has been put into service, which is mandatory for accommodation facilities with 30 rooms or more, and optional for accommodation facilities with less than 30 rooms and other areas (4). Thus, it is seen that the awareness of hygiene is not limited to the enterprises, but also started to take place in the country's policies.

Looking at the definition in the literature, it is defined as "the kitchen is one of the most important parts of food and beverage businesses, and it is the place where all kinds of food are prepared, cooked and sometimes consumed due to the structure of the business" (5). The unhygienic kitchen and production areas and the lack of attention to hygiene rules by the personnel working here can make the food produced poisonous and cause diseases and deaths in the people who consume them (6). While restaurants, motels and bars are among the places where food poisoning is most common in Europe, the World Health Organization (WHO) states that 13 of the foodborne infections are caused by the faulty hygiene behaviors of the personnel (7). In a study, when the causes of food poisoning were examined, the personnel working in the kitchen and production area were held responsible for 25-40% (8). The causes of food poisoning in the Center for Disease Control and Prevention are respectively not keeping the food at the appropriate temperature (32.0%), insufficient heating of the food (21.0%), contaminated equipment (13.0%), and insufficient personnel hygiene (14,0%) was shown. In addition, hand hygiene of kitchen staff was shown as the cause of 34 of 81 food poisoning cases in the United States between 1975-1998 (9,10).

It is stated that the main cause of food poisoning is the hands of the kitchen staff who do not know or apply the hygiene rules. Because there are various types of microorganisms that are transmitted from the outer surfaces of the clothes of the kitchen staff, from the items they come into contact with and from the hands, arms, nasal cavity and mouths. If the kitchen staff does not comply with the hygiene rules, it is possible for these microorganisms to be transmitted to the food (11). In a study conducted in Turkey, Staphylococcus aureus bacteria were found at high levels in 9.4% of foods that require more manual labor (salad-meatballs) (12), while in another study, coliform bacteria were found in 20.0% of salads, Escherichia coli bacteria were detected in 11.4% and Staphylococcus aureus bacteria in 11.4% (13). In line with this information, the importance of prioritizing personnel, equipment hygiene and food hygiene in the kitchen emerges. Today's epidemics have created an obligatory awareness of both the operators and the kitchen staff on this issue and increased their

sensitivity to comply with the rules. From this point of view, in this study, it is aimed to present information on personnel hygiene and to evaluate the data on the hygiene status of kitchen staff in various businesses in Turkey.

1.1. Staff Hygiene in the Kitchen

When the origin of the concept of hygiene is examined, it is found in mythological sources and Ancient Greece. Hygiea, the son of the god of medicine Apollon and the daughter of Asclepios, is known as the goddess of hygiene and cleanliness. In history, the Romans played an important role in the development of the concept of hygiene with their water connections, baths and spas, and sewer lines. (14). The word hygiene (hygiene) comes from the Greek "Hygies" and gives the meaning of health, healthy life. Hygiene, known as "Hıfzıssıhha" in Turkish (15), means health information in the medical language. In other words, it is described as protecting and supporting health (11). "Personal hygiene, which is one of the cleaning and hygiene practices, is expressed as the self-care practices that an individual should do in a day to protect and maintain their health". It is important for kitchen workers to pay attention to hygiene rules in terms of preventing food poisoning and ensuring customer satisfaction. The kitchen staff should keep both the health conditions and the reliability of the business by complying with the recommended hygiene behaviors (16).

Hand hygiene takes the first place in personal hygiene applications. It is stated in the researches that hand washing with the right method prevents the catching and spreading of many diseases (17,18). Effective hand washing; It should be done by rubbing vigorously between the hands and fingers with clean water at a minimum of 38°C and a hand washing agent. Afterwards, the process should be completed with the appropriate drying method. The Food and Drug Administration (FDA) stated that the use of hand sanitizer alone does not provide hygiene. It has also been reported that hand antiseptics should be used after proper handwashing procedure (19). Following this, the golden method in the drying process is stated as disposable paper towels. Cloth towels can cause cross-contamination among employees and hygiene cannot be fully achieved due to hand dryers not drying the hands sufficiently (5). In addition, it is necessary to carry out controls related to hand hygiene of the personnel in the enterprises. After the analysis, it is expected that the total number of bacteria will be less than 3000, the number of microorganisms belonging to the Enterobacteriaceae family will be less than 1000, the number of Escherichia coli will be less than 100, the number of fecal Streptococcus

will be less than 1000 and the number of pathogenic S. aureus will be less than 100 (20).

Another hygiene practice is washing the face and feet, where the bacterial load and the risk of food contamination are high. The materials used in washing the face and feet play an important role in ensuring hygiene (16). In one study, it was determined that washing hands with soap and water reduced bacterial contamination thousand times (21), while another study showed that solid soaps are more contaminated with bacteria than liquid soaps (22). In this case, it has been understood that the use of liquid soaps should be preferred especially for hand washing by kitchen staff. In addition, brushing teeth twice a day, changing underwear and socks frequently, cleaning and changing work uniforms regularly, cutting nails regularly, removing nail polish on the fingers and taking a daily shower are also important practices (23).

In addition to personal hygiene practices, it is also necessary to clean the materials such as uniforms, aprons, masks, gloves, caps that the personnel wear while working. While it is recommended to prefer light colored aprons or uniforms as work clothes, changing the clothes when moving from the raw food processing section to the parts with processed/cooked products is a precaution that can prevent cross-contamination. In addition, these clothes should not have pockets, and if there is, the materials put in the pocket should be covered in order to reduce the risk of contact with food. Jewelry such as bracelets, rings and watches that can be seen in the kitchen staff unfortunately cause cross contamination. For this reason, it is necessary to not have this jewelry. Finally, it is recommended that the kitchen staff wear non-slip, waterproof and closed, comfortable shoes, that they are only used in the kitchen area, and that the staff have regular general health and carrier checks (5,11).

2. Method

The study which was planned as a review, was created by examining the studies on hygiene knowledge, hygiene status and hygiene behaviors of kitchen staff in Turkey between the years 2016-2022. In this context 1 national thesis and 11 research articles suitable for the purpose of the study were included in the scope of the study by scanning the Higher Education Council Thesis Center and Google Scholar databases.

3. Results

When the studies on the hygiene status of the kitchen staff in Turkey are examined, it is seen that negative results are obtained in the hygiene knowledge

and hygiene behaviors of the kitchen staff in most of the studies. In only one study, the hygiene knowledge of the kitchen staff was found to be high (24), while in one study, it was seen that the cooks had a high level of awareness about hygiene (25). In other studies, examined, it has been determined that the hygiene knowledge of the kitchen staff is low (19,26), they do not pay enough attention to hand washing, hair, beard, jewelry and clothing cleaning, and their awareness is not at a sufficient level. In studies that were added differently on these themes, Simsek and Sen (27) stated that as work motivation increases, hygiene behaviors are also positively affected; Gurpınar et al. (28) determined the positive effects of proper hand washing technique (Table 1).

Table 1. Analysis of the Hygiene Status of the Kitchen Staff

STUDY	**Sener and Cılgınoglu (26)**	**Gun and Kendirci (24)**	**Arısoy et al. (29)**
SAMPLE	6 chefs between the ages of 30-50 residing in Kastamonu	114 kitchen staff working in 3- and 4-star hotels in Tokat	207 kitchen staff in 17 accommodation establishments in Sanlıurfa
ANALYSIS	Food Safety and Hygiene Behaviors	Food Safety and Hygiene Knowledge Levels	Evaluation of Personal Hygiene and Knowledge
RESULTS	While only 33.4% of the chefs were found to have an idea about cross-contamination and HACCP principles, it was determined that they paid attention to the chopping boards, counters and cabinets in kitchen hygiene. 16.7% of the chefs emphasized the importance of personal hygiene.	When the results of the study were examined, it was determined that the personnel had a very good knowledge of food and equipment hygiene, and that they had a good level of knowledge about personal hygiene and food safety.	At the end of the study, it was determined that 28.5% of the personnel did not apply personal hygiene rules, and 65.2% brushed their teeth less than twice a day. In addition, it was observed that 61.4% shared a hand towel, 34.3% a face towel, 22.2% a foot towel, and generally did not know how to cut fingernails and toenails and how to clean their hair.

STUDY	Simsek and Sen (27)	Catar and Yıldırım (30)	Ozgel and Yıldız (31)	Kemer and Etyemez (25)
SAMPLE	466 kitchen workers from the Turkish Cooks and Chefs Federation	25 canteen employees working on Erciyes University Campus	20 Catering Companies	23 Chef Chefs Working in 4-5 Star Hotel in Nevsehir
ANALYSIS	Examining the Relationship Between Work Motivation and Hygiene Behavior	Hand Hygiene Conditions and Examining Hygiene Behaviors	Evaluation of Kitchen Hygiene Conditions	A Qualitative Study on Personal Hygiene Practices
RESULTS	While the average work motivation score of the kitchen staff was 3.61, the average of the hygiene behavior score was determined as 4.46. It has been determined that there is a positive relationship between both variables.	At the end of this study, the number of bacteria above the target value was detected on the hands of 40.47% of the personnel, and it was determined that the food offered for sale in the canteens, the personnel's hands and food contact surfaces were contaminated with indicator and some pathogenic bacteria. In addition, only 48% of the employees use bonnets, 44% use gloves, and 24% do not have hair and beard care.	When the results of the study are examined, the most common problem among the personnel is shaving the beard and mustache with a rate of 70.0%, followed by the use of gloves with a rate of 60.0% and the use of jewelry at a rate of 50.0%.	The themes created within the scope of the answers given by the kitchen chefs are as follows. Beard shaving, hair care, Daily showering Hand and nail hygiene (use of gloves) Cleaning of the used apron, Things to be applied regularly Cleaning of clothes Absence of jewelry such as necklaces, rings Shoes and slippers are appropriate.

STUDY	Gurpınar et al. (28)	Ertopcu et al. (32)	Dere (19)
SAMPLE	50 Gastronomy and Culinary Arts Department Students	260 students studying in Cookery Programs	147 staff working in cafeteria kitchens
ANALYSIS	Effect of proper hand washing technique on bacterial count	A Study on Hygiene Knowledge Levels	The Effect of Hygiene Education on Hygiene Behaviors
RESULTS	According to the results of the study, almost all of the bacterial load (99%) was destroyed in the majority of the students (41 students). A decrease in bacterial load was detected in the range of 75-85% in only 4 students and 50% and below in 5 students.	Although the general kitchen hygiene knowledge level of the students is good, their knowledge level about the items "I will postpone washing my hands if there is busyness while working", "I taste the food with my hands to make sure it is cooked" and "the probability of transmission of food-borne diseases is low" was found to be low.	87.7% of the participants stated that they had received hygiene training before. However, while the median hygiene knowledge score of all participants was 58 before the hygiene training, it increased to 82 after the training. In addition, before the hygiene training, 75.5% of the participants were at an insufficient level of knowledge, while after the training, 32% of them were at an insufficient level of knowledge.

STUDY	Sormaz and Sanlıer (33)	Kızılcık and Giritoglu (34)
SAMPLE	1150 food and beverage services personnel in Istanbul	213 production personnel working in the units where Maraş ice cream is produced
ANALYSIS	The Effect of Hygiene Education on Hygiene Behaviors	Level of Knowledge and Practice on Hygiene and Food Safety
RESULTS	The results of the study are summarized as follows. • Washing the hands at the right time (34.5% before the training, 72.5% after the training) • While the handwashing place before the training was 50.5% kitchen sink, the handwashing place after the training was determined as the kitchen handwashing sink at a rate of 76.6%. • While 55.7% of them wash their hands with liquid soap before the training, 73.7% of them wash their hands with disinfectant liquid after the training. • While 33.9% could not dry their hands before the training, 79.9% of them dry their hands with disposable paper towels after the training. Workwear change was determined once a week in 41.0% of the pre-training, while it was determined as every day in 47.4% after the training.	Among the personal hygiene behaviors, the personnel were found to have a high level of awareness about “long nails are extremely effective in the transmission of bacteria” and “the most ideal method for drying hands is the use of paper towels”. However, “there are bacteria that can cause foodborne poisoning on human skin” and “there are various bacteria in the hair that can contaminate food” are the least known statements by the ice cream production staff.

4. Discussion and Recommendations

The tourism sector, which has been damaged in the face of epidemics, wars, global and economic problems, has some issues to be aware of in order to protect itself. In order to prevent possible loss of tourists, tourism enterprises should meet the expectations of their customers despite various negativities and provide quality service, as well as comply with health conditions and hygiene rules (29). In this context, personnel hygiene is at the forefront of the steps that nutritional service businesses should take. In order for the personnel to take part in kitchen production, they must not be sick, their uniforms must be clean, their hygiene knowledge, behaviors and habits must be appropriate. Because food-related disturbances during travel will negatively affect both the health of the visitors and the enjoyment of the service they receive, causing the travel program to deteriorate. In addition, it reduces customer satisfaction and will lead visitors to obtain a negative impression of the destination (35).

In addition to tourism, the public's access to healthy and clean food plays an important role in maintaining public health. In the studies conducted on kitchen staff in the last six years in Turkey, it has been found that the hygiene knowledge of the kitchen staff is low, they do not pay enough attention to hand washing, hair, beard, jewelry and clothing cleaning, and their awareness is not enough. However, in studies where hygiene training was given to kitchen personnel (19,33), it was determined that the hygiene knowledge level of the personnel increased after the training and their behavior on this subject improved. In addition, it has been determined that the kitchen staff working in 3-4 star and 4–5 star hotels have a good level of knowledge about hygiene and give importance to their behavior (24,25). It is thought that this situation shows that as the quality of the enterprise increases, the quality of the personnel also increases. On the other hand, Sonmez et al. (7), in the study they conducted in 3-4-5-star hotels in Bursa, supports this situation. When it is examined whether the employees pay attention to their personal care (hair, beard, nails), the attention rates are determined as 67.6% (3-star hotel), 82.1% (4-star hotel), 90.3% (5-star hotel), respectively. Effectively washing their hands after using the toilet was 43.2% (3-star hotel), 75.0% (4-star hotel), 72.6% (5-star hotel); the status of paying attention to the cleanliness of work clothes was found to be 45.9% (3-star hotel), 75.0% (4-star hotel), 77.4% (5-star hotel). However, there are studies with conflicting findings. For example, only 22.5% of the personnel working in the restaurants of 4–5-star hotels in Gaziantep take a bath every day; It is seen that the personnel use bar soap at a high rate (42%-71%) for hand-face

washing, and most of the personnel brush their teeth only in the morning and do not brush their teeth during the day and after the meal (16).

As a result, the following suggestions were presented within the scope of the study.

- The kitchen staff must have a good level of hygiene knowledge and this information must be up-to-date and checked at regular intervals.
- The kitchen staff should be given regular hygiene training, and their knowledge level should be checked after these trainings.
- The working ones of the kitchen staff should be inspected and cleaned daily.
- The kitchen staff should not have jewelry, rings, hands should be washed in accordance with the procedure when necessary, and should pay attention to the use of uniforms and aprons. Gurpınar et al. (28) found that in most of the participants, the appropriate hand washing technique reduces the bacterial load on the hand by 99.0%.
- Gloves should be changed and hands should be disinfected after every operation in the kitchen.
- In the recruitment of kitchen staff, personnel who receive hygiene training should be selected or it should be mandatory to provide hygiene training in recruitment.
- In a study, it was seen that as the work motivation of kitchen workers increases, their hygiene behaviors are also affected positively (27). For this reason, it is recommended that employers offer conditions that will increase the motivation of employees.

References

1. Guneri B, Yay O. Reflections of the COVID-19 outbreak on the hotel kitchen department: A qualitative research on five-star hotels in Antalya. Journal of Tourism and Gastronomy Studies. 2022;10(1):225-247.

2. World Tourism Organization (UNWTO) (2020). International Tourism Highlights. https://www.e-unwto.org/doi/book/10.18111/9789284422456 (access date: 10.11.2022)

3. Turkish Statistical Institute (TUIK) (2021). Tourism Statistics. https://data.tuik.gov.tr/Bulten/Index?p=Turizm-Istatistikleri-IV.Ceyrek:-Ekim-Aralik-ve-Yillik,-2021-45785 (access date: 13.10.2022).

4. Turker GO, Erturk N. Covid-19 effects on the hospitality industry: an assessment from the perspective of managers. Journal of Tourism Economics and Business Studies. 2020;2(2):89-101.

5. Değirmencioglu N, Cicek D. Personnel hygiene and HACCP practices in the kitchen of hotel businesses. Anatolia: A Journal of Tourism Research. 2004;15(1):21-35.

6. Bayram F, Ersoy Y. Knowledge level of kitchen staff in hotels about food safety (Turkey, Germany, Spain and Dubai Cases). Journal of Tourism and Gastronomy Studies. 2020;8(4):565-580.

7. Sonmez O, Ozdemir Y, Serengil M. A study on the consciousness of applying the hygiene rules of the staff working in the kitchens of 3, 4 and 5 star hotels operating in Bursa province. PARADOKS Economics, Sociology and Policy Journal. 2015;11:72-86.

8. Troller JA. Water activity measurements with a capacitance manometer. Journal of Food Science. 1983;48(3):739-741.

9. Ekanem EP, Mafuyai-Ekanem M, Tegegne F, Adamu U. Assessing food safety training needs: findings from TN focus groups. Journal of Food Distribution Research. 2012;43(856-2016-58046): 7-11.

10. Olsen SJ, MacKinon L, Goulding J, Bean NH, Slutsker L. Surveillance for Foodborne Disease Outbreak United States, 1993–1997. Morbidity And Mortality Weekly Report. 2000;49:1-72. http://www.cdc.gov/mmwr/PDF/ss/ss4901.pdf (access date: 09.11.2022).

11. Unluonen K, Comert M. Identifying Personnel Hygiene Knowledge Levels of Kitchen Staff in Hotel Establishments. Journal of Tourism and Gastronomy Studies. 2013;1(1):3-12.

12. Aycicegi H, Cakıroglu S, Stevenson HT. Incidence of staphylococcus aureus in ready-to-eat meals from military cafeterias in Ankara, Turkey. Food Control. 2005;16:531-534.

13. Aycicegi H, Sarımehmetoglu B, Cakıroglu. Assessment of the microbiological quality of meals sampled at the meal serving units of a military hospital in Ankara, Turkey. Food Control. 2004;15: 379-384.

14. Tozun M, Sozmen MK. The historical development and the fundamental concepts of public health. Smyrna Tıp Dergisi. 2014;(3):58-62.

15. Parlak T. A Different view on the concept of hygiene in the production of food product. OHS Academy. 2020;3(2):73-101.

16. Kınacı B, Cetiner H. A Field research on personal hygiene practices of personnels working in restaurants of four and five star hotels in Gaziantep. Journal of Tourism and Gastronomy Studies. 2013;1(1):21-28.

17. Kendall A, Landers T, Kirk J, Young E. Point-of-care hand hygiene: preventing infection behind the curtain. American Journal of Infection Control. 2012;40(2012):3-10.

18. Sarter G, Sarter S. Promoting a culture of food safety to improve hygiene in small restaurants in madagascar. Food Control. 2012;25(2012):165-171.

19. Dere H. The Influence of Hygiene Education on the Hygiene Status of Foodhandlers in Catering Systems. Post Graduate Thesis Ankara: Baskent University, Institute of Health Sciences, Nutrition and Dietetics. 2018.

20. Persil Ozkan O, Sevim Y. Importance of hand hygiene in food service systems. The Journal of Health Sciences and Life. 2016;1(2):18-23.

21. Mackintosh CA, Hoffman PN. An extended model for transfer of micro-organisms via the hands: differences between organisms and the effect of alcohol disinfection. Epidemiology & Infection. 1984;92(3):345-355.

22. Sutton S. Hand washing-a critical aspect of personal hygiene in pharma. Journal of Validation Technology. 2009;15(4):50-55.

23. Cetiner H. Hygiene, Sanitation in Food and Beverage Business and Training Factor in Personal Behaviour Related to Hygiene Rules. Master Thesis. Ankara: Gazi University Institute of Educational Sciences, Department of Tourism Management Education. 2010

24. Gun S, Kendirci P. Investigation of the level of food safety and hygiene knowledge of kitchen staff working in 3 and 4 star hotels located in Tokat. Journal of Agriculture Faculty of Ege University. 2021;58(3):335-346.

25. Kemer E, Etyemez S. A Qualitative study on personal hygiene practices in hotel kitchens (example of Nevsehir province). Journal of Tourism and Gastronomy Studies. 2020;8(3):1787-1801.

26. Sener Y, Cılgınoglu H. Safe food and good hygiene practices of employees in convectional kitchens of food and beverage businesses: the case of Kastamonu. New Era International Journal of Interdisciplinary Social Researches. 2022;7(13):1-10.

27. Simsek M, Sen MA. A study on work motivation and hygiene behaviour of kitchen workers. Journal of Tourism and Gastronomy Studies. 2020;8(4):3039-3051.

28. Gurpınar S, Agan C, Ozer C. Hand hygiene applications of gastronomy and culinary arts students: the case of Istinye University. Journal of Tourism Economics and Business Studies. 2020;2(1):5-17.

29. Arısoy NÖ, Ince E, Olcay A. personal hygiene information and practices of kitchen department staff: an application in Sanliurfa province accommodation businesses. Journal of Turkish Tourism Research. 2021;5(3):2086-2106.

30. Catar O, Yıldırım Y. Hand hygiene profile of food handlers in canteens of Erciyes University campus. Kocatepe Veterinary Journal. 2020;13(1):52-59.

31. Ozgel O, Yıldız Z. Evaluation of kitchen hygiene conditions of ready-made food companies in Mersin. European Journal of Science and Technology. 2020;19:778-785.

32. Ertopcu İ, Avcıkurt A, Çetinkaya T. A Study on hygiene knowledge levels of cooking program students. Journal of Tourism and Gastronomy Studies. 2019;7(3):2187-2203.

33. Sormaz U, Sanlıer N. The impact of compulsory hygiene training given to employees working in the sector of food and beverages services on their hygiene-related habits, behaviors and knowledge. Journal of Human Sciences. 2017; 14(2):1356-1369.

34. Kızılcık O, Giritlioglu İ. A research on knowledge and practice levels about food safety and hygiene of employees in the production of ice-cream in the confectionery businesses of tourism services. Mehmet Akif Ersoy University Journal of Social Sciences Institute. 2016;8(15):301-319.

35. Eren R, Nebioglu O, Şık A. Knowledge levels on food safety of employees working in hotel enterprises' kitchen: the example of Alanya. Journal of Multidisciplinary Academic Tourism. 2017;1(2): 47-64.

CHAPTER II

MATERNAL HEALTH CARE EXPERIENCES AND OUTCOMES OF WOMEN WITH DISABILITIES: A SYSTEMATIC REVIEW

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1. Introduction

More than one billion people worldwide are recognized to have some form of disability, ten percent of whom are women of childbearing age [1,2]. Although the prevalence of pregnancy among women with disabilities is unknown [3], an increasing number of women with disabilities are having children [4,5]. Although all women have the right to receive high-quality maternity care, it has been reported that on average half of women with disabilities in the world do not have access to health care [5].

Women with disabilities are often assumed to be sexually inactive and less likely to have children [6]. This inaccurate assumption in societies has been demonstrated in qualitative studies on reproductive health and maternity care with women with physical disabilities [7–9]. Such attitudes, which reflect social stigmatization of women with disabilities as asexual and negative perceptions of their desire to become mothers, lead to limited access to reproductive health and maternal health services for women with disabilities [6,10–12]. In this direction, it has been stated that improved education and training can reduce inequalities in

access and utilization of health services for women with disabilities and improve the quality of care [13,14].

Some evidence in the literature suggests that although most women with disabilities have a pleasant birth experience, they are at higher risk of adverse birth outcomes, including pregnancy complications and preterm birth [15–18]. A recent meta-analysis by Tarasoff et al (2020) reported that women with disabilities are at high risk for adverse perinatal outcomes [19]. Although little is acknowledged regarding adverse outcomes and risks among newborns, newborns of women with disabilities have also been reported to be at high risk for preterm birth and low birth weight [20,21]. In addition, women with disabilities have been reported to have higher rates of domestic violence [22], higher rates of smoking [23] and substance use [24,25].

Along with negative attitudes of maternal care providers and lack of knowledge about care [16], there are other obstacles for women with disabilities, such as physically inaccessible facilities and examination tables [18,26]. Findings from Breckenridge et al's (2014) study suggest that poor relationships with health professionals and environmental barriers can jeopardize the utilization of maternity services by women [22].

It is essential to comprehend the maternal health care experiences of women with disabilities in order to meet their needs and evaluate the outcomes. When the literature was examined, it was observed that the number of studies conducted especially with women with physical disabilities has increased in recent years and the systematic reviews intensively addressed women with physical disabilities. For this reason, it was considered that the reviews should be updated by including studies examining the maternal healthcare experiences of women with other types of disabilities. Accordingly, the aims of this systematic review study are as follows:

- To review the published literature on maternal health care experiences of women with disabilities
- To review the literature on barriers and facilitating factors for women with disabilities in accessing maternal health care
- To review the literature on the current interventions to improve maternal health care and quality of care for women with disabilities

The findings from this study will provide researchers with the opportunity to examine the maternal health care experiences of women with disabilities and

how they evaluate these services. In addition, it is believed that it will guide the identification of interventions that improve access to maternal health services.

2. Method

This systematic review study was designed to examine the maternal health care experiences of women with disabilities, the obstacles they encounter in receiving services, and the results of maternal health care services available. The Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) protocol served as a guide for conducting the study and preparing the report [27].

PRISMA 2020 is designed for systematic reviews with or without synthesis. The PRISMA 2020 statement consists of a 27-item checklist addressing the introduction, methods, results and discussion sections of a systematic review report and a 12-item checklist for the abstract.

2.1. Research Question

What is already recognized in the existing literature about the maternal health care experiences of women with disabilities and how they evaluate the services provided?

The sub research questions are as follows:

1. What are the factors that hinder or facilitate access to maternal health care for women with disabilities and what are the existing interventions to address them?
2. What are the outcomes of maternal health services provided to women with disabilities?

This systematic review employed the PICOS framework to determine the relevance of the research question (Table 1).

Table 1. PICOS Framework

Criteria	**Determinants**
P (Population):	Women with disabilities (visual, hearing, mental, physical and walking disabilities) experienced to maternal health services (antenatal, delivery and postnatal)
I (Interventions):	Maternal health care experience (antenatal, delivery and postpartum)
C (Comparators):	In this study, the experiences of women with disabilities were examined and no comparison was made
O (Outcome):	Maternal health care experiences and evaluations of women with disabilities
S (Study desings):	Qualitative study design

2.2. Identification of Relevant Studies

In this systematic review, qualitative research studies published in peer-reviewed journals that analyze the research questions that we addressed were examined. The screening process was carried out by searching electronic databases. National level publications were scanned through Ulakbim, Türk Medline, Google Scholar search engines, and international publications were scanned through Web of Science, PubMed, Google Schooler databases.

Studies published in Turkish and English between January 2000 and December 2022 were screened to determine the studies to be included. Articles were also screened through citations in the references list of the studies included in this study. Studies published in Turkish were reviewed with the search terms ““maternal sağlık hizmeti, engelli kadınlar, doğum öncesi bakım, doğum, doğum sonrası bakım, deneyim”” and studies published in English were reviewed with the search terms “maternal health services, disabled women, antenatal care, postpartum care, birth/childbirth, experience”. Database-specific Medical Subject Headings (MeSH terms) were also utilized to identify articles. After the search, the retrieved studies were re-examined according to the inclusion and exclusion criteria.

2.3. Eligibility Criteria

Eligibility criteria were developed to obtain information within the studies identified as a result of the screening regarding the maternal health care experiences of women with disabilities, the quality of the service, the obstacles to access to the service and the answers to the research question.

2.3.1. Inclusion Criteria:

Studies were included if they fulfilled the following characteristics:

- Published in Turkish and English
- Focusing on maternal health care and women with disabilities
- Reporting the experiences, assessments and access of women with disabilities to maternal health services
- Published from 2000 to December 2022, to emphasize the reduction of maternal mortality in relation to the Millennium Development Goals and to be relevant to contemporary health care experiences
- Studies with a qualitative study design that analyzed oral testimony were included since the unique needs and experiences of people with disabilities are most authentically identified by people living with disability.

2.3.2. Exclusion Criteria:

Studies were excluded if they fulfilled the following characteristics:

- Studies that do not include women with disabilities in the population of the study,
- Studies focusing on health services outside maternal health care for women with disabilities,
- Research conducted with different study designs than qualitative study design,
- Studies that could not be accessed in full text.

To ensure the compatibility of the selected databases and keywords, a pilot study was conducted with the identified search strategies. All eligible studies were transferred to EndNote 20 reference management software to check for duplication. After eliminating duplicate studies, selection was based on title, abstract and full text, respectively.

2.4. Assessment of Methodological Quality of Studies and Analysis of Data

The methodological quality of the studies included in this systematic review was assessed with checklists published by the Joanna Briggs Institute [29]. Accordingly, a 10-item checklist used in the quality assessment of qualitative research was employed. Each item in these lists is evaluated as "yes, no, uncertain and not applicable". The checklist was utilized to examine the relevance, adequacy and methodology of the study, study design, participant recruitment,

data collection, data analysis, presentation of findings, authors' discussion and conclusions. The quality of the article was determined by examining specified aspects. The qualifications for each study are presented in Table 2.

The narrative synthesis method, which is an instrument used to synthesize qualitative studies, was employed to analyze the data. The study selection procedure is summarized through PRISMA table as presented in Figure 1.

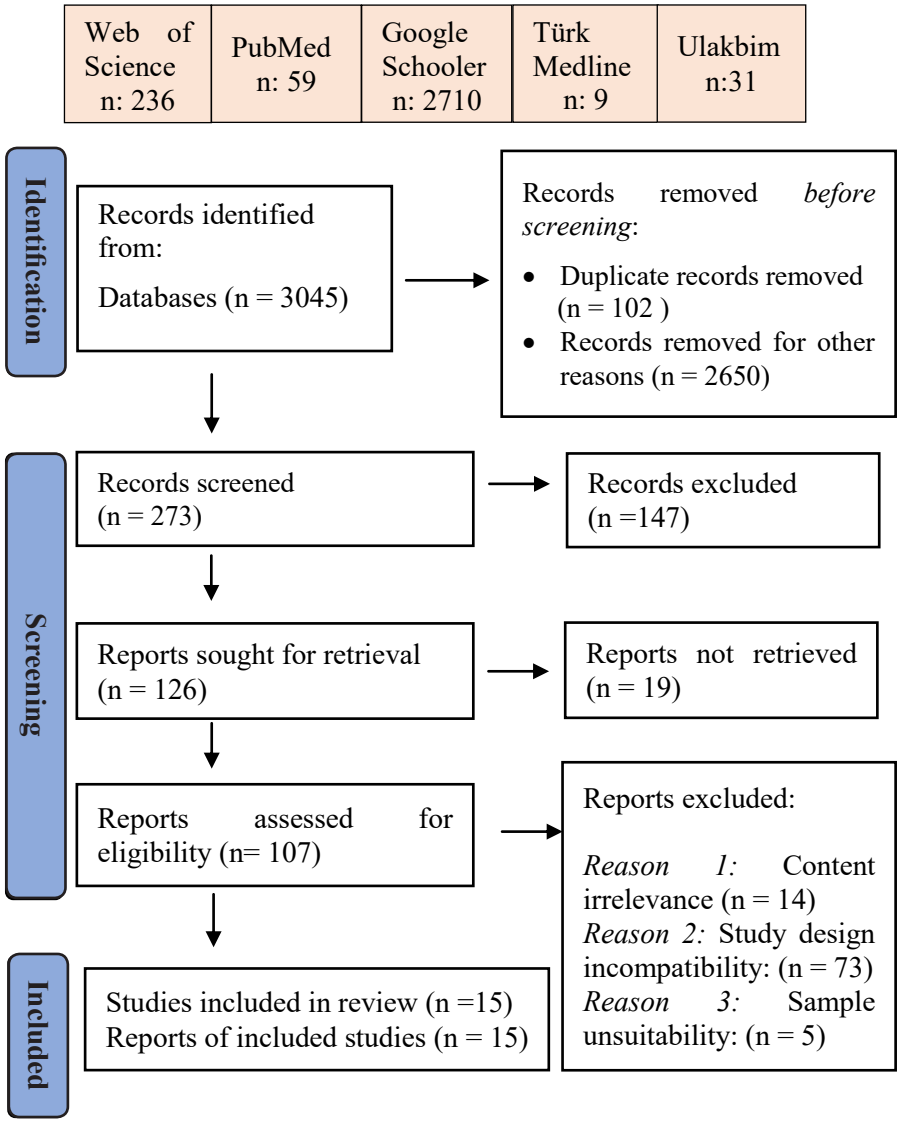

Figure 1. PRISMA Flowchart [28]

3. Findings

The systematic review included articles published in national and international journals since 2000. The author name, year and country, purpose of the study, study design, participant characteristics and sample size, important findings/major themes, conclusions/recommendations and quality score of all qualitative studies included in the systematic review are presented in Table 2.

Table 2. Characteristics and Quality Assessment Scores of Included Studies

Author, Year, Country	Aims of the study and Methodology	Participants and Sample Size	Key Findings / Main Themes	Conclusion / Recommendations	Qualitiy appraisal score (percentage)
Iezzoni et al, 2015 [30] **United States of America**	To examine the physical accessibility of antenatal care services for women with mobility disabilities and their experiences with the equipment utilized in the service *Methodology:* A qualitative study with in-depth, semi-structured, open-ended interview protocol and 2-hour telephone interviews	22 women with physical disabilities who benefited from mobility assistance devices or experienced limitation in arm/ hand movements during pregnancy	Maternal health care providers often do not possess height-adjustable examination tables. Some women were therefore physically unable to be examined or were examined in their wheelchairs. They also described being lifted onto non-adjustable tables for examination as intimidating and humiliating. Due to a lack of accessible scales, none of the participants were routinely weighed throughout their pregnancies. This caused concern for many women with disabilities as it could lead to inaccuracies in drug dose calculations during delivery.	Inadequate and inappropriate equipment led to non satisfactory care for pregnant women with disabilities. Height adjustable examination tables and weight scales will significantly improve care and comfort for pregnant women with physical disabilities.	**7/10 (70%)**
Smeltzer et al, 2017 [31] **United States of America**	To explore the birth and anesthesia experiences of women with physical disabilities to guide improvements in obstetric care	See Ieozzoni et al, 2015 [30]	Four main themes describing how they experienced labor and birth: - *Preferences of women for mode of delivery,* Birth arrangements were usually negotiated between women and their caregivers. - *Clinicians not expecting labor pain from women with disabilities,*	The study revealed that clinicians are in need of training on this issue in order for women with physical disabilities to receive more effective care during pregnancy.	**7/10 (70%)**

			Clinicians have the inaccurate assumption that women should not experience pain during labor. - *Fears that trigger active defensibility* Women experience fear related to caregivers' lack of knowledge about disability. - *Positive experiences* Women who answered the questions and provided reliable information reported positive experiences. Four themes related to anesthesia experiences: - *The importance of consultation with the anesthesia team*, The significance of discussing options, fears and preferences is highlighted. *-Decisions between epidural/spinal and general anesthesia* It is not always determined in consultation with the woman. - *Unsuccessful epidural with repeated attempts* It has been described as frightening and painful. - *Fear of anesthesia-related injury* Participants reported that they were concerned about injury during anesthesia.	It is recommended that clinicians should devote more attention to informing and consulting pregnant women with physical disabilities about treatment decisions.	

Lipson, 2000 [9] **United States of America**	To examine the pregnancy, birth and postpartum experiences of women with mobility limitations and to investigate their satisfaction with health services during these periods *Methodology:* A qualitative study with semi-structured interviews Most of the in-depth interviews lasting 1-2 hours were conducted in the houses of the participants.	12 women with mobility disabilities who gave birth in the last two years	Personal and health system factors influencing women's birth and postpartum experiences: - *Specific disability* Women with nonvisible disabilities reported that they felt less supported. - *Resource availability* They reported that access to pregnancy and infant care related information was very challenging. - *Individual approach to pregnancy and childbirth* Women who advocated for care preferences had more satisfying care experiences. - *Structural and political factors* They experienced the negative effects of lack of communication between caregivers and referrals to relevant health services. - *Attitudes of health care workers* Lack of knowledge of health workers negatively affected attitudes towards women with disabilities.	The positive attitudes and support of the staff caring for women with disabilities increase their self-confidence. This situation affects the whole process of birth experiences of women with disabilities.	**8/10 (80%)**
Long-Bellil et al, 2017 [32] **United States of America**	To explore the pain relief practices of women with physical disabilities during labor to inform future maternal care	25 physically disabled women younger than 55 years of age who gave birth within the last 10 years	Women's experiences of pain relief practices during childbirth varied tremendously. While some women were able to select methods of relief, others faced systemic barriers to access pain relief options.	While some women had positive birth experiences, systemic barriers and lack of clinician knowledge prevented some women from receiving quality care.	**8/10 (80%)**

	Methodology: It was designed as part of a larger mixed methods study (**Mitra, 2016** [18] **and Smeltzer, 2016**) [26] A qualitative study with in-depth, semi-structured and telephone interviews lasting a maximum of 2 hours		Many anesthesiologists lack experience and knowledge in the care of women with disabilities. Women who were able to plan their pain relief options in advance with their health care providers reported more positive birth experiences.	Women were most satisfied with birth experiences that provided opportunities for them to make an informed choice and included a comprehensive and individualized assessment of their options as part of an advanced planning process.	
Mitra et al, 2016 [18] **United States of America**	To examine the unmet health needs of women with physical disabilities during pregnancy and to examine their recommendations for similar women who are also considering pregnancy *Methodology:* A qualitative study with in-depth, semi-structured and telephone interviews lasting a maximum of 2 hours	Interviews were conducted with 25 physically disabled mothers younger than 55 years of age who gave birth in the last 10 years.	Three main themes identified to address unmet needs: -*Knowledge and attitudes of clinicians* Clinicians are not aware of the impact of disability on pregnancy. -*Physical accessibility of health facilities and equipment* Women reported lack of access to facilities and equipment to provide medical care. - *Necessity of information on support during pregnancy and postpartum* Women reported receiving insufficient support and information about the interaction between their disability and pregnancy and the postpartum period. Four main recommendations for other women with physical disabilities:	Participants have a wide range of negative experiences related to caregiver knowledge and attitudes, physical accessibility to health facilities and equipment, and pregnancy and postpartum support.	**7/10 (70%)**

			- Choosing a favorable clinician - Requesting peer support from other mothers with disabilities - Being assertive and self-defensive - Preparing for pregnancy, childbirth and postpartum		
Smeltzer et al, 2016 [26] **United States of America**	Investigating the perinatal experiences of women with physical disabilities with maternity care providers *Methodology:* A qualitative study with in-depth, semi-structured and telephone interviews lasting a maximum of 2 hours	Interviews with 25 physically disabled mothers younger than 55 years of age who gave birth in the last 10 years.	Three main themes including experiences with birth caregivers: *- Lack of knowledge of clinicians about the pregnancy-related needs of women with physical disabilities* Participants stated that knowledge deficiencies should be improved through training. *- Clinicians do not take into account the knowledge, experience and expertise of women about their disabilities* They stated that unless there is a medical emergency, they know their bodies best, but clinicians often take this information for granted. *- Lack of awareness among clinicians about reproductive concerns of women with physical disabilities* Women reported that clinicians often treated them in a stigmatizing way, such as labeling them as asexual or being judgmental by suggesting that they should not have children.	Training of health professionals on disability is crucial to eliminate stigma and prejudice against women with disabilities. It is emphasized that training of clinicians is essential for women with physical disabilities to receive the high quality health care that both themselves and their babies deserve during pregnancy.	**7/10 (70%)**

McKay-Moffat and Cunningham 2006 [33] **United Kingdom**	To explore the views and experiences of mothers with disabilities about childbirth, as well as the experiences of the midwives who care for them. *Methodology:* A qualitative study with semi-structured interviews, which constitutes the first stage of a two-stage mixed method study The themes generated from the data obtained from the interviews with women with disabilities were compared with the themes obtained from the interviews with midwives.	In the first phase of the study, five mothers and eight midwives from three different maternity units with mobility limitations were included. Only results from women with disabilities were evaluated in this review.	Five themes developed from the statements of mothers with disabilities about their birth experiences: *-The search for normality and independence* Participants do not want disability to be the primary focus of maternity care. *-Disability is the most important thing* Some women reported feelings of embarrassment because their disability attracted the attention of other people and this affected their self-confidence. *-Lack of knowledge about disability among midwives* Lack of information has led to inadequate care and support. *-The necessity for midwives to have disability awareness and positive attitudes* Interactions with midwives range from insensitive to respectful attitudes. *-The importance of effective communication* Open communication between midwives and women reduces anxiety and increases satisfaction.	Although services for mothers with disabilities appear to have improved, they are not yet at a sufficient level. In order to improve the quality of services, the information and demands of women with disabilities should be utilized.	**9/10 (90%)**

Prilleltensky, 2003 [34] **Canada**	To explore the pregnancy and early parenting experiences of mothers with disabilities and the availability of both formal and informal supports *Methodology:* A qualitative study including both focus groups and individual interviews Four focus groups and twenty-six individual interviews were conducted in the study. The focus groups were conducted at a symposium on women with disabilities and motherhood about 18 months before the in-depth interviews. During the symposium, women who were willing to participate in the in-depth interviews were identified and then individual interviews were conducted in the houses of the participants.	13 women with physical disabilities	-Many women felt under pressure by health care providers to terminate their pregnancies, even if they were planned. -Some women reported being excessively monitored and questioned in postnatal wards, as they had to prove their competence as a parent. -Women appreciated health professionals who included them in care decisions and provided additional support when needed. - The main author has lived experience of physical disability and conducted the interviews with the participants.	Mothers are diverse in terms of the level of support they receive and the stressors they face. The lack of funded resources to meet their needs is a major obstacle for mothers with physical disabilities. Women who need help to fulfill their caregiving duties emphasize the importance of retaining control in their parental role.	**7/10 (70%)**

Schildberger et al, 2017 [35] **Austria**	To explore the experiences and personal understandings of women with physical and sensory disabilities about pregnancy, childbirth and puerperium *Methodology:* Qualitative design involving in-depth semi-structured interviews The interviews lasted between 50 and 90 minutes, some of which were conducted in participants' homes. Interviews with hearing-impaired women were assisted by a sign language interpreter or a close relative.	10 women residing in Austria between September 2013 and March 2015, 4 with physical disabilities and 6 with sensory disabilities (4 visually impaired, 2 hearing impaired)	Three themes identified for the experiences of women with disabilities related to pregnancy, childbirth and puerperium: -*Social network* Women desire to be accepted and normalized by society as a woman and mother. - *Self-efficacy and self-awareness* Women are generally confident in their own abilities. However, lack of support and negative attitudes of health care providers undermine their confidence. -*Communication, transparency and information* There are uncertainties in communication between women and caregivers. Information on pregnancy, childbirth and puerperium is insufficient for women with disabilities.	Women experience a lack of trust and support in their parenting abilities. They also feel a violation of their personal boundaries and a sense of being monitored and controlled. This negatively affects their self-efficacy and self-awareness.	**8/10 (80%)**

Tarasoff, 2017 [8] **Canada**	To examine the perinatal care experiences and outcomes of women with physical disabilities in Ontario, Canada to identify obstacles in care *Methodology:* A qualitative study with in-depth interviews conducted partly by telephone and partly face-to-face	13 physically disabled women who gave birth in the last five years	Five interrelated themes related to obstacles to perinatal care: *-Inaccessible care settings* Women reported encountering inaccessible perinatal care settings, including inaccessible examination tables, toilets and other needed equipment. *-Negative attitudes* Maternal caregivers exhibit negative attitudes such as questioning women's parenting ability and reluctance to help when necessary. *-Lack of knowledge and experience* Lack of knowledge among maternal caregivers leads to feelings of frustration and anxiety in women. *-Lack of communication and cooperation between caregivers* There is a lack of communication and cooperation, especially among care providers, regarding childbirth and disability *-Misconception of disability-related needs* The problems faced by participants often stem from a lack of understanding of disability and disability-related needs among perinatal caregivers.	The findings from the study suggest that there is a need for a broader comprehension of disability-related needs and the issue of disability. Statements from participants suggest that disability-related mobility limitations are not always a primary concern.	**9/10 (90%)**

Tebbet and Kennedy, 2012 [36] **United Kingdom**	To examine the pregnancy and birth experiences of women with spinal cord injury *Methodology:* A study in qualitative design utilizing an interpretive phenomenological approach In-depth, semi-structured interviews with participants who gave birth in a hospital where the maternity ward and the specialist spine center were present together were conducted in the houses of the participants.	8 women who gave birth in the last eight years and had a spinal cord injury before giving birth to a child	Five main themes for women with spinal cord injury's accounts of their experiences of pregnancy and childbirth: *-Preparing for childbirth* Women seek pregnancy and childbirth information about themselves before their birth. *-Birth as a joyful event* Most women reported positive birth experiences. *-Birth as a unique experience for women with spinal cord injury* They reported unique care plans, including routine transfers to the spinal cord service before and after birth. *-The importance of support* The supportive attitudes of caregivers improved women's experiences. *-Birth as a team effort with varying degrees of controllability* Women reported that they valued being listened to and involved in care decisions by caregivers.	The narratives provided by women in this study provide information that helps normalize birth for women with spinal cord injury. The results of the study also clearly indicate the need for a more integrated service delivery model for women with spinal cord injury.	**7/10 (70%)**

Walsh-Gallagher et al, 2012 [7] **Ireland**	Describe the personal understandings that women with disabilities attach to their experiences of pregnancy, childbirth and motherhood, as perceived by them, with a particular focus on feelings of control, making choices, access to care and sustainability of care *Methodology:* A qualitatively designed study involving in-depth, semi-structured interviews using a descriptive, phenomenological approach Each participant was interviewed twice: The first interview was conducted in the second trimester of pregnancy and the second interview was conducted up to three months after delivery.	17 pregnant women with physical, sensory and/or intellectual disabilities from the island of Ireland	Two main themes were identified from the statements on the experiences of women with disabilities: -*Affirmation of Motherhood* -Women express happiness at the validation of their motherhood and the regularity of their experiences despite their obstacles. -*Perceived reactions of others* Caregivers perceive women as high risk. Being labeled as high risk increases women's anxiety. Other findings: -Women do not feel in control of their care choices and birth experiences. -Caregivers have insufficient knowledge about disability.	Pregnant women with disabilities, especially those labeled as 'high risk', expect as easy access to appropriate maternity care as those without disabilities. Maternity services should support women with disabilities, taking into account their personal preferences, and protect their identity and value as mothers.	**8/10 (80%)**

Nguyen et al, 2020 [37] **North Vietnam**	To investigate how maternal health care is practiced and accessed by women with physical disabilities *Methodology:* A study in qualitative design employing an interpretive phenomenological approach The interviews were generally conducted in the homes of the participants with the questions included in the guide prepared by the authors.	27 women with physical disabilities who gave birth in the last three years	Six themes were identified regarding the experiences of women with maternal health care: *-Technology and the search for "normal"* Many women expressed a deep desire to check whether their babies were developing "normally". *-Caregivers' belief that "people with disabilities should not give birth"* Some women reported that their right to be a mother was denied by maternal health care providers and that they faced negative attitudes. *-Information only for "normal" women* There is limited specific information for women with disabilities. *- Increased direct and indirect costs* Women reported that although they had health insurance, additional costs for travel and "thank you money" to the delivery team were very common. *- Confusing referral and waiting procedures* While there are policies prioritizing persons with disabilities in service delivery, this is not always adhered to and confusing referral systems are encountered.	Decisions by women to seek maternal health care are influenced by staff attitudes and lack of recognition of their right to quality maternal health care. Many Vietnamese health services are ill-equipped to provide disability-inclusive and responsive maternal health care due to poor structural design, limited appropriate knowledge and financial burden.	**8/10 (80%)**

			-Poor accessibility to transportation, facilities and equipment Many women spoke about the challenges of independently accessing health services during their pregnancy.		
Magagula et al, 2022 [38] **Eswatini**	To explore and describe the care experiences of women with mobility disabilities during pregnancy, childbirth and puerperium in Eswatini *Methodology:* A study in qualitative design using a descriptive and phenomenological approach Participants were purposively sampled from the entire population of women with mobility impairments residing in Hhohho and Manzini districts in Eswatini.	12 mobility impaired women over 18 years of age with childbirth experience	Five main themes related to statements by women about their experiences of receiving maternal health care: *-To report the experience with midwives as unprofessionals* Midwives were reported to behave in an unsympathetic and unprofessional manner. *- Experience of being victimized by midwives* The negative attitudes of midwives make women with disabilities feel emotionally abused. *-Lack of support during pregnancy, childbirth and puerperium* Participants feel desperate in the absence of support and assistance *- Experiencing the challenges related to lack of protocols, equipment and infrastructure before and after birth* Women face challenges with inadequate and inappropriate protocols, equipment and infrastructure to meet their needs	Women with mobility impairments experience many challenges in the antenatal, labor and postpartum period. It is recommended that guidelines be formulated to provide the necessary support to women with mobility impairments and to provide holistic maternity care.	**8/10 (80%)**

			-Experiencing the need for support from midwives They stated that they need midwives equipped with the knowledge and skills to provide adequate maternal care.		
Apolot et al, 2019 [39] **Uganda**	To investigate the maternal and newborn health needs of women with mobility disabilities in Kibuku District, Uganda *Methodology:* A qualitative study with in-depth individual interviews Participants were selected from among women with mobility disabilities using purposive sampling method. Interviews were conducted in the homes of the participants to explore the needs of women with mobility disabilities.	4 women with mobility disabilities who gave birth between 2015-2017	Four main themes developed from statements on maternal and newborn care needs: *-Psychosocial maternal-newborn needs of women with mobility disabilities* Women have a range of psychosocial needs, including recognition by partners, family, community and health professionals. *- Mobility-related needs of women with mobility disabilities* They have mobility requirements for transportation to the health facility and childcare tasks. *- Maternal and newborn care needs of women with mobility disabilities at health facilities* Due to some infrastructural inadequacies in health facilities, meeting special service needs is inadequate. *- Personal maternal and neonatal needs of women with mobility disabilities* Women reported personal needs related to childbirth preparation, baby care, personal protective clothing and basic needs.	Women with disabilities have many unmet needs for maternal health care. Policy makers and maternal service providers need to take deliberate steps to ensure appropriate infrastructure for women with disabilities.	**7/10 (70%)**

3.1. Characteristics of Studies

The characteristics of the included studies are presented in Table 2. These studies, published between 2000 and 2022, were conducted in the United States [9,18,26,30–32], United Kingdom [33,36], Canada [8,34], Austria [35], Ireland [7], Vietnam [37], Eswatini [38] and Uganda [39]. Thirteen studies conducted individual interviews [7,8,38,39,9,18,30–33,35,36]. One study combined individual interviews with focus groups [34] and one study with physical access audits [37]. Five studies conducted face-to-face interviews [9,35–37,39] and nine conducted telephone interviews [7,18,26,30–33,38–40] and one study conducted both face-to-face and telephone interviews[8]. In this study, studies conducted with participants with any type of disability were included and no distinction was made based on the type of disability.

As a result of the analysis of the main themes in the findings of these studies, three main themes were identified.

- **Knowledge and Attitudes of Maternal Health Service Providers**

The main theme "Knowledge and attitude of maternal health care providers" includes the categories of lack of knowledge, disregard for the concerns of women, inadequate support and communication, and unwillingness to help and support.

In the included studies, clinicians do not have sufficient information about pregnancy, birth and postpartum processes of women with disabilities [7–9,18,31–33], access to information is challenging [9], there is not adequate information [37], and most of all, unprofessional negative judgments and attitudes are exhibited by caregivers, such as their rights and wishes to be a mother are not considered normal by caregivers [8,26,35,38] and that these attitudes lead to less support [18] and poor communication [8,9,33,35].

In the studies examined, it was observed that the experiences of women with disabilities regarding the knowledge and behaviors of maternal health care providers constituted obstacles in accessing services and receiving quality care. These obstacles have been reported to cause dissatisfaction with the maternal health care services received by women and to have a negative impact on their decisions to receive services.

- **Structural/Physical Conditions and Political Factors**

Under this main theme, the study findings were assessed in the sub-categories of medical treatment, access to and availability of care facilities, specialized supplies and equipment, and improved integration of care.

In the included studies, women with disabilities have been affected by inaccessible care settings [8,18,37], inadequate and inappropriate equipment in institutions providing medical care [30] perplexing referral systems and inadequate protocols [9,37,38] inadequate resources and infrastructure funded for needs [34,38,39] provide evidence of factors that negatively impact women's experiences.

The findings of the studies unearthed that inadequate transportation and equipment, complicated referral systems and long waiting times make it even more difficult for women with disabilities to access inadequate maternal health services and have a negative impact on the services they receive.

• **Emotions of Women**

Under this main theme, the emotions of women were addressed under the sub-categories of vulnerable, anxious, frightened, needing self-defense, perceived stigma, lack of validation and desire for normalization.

In the included studies, women with disabilities were not consulted for their opinions during pregnancy, birth and postpartum processes, were not included in decisions and experienced lack of information, frustration, fear and anxiety from caregivers [8,30,31], focusing on disability during the birth process and being considered in the high-risk group can lead to anxiety [7], stigmatization by society, family members and healthcare professionals [26], feelings of shame[33], helplessness and emotional abuse [38], violation of personal boundaries, feeling pressured and constantly monitored [34,35], and these feelings often trigger a wide range of negative emotions such as the desire for acceptance and normalization [7,35,39] the desire to have control over their decisions and births [7] and the need for self-defense [34]. Such feelings have been found to negatively affect the self-efficacy and self-awareness of women with disabilities, making it difficult for them to access maternal health services and influencing their decisions to receive services.

In addition to these findings, there is evidence that providing women with disabilities with reliable information [31], positive attitudes and support [9], comprehensive individualized care with the opportunity to make an informed choice [32] and a more integrated service [36] positively affect their experience of maternal health care.

4. Discussion

Quality maternal health care prevents maternal mortality when it is available and accessible to those who need it. This study therefore included research that

focused on experiences of access (availability, affordability, acceptability and quality of services) and utilization.

Different studies over the last 22 years have repeatedly highlighted that existing health services for women with disabilities present structural obstacles, low-quality birthing centers and access to appropriate services. However, when the experiences of women with disabilities are examined over the years, little has changed. Therefore, the maternal health care experiences of women with disabilities are consistent across studies conducted over 22 years. These findings are confirmed by recent research, including Tarasoff's (2017) report on obstacles to accessing facilities and equipment, and care that is not tailored to the needs of women [38,39].

Based on the experiences of women, some studies have demonstrated that maternal health care providers ignore women's disability-related concerns by focusing only on pregnancy and childbirth [32,38]. However, there is also strong evidence that the supportive attitude of health professionals has a positive impact on the experiences of women with disabilities [9,18,31]. In these studies, women described health care providers who provided responsive, respectful and supportive care. Women who were involved in their own care, participated in their birth choices, and had their disability and knowledge taken into account reported more positive maternal healthcare experiences. A mother with cerebral palsy reported that "*My obstetrician knew that I was not speaking out of fear, that I had some knowledge and education to support my decisions. She really believed that I knew my body best and was willing to help me*." [31].

It has been observed in the findings of studies that the lack of knowledge of maternal health service providers reflects negatively on the experiences of women. Lack of knowledge leads to inadequate care and support [33]. However, health care providers caring for women with disabilities also acknowledge the lack of accessible care facilities, equipment and disability-specific training. Lack of information leads maternal health care providers to feel unqualified to care for women with disabilities [7,18].

It is evident from the findings that there is a need to prioritize maternity care for women with disabilities. Guidelines for the implementation of holistic maternity care are essential to facilitate support and holistic maternity care for women with disabilities. These could include empowering midwives to practice professionalism, respectful communication and compassion in the care of women with disabilities. There is also a strong need to formulate specially adapted protocols and design equipment and infrastructure to promote optimal health for women with disabilities.

5. Conclusion

Maternal health services designed to meet the needs of healthy women are not providing the flexibility necessary to meet the specific maternity care needs of women with disabilities. Current health services are far from being tailored to meet the needs and expectations of these mothers and further improvements in both policy and practice are therefore required to provide better health care for these women. Policy should provide health care providers with a framework that enables them to meet the needs of women in the most effective way.

There is a requirement to ensure disability-friendly transportation, health facilities and services, as well as filling the disability-related training gap for health care providers. To ensure the development of disability-friendly maternal health services, there is also a need for research on maternal health needs, obstacles and factors that facilitate access to maternity services for women with disabilities.

References

1. WHO. *World Report on Disability*.; 2011.

2. United Nations. Maternal health. Published 2019. https://www.unfpa.org/maternal-health

3. Iezzoni LI, Yu J, Wint AJ, Smeltzer SC, Ecker JL. Prevalence of current pregnancy among us women with and without chronic physical disabilities. *Med Care*. 2013;51(6):555-562.

4. Iezzoni LI, Wint AJ, Smeltzer SC, Ecker JL. Recommendations about pregnancy from women with mobility disability to their peers. *Women's Heal Issues*. 2017;27(1):75-82.

5. Malouf R, Henderson J, Redshaw M. Access and quality of maternity care for disabled women during pregnancy, birth and the postnatal period in England: data from a national survey. *BMJ Open*. 2017;7(7):e016757.

6. Ahumuza SE, Matovu JKB, Ddamulira JB, Muhanguzi FK. Challenges in accessing sexual and reproductive health services by people with physical disabilities in Kampala, Uganda. *Reprod Health*. 2014;11(1):59.

7. Walsh-Gallagher D, Sinclair M, Mc Conkey R. The ambiguity of disabled women's experiences of pregnancy, childbirth and motherhood: A phenomenological understanding. *Midwifery*. 2012;28(2):156-162.

8. Tarasoff LA. "We don't know. We've never had anybody like you before": Barriers to perinatal care for women with physical disabilities. *Disabil Health J*. 2017;10(3):426-433.

9. Lipson G. J. Pregnancy, bırth, and dısabılıty: women's health care experıences. *Health Care Women Int*. 2000;21(1):11-26.

10. Milligan MS, Neufeldt AH. The myth of asexuality: A survey of social and empirical evidence. *Sex Disabil*. 2001;19(2):91-109.

11. Morrison J, Basnet M, Budhathoki B, et al. Disabled women's maternal and newborn health care in rural Nepal: A qualitative study. *Midwifery*. 2014;30(11):1132-1139.

12. Eisenberg NW, Andreski SR, Mona LR. Sexuality and physical disability: a disability-affirmative approach to assessment and ıntervention within health care. *Curr Sex Heal Reports*. 2015;7(1):19-29.

13. Amir N, Smith LD, Valentine AM, Mitra M, Parish SL, Moore Simas TA. Clinician perspectives on the need for training on caring for pregnant women with intellectual and developmental disabilities. *Disabil Health J*. 2022;15(2):101262.

14. Smeltzer SC, Mitra M, Long-Bellil L, Iezzoni LI, Smith LD. Obstetric clinicians' experiences and educational preparation for caring for pregnant women with physical disabilities: A qualitative study. *Disabil Health J*. 2018;11(1):8-13.

15. Signore CC. Pregnancy in women with physical disabilities. In: John T. Queenan MD, Catherine Y. Spong MD CJLM, ed. *Queenan's Management of High-Risk Pregnancy*. Wiley; 2012:253-259.

16. Mitra M, Long-Bellil LM, Smeltzer SC, Iezzoni LI. A perinatal health framework for women with physical disabilities. *Disabil Health J*. 2015;8(4):499-506.

17. Clements KM, Mitra M, Zhang J, Iezzoni LI. Pregnancy characteristics and outcomes among women at risk for disability from health conditions ıdentified in medical claims. *Women's Heal Issues*. 2016;26(5):504-510.

18. Mitra M, Long-Bellil LM, Iezzoni LI, Smeltzer SC, Smith LD. Pregnancy among women with physical disabilities: Unmet needs and recommendations on navigating pregnancy. *Disabil Health J*. 2016;9(3):457-463.

19. Tarasoff LA, Ravindran S, Malik H, Salaeva D, Brown HK. Maternal disability and risk for pregnancy, delivery, and postpartum complications: a systematic review and meta-analysis. *Am J Obstet Gynecol*. 2020;222(1):27.e1-27.e32.

20. Gleason JL, Grewal J, Chen Z, Cernich AN, Grantz KL. Risk of adverse neonatal outcomes among pregnant women with disabilities. *Int J Epidemiol*. 2022;52(1):203-213.

21. Tarasoff LA, Ravindran S, Malik H, Salaeva D, Brown HK. Maternal disability and risk for pregnancy, delivery, and postpartum complications: a systematic review and meta-analysis. *Am J Obstet Gynecol*. 2020;222(1):27.e1-27.e32.

22. Breckenridge JP, Devaney J, Kroll T, Lazenbatt A, Taylor J, Bradbury-Jones C. Access and utilisation of maternity care for disabled women who experience domestic abuse: a systematic review. *BMC Pregnancy Childbirth*. 2014;14(1):234.

23. Mitra M, Lu E, Diop H. Smoking among pregnant women with disabilities. *Women's Heal Issues*. 2012;22(2):e233-e239.

24. Kitsantas P, Adams AR, Cheema J, Kornides ML. Opioid use in pregnant women with mental health-related disabilities. *Arch Gynecol Obstet*. 2021;303(6):1531-1537.

25. Kitsantas P, Aljoudi SM, Gimm G. Marijuana use in pregnant women with disabilities in the United States. *Matern Child Health J*. 2022;26(2):242-249.

26. Smeltzer SC, Mitra M, Iezzoni LI, Long-Bellil L, Smith LD. Perinatal experiences of women with physical disabilities and their recommendations for clinicians. *J Obstet Gynecol Neonatal Nurs*. 2016;45(6):781-789.

27. Page MJ, McKenzie JE, Bossuyt PM, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *BMJ*. 2021;372:n71.

28. Page MJ, McKenzie JE, Bossuyt PM, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *Syst Rev*. 2021;10(1).

29. Lockwood C, Munn Z, Porritt K. Qualitative research synthesis. *Int J Evid Based Healthc*. 2020;13(3):179-187.

30. Iezzoni LI, Wint AJ, Smeltzer SC, Ecker JL. Physical Accessibility of Routine Prenatal Care for Women with Mobility Disability. *J Women's Heal*. 2015;24(12):1006-1012.

31. Smeltzer SC, Wint AJ, Ecker JL, Iezzoni LI. Labor, delivery, and anesthesia experiences of women with physical disability. *Birth*. 2017;44(4):315-324.

32. Long-Bellil L, Mitra M, Iezzoni LI, Smeltzer SC, Smith LD. Experiences and unmet needs of women with physical disabilities for pain relief during labor and delivery. *Disabil Health J*. 2017;10(3):440-444.

33. McKay-Moffat S, Cunningham C. Services for women with disabilities: mothers' and midwives' experiences. *Br J Midwifery*. 2006;14(8):472-477.

34. Prilleltensky O. A ramp to motherhood: The experiences of mothers with physical disabilities. *Sex Disabil*. 2003;21(1):21-47.

35. Schildberger B, Zenzmaier C, König-Bachmann M. Experiences of Austrian mothers with mobility or sensory impairments during pregnancy, childbirth and the puerperium: A qualitative study. *BMC Pregnancy Childbirth*. 2017;17(1):1-11.

36. Tebbet M, Kennedy P. The experience of childbirth for women with spinal cord injuries: an interpretative phenomenology analysis study. *Disabil Rehabil*. 2012;34(9):762-769.

37. Nguyen T V., King J, Edwards N, Dunne MP. "Nothing suitable for us": experiences of women with physical disabilities in accessing maternal healthcare services in Northern Vietnam. *Disabil Rehabil*. 2020;44(4):573-581.

38. Magagula F, Temane A, Nolte AGW. Women with mobility disabilities' experiences of maternity care during pregnancy, labour and puerperium in Eswatini. *Heal SA Gesondheid*. 2022;27:1-9.

39. Apolot RR, Ekirapa E, Waldman L, et al. Maternal and newborn health needs for women with walking disabilities; "the twists and turns": a case study in Kibuku District Uganda. *Int J Equity Health*. 2019;18(1):43.

40. Mitra M, Akobirshoev I, Moring NS, et al. Access to and satisfaction with prenatal care among pregnant women with physical disabilities: Findings from a national survey. *J Women's Heal*. 2017;26(12):1356-1363.

CHAPTER III

ZINC'S EFFECT ON MENTAL HEALTH: A REVIEW

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1. Introduction

Zinc is a vital micronutrient for the optimal function of body tissues, particularly of brain. The amount of zinc (Zn^{2+}) present in the central nervous system is estimated to be 1.5% of the body's zinc content. This essential element plays a vital role in the regulation of signaling in the central nervous system. (1) It is abundant in the hippocampal and amygdala regions that constitute the limbic system of the brain and are related to learning, memory and emotional responses. (2) In addition to its role in the structure and function of many proteins and enzymes, zinc is important for many biological activities, including epigenetic regulations. (3) As more than 300 enzymes and more than 1000 transcription factors need zinc for their activities, the zinc deficiency in

human can lead to many health problems. Therefore, zinc supplementation may play a therapeutic role in the management of certain diseases. (4)

The best-known sources of zinc include meat products and seafood. (3) Zinc deficiency may occur due to inadequate dietary intake, absorption problems (malabsorption, excessive consumption of dietary phytate-rich cereals), hyperzincuria caused by some diseases (liver cirrhosis, sickle cell anemia), and over-sweating in tropical climates. (5) In this regard, especially infants and the elderly are more prone to zinc deficiency. (6) Cognitive impairment, immune dysfunction, delay in wound healing, loss of smell and taste may occur in all age groups depending on zinc deficiency. (7-8) Although zinc deficiency can be challenging diagnostically, it is a very common condition in many places of the world. (9) It has been reported that the prevalence of zinc deficiency is high (41%), especially in psychiatric diseases in which mental health is impaired. (10) The high prevalence of zinc deficiency can be explained by a dual link between zinc level and mental disorders. Low zinc levels trigger mental disorders. Some clinical trials show, however, that mental disorders can reduce zinc levels. (11-12)

In addition to its deficiency, excessive intake of zinc can also cause pathological effects on the nervous system. It has been shown that zinc deficiency in vivo leads to mental lethargy, reduced nerve conduction and neuropsychiatric disorders while an excess of zinc intake causes neurotoxicity and neuron death. (12) The most studied central nervous system disorders associated with the impairment of zinc homeostasis are depression, schizophrenia, attention deficit hyperactivity disorder and Alzheimer's disease. This review discusses the effect of zinc on each of these disorders and the potential underlying mechanisms in the light of current literature.

2. Potential Mechanism of Zinc Affecting Mental Health

Potential mechanisms of zinc for mental health could be explained through its high affinity for metalloproteins in synaptic vesicles, its antioxidant and anti-inflammatory properties, its role in regulating neurotransmitter and its properties of glutamate receptor inhibition and protein kinase inhibition.

2.1. Property of Binding to Metalloproteins in Synaptic Vesicles

Zinc ions are transported into synaptic vesicles through binding to the zinc transporter protein (Zinc Transporter Protein, ZnT3) in the brain and are implicated in neural function. The zinc concentration in the synaptic vesicles

depends on the expression of gene encoding the ZnT3 protein (Slc30a3 gene) and the concentration of ZnT3 protein. (13) A reduction in ZnT3 protein concentration brings about a decrease in zinc (Zn^{+2}) level in synaptic vesicles, and, therefore, a decline in cognitive performance. (14)

ZnT proteins are present in many other tissues such as adipose tissue, pancreatic beta cells, retina and testicles, as well as brain. Synthesis of the ZnT protein, which modulates the flow of zinc in neurons and other cells, is affected by age, sex, hormones and nutritional factors. Advanced age and female gender are non-modifiable risk factors that reduce ZnT protein synthesis. (15) It has been observed that ZnT protein synthesis increased in the brain of mice with lowered estrogen levels as a result of ovariectomy, whereas estrogen supplementation decreased the ZnT protein level in the brain. Therefore, with the effect of estrogen hormone on expression of ZnT protein, this hormone may make women more prone to neurodegenerative diseases than men. (16)

Inadequate and unbalanced nutrition is one of the modifiable risk factors that influence the synthesis of ZnT protein. Particularly fatty acids can affect both brain and plasma zinc homeostasis. The reason is that fatty acids and zinc are transported via the same protein (albumin) in the plasma. When fatty acids are high in plasma, the binding of zinc to albumin is impaired and plasma zinc level decreases, which in turn may influence zinc flow and signaling in various cells. (17) It has been reported that docosahexaenoic acid (DHA) deficiency, one of the omega-3 fatty acids, may lead to apoptotic cell death through changes in neuronal zinc flow and increase the risk of neurodegenerative diseases. (18)

Similarly, one of the underlying reasons for the increased risk of neurodegenerative diseases in individuals with chronic diseases like obesity and Type 2 Diabetes is explained by ZnT proteins. Insulin synthesis, release and function rely on zinc and ZnT proteins. Alterations in zinc and ZnT protein may have an effect on blood glucose levels and insulin activity. As such, blood glucose and insulin levels can negatively affect the synthesis of ZnT proteins. Chronic diseases such as obesity and Type 2 diabetes downregulate ZnT proteins, which in turn increases the risk of neurodegenerative diseases accompanying chronic diseases such as obesity and type 2 diabetes. (19-20) Zinc supplementation favorably alters the course of mental changes in obese and diabetic patients. Cavalcanti et al. (2020) found that 4 weeks-zinc supplementation (15 mg / kg of zinc gluconate or zinc sulfate per body weight per day) brought neurobehavioral and neuroprotective benefits to diabetic rats. (21) Another study reported that 12

weeks zinc gluconate supplementation (30 mg / day) significantly decreased the depression score in obese / overweight patients with depression symptoms. (22)

2.2. Antioxidant and Anti-inflammatory Properties of Zinc

Zinc is incorporated into the structure of metallothioneins that have a defensive function against oxidative damage and inflammation. (23) Metallothionines, acting as zinc reserve, bind cellular zinc with a high affinity. Although metallothioneins bind some other metals (copper, cadmium, etc.), the zinc-bound form is predominant in human tissue. (24) They control the concentration of zinc ions as metallothionines serve as both zinc acceptor and zinc donor. (23) Abnormal distribution and altered expression of metalloproteins (ZnT, metallothionein) regulating cellular zinc levels are observed in mental disorders. (25) Dysregulation of cellular zinc homeostasis and downregulation of metallothioneins in different neurological diseases increase susceptibility to oxidative stress and metal-induced neurotoxicity, which may negatively alter the course of central nervous system diseases. (26) Zinc can also protect the mental health by removing reactive oxygen species and preventing lipid peroxidation. (27) because increased malondialdehyde levels, which are indicator of lipid peroxidation, have been associated with the risk of major depressive disorder, post-stroke cognitive impairment and dementia. (28-30)

Recent human and animal studies have investigated the association of the positive effect of zinc on mental state with its antioxidant and anti-inflammatory properties. A double-blind placebo-controlled study by Jafari et al. (2020) revealed that 12 weeks zinc supplementation (30 mg of zinc gluconate) increased antioxidant capacity and brain-derived neurotropic factor in women with premenstrual syndrome while alleviating their physical and psychological symptoms. (31) Brzóska et al. (2021) found an increase in pro-oxidants and a decrease in antioxidants in the brains of rats that were administered with Cd in drinking water at the concentration of 5 mg/L for six months. Simultaneous administration of zinc (30 mg / L, 60 mg / L) and cadmium via drinking water provided a protection against cadmium accumulation and oxidative damage in brain tissue. Increasing zinc intake with cadmium exposure prevented oxidative / antioxidative imbalance in the brain. (32) De Oliveira et al. (2021) investigated the effects of zinc supplementation (10 mg / kg / day) on metabolic and neuroinflammatory parameters in cafeteria diet-induced obesity in rats. Zinc supplementation decreased the increase in interleukin-6 (IL-6) caused by cafeteria diet and the increase in toll-like receptor-4 (TLR-4) in the hippocampus

region of the brain. While recognition memory was impaired in cafeteria diet-fed rats, zinc supplementation reversed it. (33)

2.3. Role of Zinc in Neurotransmitter Regulation

Zinc is found at high concentrations in vesicles containing neurotransmitters. There, it affects the vesicle content and exocytotic oscillation dynamics. (34) It changes the distribution of lipids (phospholipids and cholesterol) in the cell membrane to produce that effect. (35) Fusion pores regulate synaptic power through the rate of neurotransmitter release and other changes in the exocytotic process. (34-35) In a study on aluminum-induced neurotoxicity, Lu et al. (2013) administered a diet containing zinc at different doses to rats for nine weeks. Dopamine and 5-hydroxytryptamine levels of brain were found to be higher in the group treated with high doses of zinc (100 mg / kg, 200 mg / kg) than in the group with lower dose of zinc (50 mg / kg). A study on the role of zinc in neurotransmission revealed that zinc supplementation may affect learning and memory while reducing neuronal apoptosis. (36) In another study, rats undergoing a chronic unpredictable stress procedure for 14 days were treated with zinc chloride (10 mg / kg) or fluoxetine (10 mg / kg) once a day between 8-14 weeks. The study showed that zinc supplementation could strongly reverse the behavioral change induced by the chronic unpredictable stress procedure through modulating glutamatergic neurotransmission. (37)

2.4. Role of Zinc as a Receptor and an Enzyme Inhibitor

Zinc, an N-methyl D-aspartate (NMDA) receptor antagonist, protects mental health by preventing glutamate excitotoxicity. Neuronal death resulting from glutamate exposure occurs through NMDA that is one of the glutamate receptors. Zinc, which inhibits NMDA activity, prevents neuronal damage and death by avoiding extracellular glutamate accumulation. (38) Doboszewska et al. (2015) examined changes in NMDA receptor in the brain regions of rats fed a zinc deficient diet (3 mg Zn / kg) and a zinc sufficient diet (50 mg Zn / kg) for 4-6 weeks. Insufficient dietary zinc intake caused up-regulation of NMDA receptor subunits (GluN2A and GluN2B) in the hippocampus of rats, thus provoking behavioral despair and a reduction of social behavior. They showed that zinc deficiency-induced depressive-like behavior is linked with alterations in the NMDA receptor signaling pathway. (39) Another study compared the hippocampus of people who died of suicide with that of people

in the sudden death control group. The potency of zinc to prevent [H^3]MK-801 binding to NMDA receptor in the hippocampus in suicide tissue was found to be 29% lower. It has been reported that changes in the zinc and NMDA receptor complex in the hippocampus play a role in the pathophysiology of suicide-related disorders. (40)

Zinc is also an inhibitor of glycogen synthase kinase 3 beta (GSK-3B), one of the protein kinase enzymes. Protein kinases phosphorylate proteins leading to functional changes. GSK-3B activity plays a role in the pathogenesis of Alzheimer's disease by leading to hyperphosphorylation of especially tau proteins. Furthermore, GSK-3B activity is regarded as one of the causes of bipolar disorder, schizophrenia, stroke, and depression. (41) It has been considered that, in diabetic patients, insulin signaling is impaired as well as abnormal GSK-3B activity and the risk of neurodegenerative disease increases. Zinc has been shown to inhibit GSK-3B activity and enhance glucose uptake under in vitro conditions. It has been recommended that zinc supplementation could be used therapeutically for glucose homeostasis and mental health in diabetic patients. (42)

3. Effect of Zinc on Different Mental Disorders

Zinc has an impact on depression, schizophrenia, attention deficit hyperactivity disorder and Alzheimer's disease.

3.1. Depression

Depression is a serious psychiatric disorder that affects millions of people worldwide. (43) Zinc is considered to play an important role in the pathophysiology and treatment of the disease. (44) Serum zinc levels are generally low in individuals diagnosed with depression. A meta-analysis study found the average zinc concentration of patients diagnosed with depression to be 1.85 μmol/L lower than the healthy group. As the severity of depression increased, the difference in the serum zinc concentrations between the group diagnosed with depression and the healthy group had risen. (45) Another meta-analysis study showed high levels of zinc intake were associated with a 28% reduction in the risk of depression. (46)

The fact that zinc is an antagonist of glutamatergic system is viewed as the main mechanism explaining its antidepressant property. (43) In addition, it has been shown that zinc can play antidepressant roles by increasing the

expression of brain-derived neurotropic factor (BDNF), which is responsible for the growth and function of neuronal cells. Antidepressant drugs also exert their effects through increasing BDNF expression. In this context, zinc might be a safe and effective treatment method that would be used in combination with antidepressant drugs or used instead of these drugs. (47) Irmisch et al. (2010) suggested zinc may produce an effect on depression by altering fatty acid metabolism. They observed that as the zinc concentration decreased in depressive patients, the concentration of dihomo-gamma linoleic acid (DGLA) increased. This fatty acid promotes the synthesis of prostaglandin E2 (PGE2), which is an important trigger for major depression. Therefore, they reported that zinc may exert its effect on depression through fatty acids that increase PGE2 synthesis. (48)

When the nutritional status of depression patients is evaluated by the 24-hour dietary recall method, it is reported that increase their unhealthy food intake as a response to negative emotions and their intakes of micronutrients, including zinc, are lower than those of healthy controls. (49) Zinc supplementation has a positive effect on depressive patients. In a study by Ranjbar et al. (2013), 44 patients diagnosed with major depression were divided into two groups, in which one took zinc supplements (25 mg) in addition to antidepressant medication for 12 weeks. They observed that Beck depression inventory score of the zinc supplement group decreased significantly compared to the placebo group by the end of the 12th week. (11) In a study of a patient with opioid use disorder receiving methadone, the treatment group was given 25 mg zinc sulfate combined with methadone. As a result of the study, it was found that there was a significant decrease in depression, anxiety and stress in the treatment group compared to the control group who received only methadone. (50) In another study by Salari et al. (2015), 43 patients diagnosed with multiple sclerosis with major depressive disorder were divided into two groups, in which one received 220 mg of zinc sulfate (containing 50 mg of zinc) for 12 weeks. Although there was no difference between the neurological examination results of the control and treatment groups by the end of the 12th week, the depression score of zinc supplement group decreased compared to the control group. (51) Available evidence from observational studies and randomized controlled trials has suggested that it supports the potential benefits of zinc for reducing the risk of and alleviating depression. (22) Antidepressant drugs could have side effects that stimulate the release of zinc from zinc stores in body tissues such as muscle and bone. For this reason, it is also considered that, especially when

administered with drugs, zinc supplements could replenish depleted tissue zinc pools and promote a reduction in symptoms of depression. (11)

3.2. Schizophrenia

Multiple risk factors contribute to schizophrenia, including genetic susceptibility and environmental factors. It can occur in childhood, adolescence, or adulthood and cause disabling conditions that affect the quality of life for those who suffer from it. (52) Although there are many etiological factors that induce the development of schizophrenia, maternal infection is regarded as an important risk factor for the development of schizophrenia in children. There is mounting evidence that zinc levels may be associated with maternal exposure to infection. In an animal study in which lipopolysaccharides were used as infectious agents in the prenatal period, changes were observed in the zinc level in some brain regions. (53) A more recent animal study has concluded that zinc supplementation in combination with exposure to lipopolysaccharides during pregnancy might mitigate neurodevelopmental disorders caused by lipopolysaccharides. (54)

Patients with schizophrenia make poor dietary choices, so they are at higher risk of obesity, malnutrition, nutritional deficiencies and excesses. (55-56) Similar to patients diagnosed with depression, serum zinc levels are generally low in patients diagnosed with schizophrenia. A meta-analysis study determined that the mean serum zinc concentration of patients diagnosed with schizophrenia was 12.81 μg / dL (1.96 μmol / L) lower than the healthy group. (57) Many factors may lead to changes in zinc level in this patient group. According to one opinion, changes in zinc level may result from transporters affecting inflammation and the glutamatergic system. (57) The drugs used can also affect this condition. Valproate drug used to treat schizophrenia has been shown to reduce zinc levels in neural cells. (58) It has also been reported that 50 mg of zinc supplementation per day combined with medications in schizophrenic patients could increase response to antipsychotic drugs without any significant side effects. (59) In a study conducted with schizophrenia patients, the relationship between serum concentrations of 11 essential metal elements such as Ca, K, Zn, and Fe and the risk of schizophrenia was investigated. The findings of the study showed that although the Zn concentration had a significant difference between patients and healthy group, it was not statistically significant. In conclusion, essential metal elements have been suggested to have the potential to be biomarkers for improving the diagnosis and treatment of schizophrenia. (52)

3.3. Attention Deficit Hyperactivity Disorder

Attention deficit hyperactivity disorder (ADHD) is the most common neurobehavioral disorder of childhood. It is thought that micronutrient deficiencies may contribute significantly to the pathogenesis of this disease, as in other mental disorders. (60) Patients with ADHD may not be able to provide the attention needed to sit through a meal to ensure adequate nutrient intake. Moreover, they may be at risk for a variety of nutrient deficiencies because of appetite suppressant effect of treatment medication. (61) Different results have been reported regarding the zinc levels of children with ADHD. A meta-analysis study comparing 2177 children with ADHD and 2900 healthy children in China reported that serum zinc levels were lower in children with ADHD. Low serum zinc levels have been associated with susceptibility to ADHD. (62) On the contrary, a more recent meta-analysis study found that blood and hair zinc levels of children and adolescents with ADHD were not different from those of healthy children and adolescents. (63) Yet another study showed that serum zinc concentrations were low in children diagnosed with ADHD, and reduced concentrations were associated with the inattention subscale score of the ADHD scale developed by parents and teachers. (64)

It has been supported by many studies that zinc supplementation could be effective in alleviating symptoms of ADHD and might increase response to drug therapy in children diagnosed with Attention Deficit Hyperactivity Disorder. After children with zinc deficiency and ADHD took 55 mg / kg of zinc supplements for six months, their hyperactivity, impulsivity, and emotional lability scores improved according to Conners' parental rating scale. However, there was no change in IQ scores according to the Wechsler IQ test. (65) Children with ADHD treated with methylphenidate were split into two groups in a double-blind randomized controlled trial. One group was supplemented with zinc for six weeks whereas the other group was designated as the control group. While zinc supplementation did not affect their hyperactivity and impulsivity scores assessed with the ADHD scale, it improved their inattention scores. (66) 150 children aged 6-15 years, diagnosed with ADHD and used methylphenidate, were divided into one control group and two treatment groups in another randomized double-blind study. Treatment groups received zinc sulfate or omega-3 supplements for eight weeks. Then, groups were compared with the Conners' Parent and Teacher Rating Scale. Whereas there was no difference in the average Conners' scale scores between the control group and the group receiving omega-3 supplement, a significant improvement was observed in

the scores of the zinc supplement group compared to the control group. Zinc supplementation accompanied by the basic therapy significantly improved symptoms of ADHD. (67)

3.4. Alzheimer's Disease

Alzheimer's disease (AD) is one of the most common types of dementia. Dysregulation of metals in the brain is one of its causes. Zinc in the brain is implicated in the pathogenesis of AD by altering amyloid metabolism. Zinc also regulates synaptic function, neurotrophic signals, synaptic plasticity, and brain copper homeostasis. (68) In previous study, serum glutamate decarboxylase (GADA), Zinc transporter 8 autoantibodies (ZnT8A), Zn and vitamin D levels were evaluated in 40 AD patients and control groups. GADA and ZnT8A were found to show higher serum levels and lower serum Zn concentrations in the patient group. Accordingly, the authors suggested that the parameters could be discussed as possibly diagnostic in AD cases. (69)

As the alzheimer's disease advances, patients start to have difficulty swallowing and lose interest in eating. (70) This leads to inadequate nutrient intake and undesirable weight loss. (71) Serum zinc levels are generally low in AD patients similar to many neurological diseases. A meta-analysis study reported that the average serum and plasma zinc levels in AD patients were 0.46 μmol / L and 0.18 μmol / L, respectively, lower than healthy controls. The results have been interpreted in the manner of inadequate dietary zinc intake in AD patients and / or of the binding of zinc to plaques formed in AD. (72)

Animal and human studies have shown that zinc supplementation may positively alter the course of AD. AD patients with mild dementia received 150 mg / day zinc formula (adeona) for 6 months. A significant improvement was observed in the results of 'Alzheimer's Disease Rating Scale' and 'Clinical Dementia Rating Scale' measured in patients aged 70 and over. (73) Although there was no change in the behavior of mice that were administered with 2g / L of zinc acetate in drinking water for six months, a decrease was observed in beta amyloid accumulation. (74) In another study, mice received 30 mg / L of zinc sulfate in tap water for 11-13 months. While there was an improvement in mitochondrial function and spatial memory and an increase in brain-derived neurotrophic factor, the pathology of beta amyloid and tau protein in the hippocampus considerably reduced. (75) It is also considered that zinc supplementation might prevent copper toxicity in plasma and brain and regulate neuronal health in this way. (68)

4. Conclusion

Recent studies have provided robust evidence that link impaired zinc homeostasis to numerous neuropsychiatric disorders. Zinc affects mental health in a multifaceted manner, including neurotransmitter system, glutaminergic system, antioxidant and anti-inflammatory pathways. People with of severe mental illness have poor dietary patterns. The changes in zinc levels seen in these diseases may be the result of poor dietary quality. Intervention programs to change dietary patterns among people with mental illness might be useful for positively affect the course of the disease and meet the nutrient requirement, including zinc. In addition, zinc levels may decrease with the effect of drugs used for treatment in patients. Therefore, it is important for serum zinc levels to routinely evaluate in neurology and psychiatric practices. Moreover, the negative effects of drugs used in the treatment of mental disorders on zinc levels indicate the necessity of targeting new pharmacological approaches to treat these disorders. Pharmacotherapy is a costly treatment with side effects. Due to its positive effects on the course of mental disorders, zinc supplement can be deemed in terms of use instead of drugs. Or, the use of zinc supplements with drugs to reduce the side effects of drugs and mitigate the symptoms of neuropsychiatric diseases may be complementary. Further studies are needed that clarify the effect of zinc on the central nervous system diseases and determine the groups that would most benefit from zinc supplementation.

REFERENCES

1. Mansour HM. Nutrition and Brain Functions in Health and Disease. Mohamed W, Kobeissy F, Eds. Nutrition and Psychiatric Disorders. Springer Nature Singapore: Singapore; 2022: 3-26.

2. Takeda A, Tamano H. Insight into zinc signaling from dietary zinc deficiency. Brain Res Rev. 2009; 62(1): 33-44.

3. Gu HF, Zhang X. Zinc Deficiency and Epigenetics. Preedy V, Patel VB, Eds. Handbook of Famine, Starvation, and Nutrient Deprivation: From Biology to Policy. Sweden, Springer International Publishing AG: Switzerland; 2017: 1-18

4. Prasad AS. Discovery of human zinc deficiency: its impact on human health and disease. Adv Nutr. 2013; 4(2): 176-190.

5. Prasad AS. Zinc in human health: effect of zinc on immune cells. Mol Med. 2008; 14(5): 353-357.

6. Yasuda H, Tsutsui T. Infants and elderlies are susceptible to zinc deficiency. Sci Rep. 2016; 6: 21850.

7. Lin PH, Sermersheim M, Li H., et al. Zinc in wound healing modulation. Nutrients, 2018;10(1): 16.

8. Pisano M, Hilas O. Zinc and taste disturbances in older adults: a review of the literature. Consult Pharm, 2016; 31(5): 267-270.

9. Corbo MD, Lam J. Zinc deficiency and its management in the pediatric population: a literature review and proposed etiologic classification. J Am Acad Dermatol. 2013; 69(4): 616-624.

10. Grønli O, Kvamme JM, Friborg O, Wynn R. Zinc deficiency is common in several psychiatric disorders. PloS One, 2013; 8(12): e82793.

11. Ranjbar E, Kasaei MS, Mohammad-Shirazi M, et al. Effects of zinc supplementation in patients with major depression: a randomized clinical trial. Iran J Psychiatry. 2013; 8(2): 73.

12. Bartzatt R. Neurological impact of zinc excess and deficiency in vivo. Eur J Nutr Food Saf. 2017; 155-160.

13. Linkous DH, Flinn JM, Koh JY, et al. Evidence that the ZNT3 protein controls the total amount of elemental zinc in synaptic vesicles. J Histochem Cytochem. 2008; 56(1): 3-6.

14. Sindreu C, Palmiter RD, Storm DR. Zinc transporter ZnT-3 regulates presynaptic Erk1/2 signaling and hippocampus-dependent memory. Proc Natl Acad Sci. 2011; 108(8): 3366-3370.

15. Smidt K, Rungby J. ZnT3: a zinc transporter active in several organs. Biometals. 2012; 25(1): 1-8.

16. Lee JY, Kim JH, Hong SH, et al. Estrogen decreases zinc transporter 3 expression and synaptic vesicle zinc levels in mouse brain. J Biol chem, 2004; 279(10): 8602-8607.

17. Coverdale JP, Khazaipoul S, Arya S, Stewart AJ, Blindauer CA. Crosstalk between zinc and free fatty acids in plasma. Biochim Biophys Acta Mol Cell Biol Lipids. 2019; 1864(4): 532-542.

18. Suphioglu C, De Mel D, Kumar L, et al. The omega-3 fatty acid, DHA, decreases neuronal cell death in association with altered zinc transport. FEBS Lett. 2010; 584(3): 612-618.

19. Myers SA. Zinc transporters and zinc signaling: new insights into their role in type 2 diabetes. Int J Endocrinol. 2015.

20. Olesen RH, Hyde TM, Kleinman JE, Smidt K, Rungby J, Larsen A. Obesity and age-related alterations in the gene expression of zinc-transporter proteins in the human brain. Transl Psychiatry. 2016; 6(6): e838-e838.

21. Cavalcanti CL, Gonçalves MCR, Alves AF, et al. Antidepressant, anxiolytic and neuroprotective activities of two zinc compounds in diabetic rats. Front Neurosci. 2020; 13: 1411.

22. Yosaee S, Soltani S, Esteghamati A, et al. Effects of zinc, vitamin D, and their co-supplementation on mood, serum cortisol, and brain-derived neurotrophic factor in patients with obesity and mild to moderate depressive symptoms: A phase II, 12-wk, 2× 2 factorial design, double-blind, randomized, placebo-controlled trial. Nutr. 2020; 71: 110601.

23. Jarosz M, Olbert M, Wyszogrodzka G, Mlyniec K, Librowski T. Antioxidant and anti-inflammatory effects of zinc. Zinc-dependent NF-κB signaling. Inflammopharmacology, 2017; 25(1): 11-24.

24. Hennigar SR, Kelley AM, McClung JP. Metallothionein and zinc transporter expression in circulating human blood cells as biomarkers of zinc status: a systematic review. Adv Nutr. 2016; 7(4): 735-746.

25. Prakash A, Bharti K, Majeed ABA. Zinc: indications in brain disorders. Fundam Clin Pharmacol. 2015; 29(2): 131-149.

26. Juárez-Rebollar D, Rios C, Nava-Ruíz C, Méndez-Armenta M. Metallothionein in brain disorders. Oxid Med Cell Longev. 2017; Article ID 5828056

27. Mansour SA, Mossa ATH . Lipid peroxidation and oxidative stress in rat erythrocytes induced by chlorpyrifos and the protective effect of zinc. Pestic Biochem and Physiol. 2009; 93(1): 34-39.

28. Islam MR, Islam MR, Ahmed I, et al. Elevated serum levels of malondialdehyde and cortisol are associated with major depressive disorder: A case-control study. SAGE Open Med. 2018; 6: 2050312118773953.

29. Mahmoud W, Abd-Elgawad E, Mesallam D, El- Serafy T. Serum malondialdehyde as a predictor of post-stroke cognitive impairment in ischemic stroke patients. Zagazig Univ. Med J. 2022; 28: 326-332.

30. López-Riquelme N, Alom-Poveda J, Viciano-Morote N, Llinares-Ibor I, Tormo-Díaz C. Apolipoprotein E ε4 allele and malondialdehyde level are independent risk factors for Alzheimer's disease. SAGE Open Med. 2016; 4: 1-7.

31. Jafari F, Amani R, Tarrahi MJ. Effect of zinc supplementation on physical and psychological symptoms, biomarkers of inflammation, oxidative stress, and brain-derived neurotrophic factor in young women with premenstrual syndrome: A randomized, double-blind, placebo-controlled trial. Biol Trace Elem Res. 2020; 194(1): 89-95.

32. Brzóska MM, Kozłowska M, Rogalska J, Galazyn-Sidorczuk M, Roszcsenko A, Smereczański NM. Enhanced Zinc Intake Protects against Oxidative Stress and Its Consequences in the Brain: A Study in an In Vivo Rat Model of Cadmium Exposure. Nutrients. 2021; 13(2): 478.

33. de Oliveira S, Feijó GDS, Neto J, et al. Zinc Supplementation Decreases Obesity-Related Neuroinflammation and Improves Metabolic Function and Memory in Rats. Obes. 2021; 29(1): 116-124.

34. Ren L, Pour MD, Majdi S, Li X, Malmberg P, Ewing AG. Zinc Regulates Chemical-Transmitter Storage in Nanometer Vesicles and Exocytosis Dynamics as Measured by Amperometry. Angew Chem Int Ed. 2017; 56(18): 4970-4975.

35. Ren L, Dowlatshahi Pour M, Malmberg P, Ewing AG. Altered lipid composition of secretory cells following exposure to zinc can be correlated to changes in exocytosis. Chem Eur J, 2019; 25(21): 5406-5411.

36. Lu H, Hu J, Li J, et al. Optimal dose of zinc supplementation for preventing aluminum-induced neurotoxicity in rats. Neural Regen Res. 2013; 8(29): 2754.

37. Manosso LM, Moretti M, Colla AR, et al. Involvement of glutamatergic neurotransmission in the antidepressant-like effect of zinc in the chronic unpredictable stress model of depression. J Neural Transm. 2016; 123(3): 339-352.

38. Petrilli MA, Kranz TM, Kleinhaus K, et al. The emerging role for zinc in depression and psychosis. Front Pharmacol. 2017; 8: 414.

39. Doboszewska U, Sowa-Kućma M, Młyniec K, et al. Zinc deficiency in rats is associated with up-regulation of hippocampal NMDA receptor. Prog Neuropsychopharmacol Biol Psychiatry. 2015; 56: 254-263.

40. Sowa-Kućma M, Szewczyk B, Sadlik K, et al. Zinc, magnesium and NMDA receptor alterations in the hippocampus of suicide victims. J Affect Disord. 2013; 151(3): 924-931.

41. Khan I, Tantray MA, Alam MS, Hamid H. Natural and synthetic bioactive inhibitors of glycogen synthase kinase. Eur J Med Chem. 2017; 125: 464-477.

42. Ilouz R, Kaidanovich O, Gurwitz D, Eldar-Finkelman H. Inhibition of glycogen synthase kinase-3β by bivalent zinc ions: insight into the insulin-mimetic action of zinc. Biochem Biophys Res Commun. 2002; 295(1): 102-106.

43. Mlyniec K. Zinc in the glutamatergic theory of depression. Curr Neuropharmacol. 2015; 13(4): 505-513.

44. Nowak G. Zinc, future mono/adjunctive therapy for depression: mechanisms of antidepressant action. Pharmacol Rep. 2015; 67(3): 659-662.

45. Swardfager W, Herrmann N, Mazereeuw G, Goldberger K, Harimoto T, Lanctôt KL. Zinc in depression: a meta-analysis. Biol Psychiatry. 2013; 74(12): 872-878.

46. Yosaee S, Clark CC, Keshtkaran Z, Ashourpour M, Keshani P, Soltani S. Zinc in depression: From development to treatment: A comparative/dose response meta-analysis of observational studies and randomized controlled trials. Gen Hosp Psychiatry, 2022: 110-117.

47. Solati Z, Jazayeri S, Tehrani-Doost M, Mahmoodianfard S, Gohari MR. Zinc monotherapy increases serum brain-derived neurotrophic factor (BDNF) levels and decreases depressive symptoms in overweight or obese subjects: a double-blind, randomized, placebo-controlled trial. Nutr Neurosci. 2015; 18(4): 162-168.

48. Irmisch G, Schlaefke D, Richter J. Zinc and fatty acids in depression. Neurochem Res. 2010; 35(9): 1376-1383.

49. Kaner G, Soylu M, Yüksel N, Inanç N, Ongan D, Başmısırlı E. Evaluation of nutritional status of patients with depression. Biomed Res Int. 2015; 2015:521481

50. Amini Z, HeidariFarsani E. Investigating the effect of zinc supplementation on probability of relapse and mental health in patients with opioid use disorder undergoing methadone maintenance treatment. Subst Abuse Treat Prev Policy. 2023; 18(1): 1-8.

51. Salari S, Khomand P, Arasteh M, Yousefzamani B, Hassanzadeh K. Zinc sulphate: a reasonable choice for depression management in patients with multiple sclerosis: a randomized, double-blind, placebo-controlled clinical trial. Pharmacol Rep. 2015; 67(3): 606-609.

52. Ma J, Yan L, Guo T et al. Association between serum essential metal elements and the risk of schizophrenia in China. Sci Rep. 2020; 10(1): 1-11.

53. Tellez-Merlo G, Morales-Medina JC, Camacho-Ábrego I, et al. Prenatal immune challenge induces behavioral deficits, neuronal remodeling, and increases brain nitric oxide and zinc levels in the male rat offspring. Neurosci. 2019; 406: 594-605.

54. Alizadeh F, Davoodian N, Kazemi H, Ghasemi-Kasman M, Shaerzadeh F. Prenatal zinc supplementation attenuates lipopolysaccharide-induced behavioral impairments in maternal immune activation model. Behav Brain Res. 2020; 377: 112247.

55. Kim SR, Kim JY, Kim HY, Nho JH, Kim YH, Min SY. Factors related to malnutrition in community-dwelling patients with schizophrenia. Perspect Psychiatr Care. 2018; 55(3): 415-423

56. Onaolapo OJ, Onaolapo AY. Nutrition, nutritional deficiencies, and schizophrenia: An association worthy of constant reassessment. World J Clin Cases. 2021; 9(28): 8295.

57. Joe P, Petrilli M, Malaspina D, Weissman J. Zinc in schizophrenia: a meta-analysis. Gen Hosp Psychiatry, 2018; 53: 19-24.

58. da Silveira Paulsen B, Cardoso SC, Stelling MP, Cadilhe DV, Rehen SK. Valproate reverts zinc and potassium imbalance in schizophrenia-derived reprogrammed cells. Schizophr Res. 2014; 154: 30-35.

59. Mortazavi M, Farzin D, Zarhghami M, Hosseini SH, Mansoori P, Nateghi G. Efficacy of zinc sulfate as an add-on therapy to risperidone versus risperidone alone in patients with schizophrenia: a double-blind randomized placebo-controlled trial. Iran J Psychiatry Behav Sci. 2015; 9(3).

60. Skalny AV, Mazaletskaya AL, Ajsuvakova OP et al. Serum zinc, copper, zinc-to-copper ratio, and other essential elements and minerals in children with attention deficit/hyperactivity disorder (ADHD). J Trace Elem Med Biol. 2020; 58:126445

61. Kiddie JY, Weiss MD, Kitts DD, Levy-Milne R, Wasdell M.B. Nutritional status of children with attention deficit hyperactivity disorder: a pilot study. Int J Pediatr. 2010; 2010:767318.

62. Sun GX, Wang BH, Zhang YF. Relationship between serum zinc levels and attention deficit hyperactivity disorder in children. Zhongguo Dang Dai Er Ke Za Zhi. 2015; 17(9): 980-983.

63. Luo J, Mo Y, Liu M. Blood and hair zinc levels in children with attention deficit hyperactivity disorder: A meta-analysis. Asian J Psychiatry. 2020; 47: 101805.

64. Arnold LE, Bozzolo H, Hollway J, et al. Serum zinc correlates with parent-and teacher-rated inattention in children with attention-deficit/hyperactivity disorder. J Child Adolesc Psychopharmacol. 2005; 15(4): 628-636.

65. El-Baz FM, Youssef AM, Khairy E, Ramadan D, Youssef WY. Association between circulating zinc/ferritin levels and parent Conner's scores in children with attention deficit hyperactivity disorder. Eur Psychiatry. 2019; 62: 68-73.

66. Noorazar SG, Malek A, Aghaei SM, Yasamineh N, Kalejahi P. The efficacy of zinc augmentation in children with attention deficit hyperactivity disorder under treatment with methylphenidate: A randomized controlled trial. Asian J Psychiatr, 2020; 48: 101868.

67. Salehi B, Mohammadbeigi A, Sheykholeslam H, Moshiri E, Dorreh F. Omega-3 and Zinc supplementation as complementary therapies in children with attention-deficit/hyperactivity disorder. J Res Pharm Pract. 2016; 5(1): 22.

68. Sensi SL, Granzotto A, Siotto M, Squitti R. Copper and zinc dysregulation in Alzheimer's disease. Trends Pharmacol Sci. 2018; 39(12): 1049-1063.

69. Sadrirad S, Samadi N, Nikanfar M, et al. Evaluation of the Diagnostic and Predictive Value of Glutamic Acid Decarboxylase, Zinc Transporter 8 Serum Levels in Alzheimer's Disease. Clin Lab. 2022; 68(1).

70. Kalia M. Dysphagia and aspiration pneumonia in patients with Alzheimer's disease. Metab., 2003; 52:36-38.

71. Aziz NA, van der Marck MA, Pijl H, Olde Rikkert MGM, Bloem BR, Roos RAC. Weight loss in neurodegenerative disorders. J Neurol. 2008; 255(12): 1872-1880.

72. Ventriglia M, Brewer GJ, Simonelli I, et al. Zinc in Alzheimer's disease: a meta-analysis of serum, plasma, and cerebrospinal fluid studies. J Alzheimers Dis. 2015; 46(1): 75-87.

73. Brewer GJ. Copper excess, zinc deficiency, and cognition loss in Alzheimer's disease. Biofactors. 2012; 38(2): 107-113.

74. Harris CJ, Voss K, Murchison C, et al. Oral zinc reduces amyloid burden in Tg2576 mice. J Alzheimers Dis. 2014; 41(1): 179-192.

75. Corona C, Masciopinto F, Silvestri E, et al. Dietary zinc supplementation of 3xTg-AD mice increases BDNF levels and prevents cognitive deficits as well as mitochondrial dysfunction. Cell Death Dis. 2010; 1(10): e91-e91.

CHAPTER IV

BRONCHIECTASIS

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1. Introduction

Bronchiectasis usually refers to abnormal permanent dilatation of the bronchi accompanied by cough, sputum and recurrent infections. (1,2) The cause of dilatation is the destruction of the structures that provide muscle or elasticity in the bronchial wall. It is an important cause of morbidity that affects respiratory function and decreases the quality of life. (3,4) It was first reported by Laennac in 1819.(5,6) There is no definite information about its prevalence in the population. (7,8) While it was a disease with high morbidity in the past, it is thought that its prevalence decreased due to reasons such as increased access to and use of antibiotics (9), developments in preventive health services, and increased vaccination activities. However, there are some opinions that its prevalence has increased recently. (4,10) Although there are positive developments in the health system and access to the health system,(11) it is still an important health problem in underdeveloped and developing countries. (12,13) While the main cause is generally infection in underdeveloped or developing countries, it is reported as cystic fibrosis in developed countries.(14) It may be asymptomatic or severe such as massive haemoptysis. Radiological imaging methods play the main role in the diagnosis. Depending on the current medical condition, there are many options for treatment ranging from lifestyle changes to medical treatment and even surgery.

2. Etiology and Pathophysiology

Although it can be seen in all age groups, it is more common in the elderly population and female gender.(1,3,10) It is most commonly observed in the

lower lobe of the left lung. (12,15) It is more common in the lower lobe of the left lung because of the better drainage of the right main bronchus, vascular compression to the left main bronchus, and the anatomical structure of the left main bronchus. (12) Although it is usually unilateral, it has been reported to be bilateral in 30-50% of cases. (11) Medium-diameter bronchi are most commonly affected. (12)

Permanent abnormal expansion occurs after the structures that provide elasticity in the bronchial wall lose their strength by being affected by current processes. It is thought that the causes of the pathology are obstruction in the respiratory tract (intrabronchial or extra bronchial), recurrent infection, impairment of mucociliary activity, immune response, peribronchial fibrosis processes. (3,12,15) Pulmonary functions are impaired due to these causes.

The main reasons are; Idiopathic, causes of airway obstruction (foreign body, neoplasm, mucoid plugs, acquired tracheal stenosis, mediastinal lymph adenopathy), infectious diseases (childhood, bacterial, viral, fungal), congenital-immune-genetic-hereditary diseases (Mounier-Khun, Kartagener, cystic fibrosis, alpha 1 antitrypsin deficiency, agammaglobulinaemia, etc.), inflammation, pulmonary diseases, inhalation damage. The most common cause is recurrent infections. (1,3,4,15)

3. Clinic

It may be asymptomatic in some patients or it may be very severe. Patients classically complain of cough and sputum. The most common symptoms are productive cough with copious sputum (most common), haemoptysis, chest pain, dyspnoea, cyanosis, clubbing, bad breath, weakness, and weight loss. (1,3,15) Haemoptysis may sometimes be minor enough to be mixed with sputum, while sometimes massive haemoptysis may be seen at a life-threatening level. The main complications are infectious processes (abscess, pneumonia, empyema, sepsis, mediastinitis) and haemoptysis and fistula. (15) Different classifications are used according to their prevalence, etiological cause and clinical severity. One of the most well-known classifications is the radiology-based classification defined by Reid.[8] In this classification, types are cylindrical, varicose, and saccular (cystic). The cylindrical type is less severe, while the saccular type is more severe. In some cases, vanishing bronchiectasis (pseudo bronchiectasis) may be observed. (12,15)

Table 1: Classification

Cylindrical	Bronchial branching number within normal limits, minimal dilatation
Varicose	Bronchial branching number moderately reduced, mild diffuse dilatation
Saccular	Bronchial branching number is severely reduced, common dilatation
Pseudobronchiectasis	Temporary and disappears after a while

(15)

4. Diagnosis

Physical examination may not detect any pathological findings. Coarse rales may be heard in expiration. Clubbing, wheezing, cyanosis, and cor pulmonale findings may be observed. Haemogram, Ig levels, and allergic aspergillus test can be examined in laboratory tests. Some speciality associations recommend serological-immunological testing for diagnostic purposes. (3) However, there is no biomarker accepted by all groups in the used clinical follow-up of the disease. (10)

Although clinical and laboratory findings create suspicion, the definitive diagnosis is made by radiological methods. While bronchography was used for diagnostic purposes in the past, chest radiography and HRCT (the gold standard) are used in the current approach. No obvious pathology may be seen on chest radiography, tram track appearance (thin parallel lines) and honeycomb appearance can be seen. Bronchiectatic areas are best visualised on tomography (high resolution). The typical appearance of bronchiectasis is ‘signet ring sign’ (with a large bronchial area and accompanying pulmonary artery branch) in the tomography section.

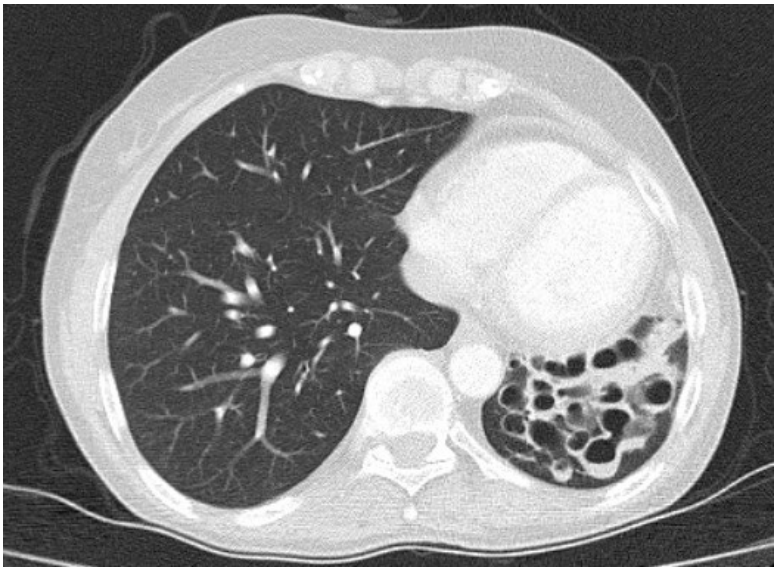

Figure 1: Bronchiectasis in the lower lobe of the left lung on thorax CT

5. Treatment

Treatment methods are; Elimination of the cause if possible, patient education, preventive measures, control of infection, airway cleaning, medical treatments (anti-biotherapy, anti-inflammatory therapy, bronchodilator, vaccination), respiratory exercises, physical therapy, bronchoscopy, surgery and symptomatic-supportive treatment (in advanced disease). (1,12,15)

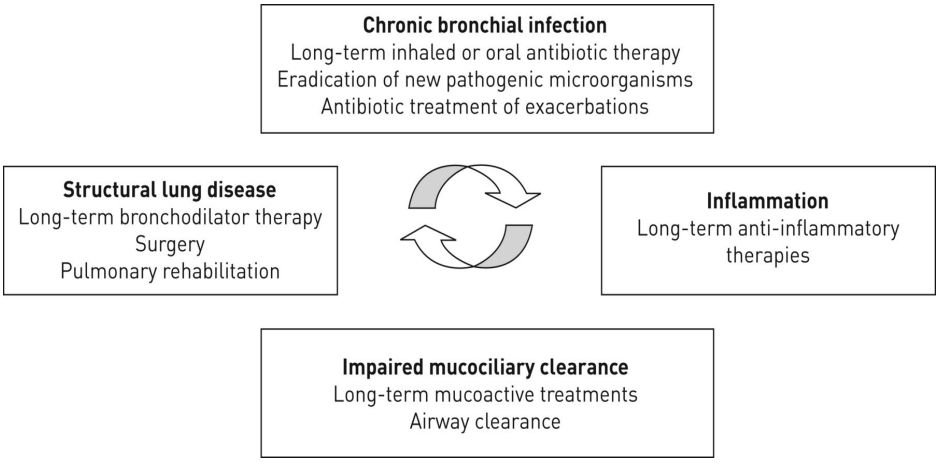

Figure taken from the European Respiratory Society Guideline 2017. (16)

Figure 2: Treatment strategy

Surgical treatment is usually performed due to failure of medical treatment and complications. (5) Surgical treatment can be performed for complications, resection, and transplantation. The aim of surgical treatment is to remove the tissue that has lost its function and to prevent contamination of the surrounding tissue (15), and combat complications such as haemoptysis. Surgical treatment should have low mortality and morbidity and improve the patient's quality of life. (16) Anatomical resections should be performed in patients in whom resection is considered. Successful results are obtained with anatomical resection in patients with localised disease. Another consideration is to pay attention to the preoperative nutrition of the patient and to plan the operation during the dry period of the disease. Resection treatment cannot be performed in all patients. The patient decision, the patient's current respiratory reserve, medical condition and the extent of the disease affect the surgical decision. Although bilaterality is not a definite contraindication, surgery is not considered in bilateral diffuse disease. (15)

6. Conclusion

Bronchiectasis is a disease characterised by abnormal enlargement of the bronchial system, diagnosed mostly by radiological methods, with medical and surgical treatment options.

References

1. Abul Y. Bronşektazi. In: Dalar L, Süerdem M, Öztürk C, Saygı A eds. Göğüs Hastalıkları. İstanbul: İstanbul Medikal Sağlık ve Yayıncılık; 2015:429-435.

2. Mitchell JD, Sherwood JT (Çev:Karamustafaoğlu YA). Bronşektazide Cerrahi. In: Erişkin göğüs Cerrahisi. Sugarbaker DJ, Bueno R, Krasna MJ, Mentzer SJ, Zellos L (Çev:Yüksel M) eds . Nobel Tıp Kitapevleri; 2011:702-708.

3. Bird K, Memon J. Bronchiectasis. [Updated 2023 Feb 23]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2023 Jan-. Available from: https://www.ncbi.nlm.nih.gov/books/NBK430810/

4. Visser SK, Bye P, Morgan L. Management of bronchiectasis in adults. Med J Aust. 2018 Aug 20;209(4):177-183. doi: 10.5694/mja17.01195. PMID: 30107772.

5. Özkan M, Sakallı MA, Yenigün BM, Kocaman G, Yüksel C et al. Surgical Treatment in Bronchiectasis: Results of 191 Patients". J. Ank. Univ. Fac. Med. 2019; 72(2): 241-246 . doi: 10.4274/atfm.galenos.2019.35744.

6. McShane PJ, Tino G. Bronchiectasis. Chest. 2019: 155(4): 825-833 doi: 10.1016/j.chest.2018.10.027.

7. Ulutaş H. Bronşektazi. In: Hacıevliyagil SS, Ulutaş H eds. Göğüs Hastalıkları&Cerrahisi Temelleri. Malatya: İnönü Üniversitesi Yayınevi; 2022:225-232.

8. Eldem B, Yıldızeli B. Bronşektazi: Etiyoloji ve Sınıflandırma. Turkiye Klinikleri J Thor Surg-Special Topics. 2012;5(1):1-8.

9. Bilgin M. Akciğerin enfeksiyon Hastalıklarında Cerrahi Yaklaşımların Temel Prensipleri. In: Özdülger A ed. Stajer Kitabı. İstabul: Aves; 2012:345.

10. O'Donnell AE. Bronchiectasis update. Curr. Opin. Infect. Dis. 2018; 31(2): 194-198. doi: 10.1097/QCO.0000000000000445.

11. Sayır F. Our Results of Surgical Management in Bronchiectasis. The Eurasian Journal of Medicine. 2007; 39:109-111.

12. Enön S, Kavukçu HŞ. Bronşektazi. In: Ökten İ, Kavukçu HŞ eds. Göğüs Cerrahisi. İstabnul: İstanbul tıp Kitabevi;2013:997-1006.

13. Çokuğraş H, Akçakaya N, Söylemez Y, Dayıoğlu E, Kulak K et al. Evaluation of Our Cases With Bronchiectasis in The Last Ten Years. GKD Cer. Derg. 1994; 2:371-374.

14. Bülbül Y, Erçen Diken Ö, Uğur Chousein EG; ASYOD Akciğer İnfeksiyonları ve Tüberküloz Bilim Kurulu. Ulusal yayınlar ışığında Türkiye'de bronşektazi. Tuberk Toraks 2020;68(1):48-65. doi: 10.5578/tt.69231.

15.Ulutaş H. Bronşektazi. In: Yüksel M, Balcı AE eds. Göğüs Cerrahisi 'Kırmızı Kitap'. 2nd edition. İstanbul: Nobel Tıp Kitapevleri;2015:265-272

16. Polverino E, Goeminne PC, McDonnell MJ, et al. European Respiratory Society guidelines for the management of adult bronchiectasis. Eur Respir J 2017; 50: 1700629 [https://doi.org/ 10.1183/13993003.00629-2017].

CHAPTER V

PULMONARY HYDATID CYST

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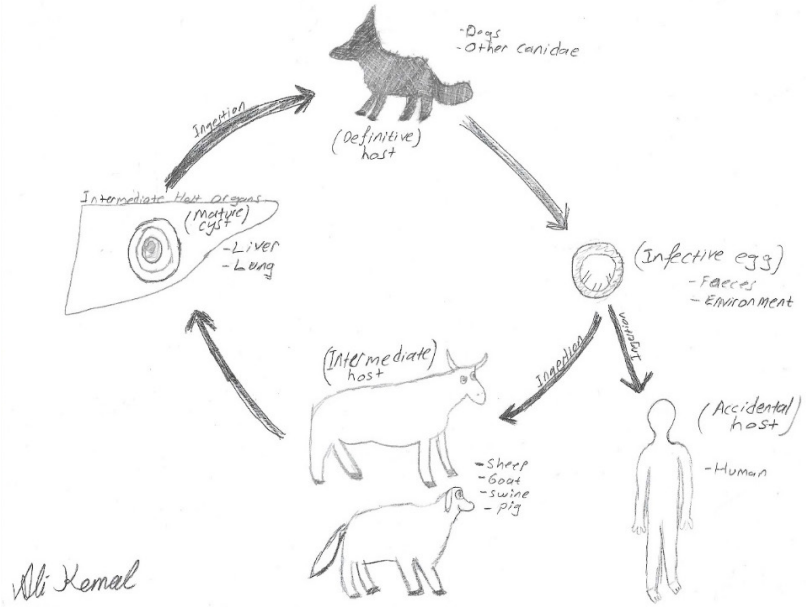
1. Introduction

Hydatid cyst is a parasitic disease caused by Echinococcus larvae (most commonly Echinococcus granulosus). (1,2) The parasite eggs spread with the faeces of the carnivores, the last host, are taken by the intermediate host and these eggs are absorbed from the intestines and reach the internal organs and cause cystic disease. (Figure 1)



(3)

Figure 1: Schematisation of the Hydatid Cyst Cycle

The eggs, which enter the circulation through the portal venous system, most commonly cause disease in the liver (60-70%) and less frequently in the lungs (30%).(4,5) In the pediatric age group, on the contrary, it is frequently localised in the lung.(4,6,7) It can infect many organs.(5) It is endemic in some regions.(8,9) Dealing with animal-related professions is a risk factor.(1) The probability of occurrence increases in regions where animal husbandry is common and in the male gender.(10) The prevalence in the world can not be known precisely since proper recording is impossible.(5) The prevalence in our country is thought to be 0.00050. (11) In general, the ratio is thought to be 0.8-11%, although it varies between regions. (4,5)

The wall structure of the hydatid cyst consists of 3 layers: pericyst (adventitia), laminar membrane and endoxide (germinal membrane). (Figure 2) Pericyst is a structure belonging to the intermediate host itself, it protects the cyst. Laminar membrane has a permeable structure. The germinative membrane is the structure that allows the parasite to reproduce.

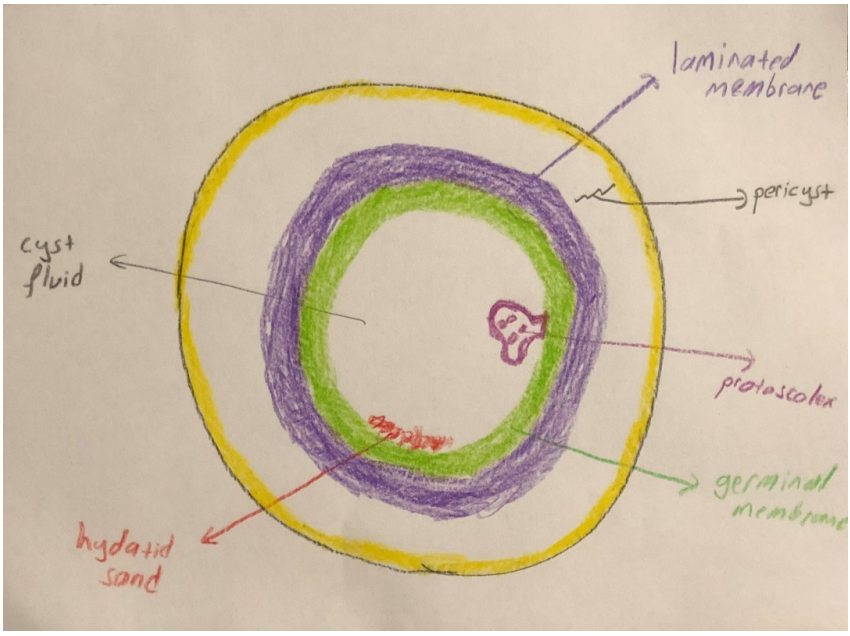


Figure 2: Wall structure of hydatid cyst

2. Clinic

It may be detected incidentally. It may be asymptomatic (approximately half of them) (5) or it may cause a severe clinic by being affected by many

factors such as its localisation, size, whether it ruptures or not, and the function of the organ in which it is located. The main symptoms are abdominal pain, jaundice, and vomiting, but the symptoms of the respiratory system are shortness of breath, chest pain, cough, and haemoptysis. (4,5,10) When the lesions in the lung reach 5 cm in diameter, bronchial compression is thought to occur. (9) The lesions in the lung are usually solitary, more common in the right lung (52.7-63.2%) and more common in the lower lobes. (5,10,11) Bilateral rates have been reported as 2-14.8%. (11)

Due to the elastic and soft structure of the lung, it can grow rapidly and reach large sizes. Perhaps this is why they give symptoms earlier than the liver. Cysts larger than 10 cm are called giant hydatid cysts by some authors. (12) When infected, it may lead to infection findings such as a lung abscess. It is also reported that they may cause bronchiectasis, empyema, fistula, amyloidosis etc. secondary to infection. (5) Cysts close to the respiratory tract may be expectorated; clear cyst fluid and membrane may come out with coughing. It is reported that it may cause horner or vena cava superior syndrome due to compression. (5) One of the most feared complications is anaphylaxis due to the rupture of the cyst (4). Rupture of pulmonary hydatid cysts has been reported with a rate of 6.2-48.3%. (11) When ruptured, it may cause cough, haemoptysis, chest pain, fever, symptoms related to anaphylaxis and pathologies such as hydropneumothorax. (5,9) Rupture may lead to very serious consequences and may even cause death.

3. Diagnosis

Although symptoms are important for the diagnosis, radiological imaging is the main factor. Auxiliary laboratory and serological tests are also used. However, the use of these tests for diagnostic purposes is controversial due to the fact that they are diagnostic in half of the cases and have high false positivity (10). Diagnosis may be difficult in some cases and may not even be diagnosed before the operation (5). In laboratory studies, haemogram (eosinophilia is less common), or staining of expectoration material can be examined. (5) In addition, complement fixation test, indirect haemagglutination (IHA) test, ELISA etc. are used among serological tests, but these tests are mostly used in treatment follow-up. (5)

The leading role in the diagnosis is radiological imaging. On radiological imaging (X-ray), they present themselves as oval, smoothly circumscribed, homogenous lesions (Figure 3). Sometimes they have specific appearances

such as Sunset sign, Escurado-Nemerow, crescent sign, lotus, double dome. Computed tomography and MRI can be used diagnostically and to evaluate the relationship with neighbouring organs. Although good information can be obtained with USG in the evaluation of liver lesions, USG is not routinely used in the evaluation of the lung. Unlike the liver, biopsies should be avoided to prevent rupture of lesions in the lung. (10)

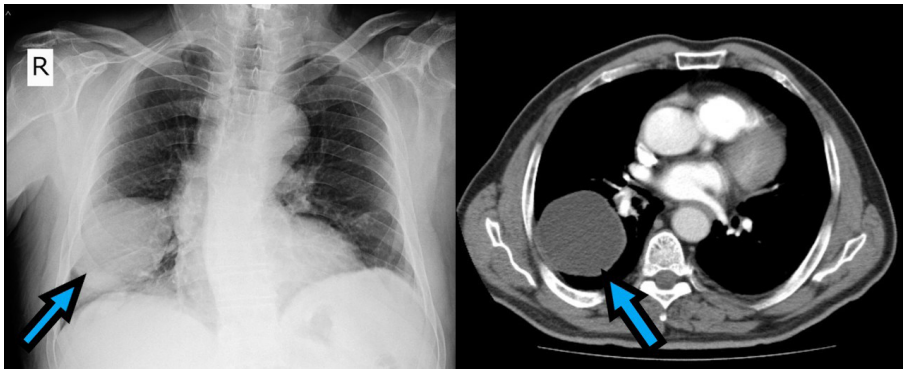


Figure 3: Radiological images of pulmonary hydatid cyst

4. Treatment

Apart from the measures to be taken to prevent the disease, there are medical and surgical treatment options. Surgery is the main treatment for pulmonary hydatid cysts (6, 10, 13, 14). Medical treatment can be used in some special cases such as when surgery cannot be performed due to cardiac-pulmonary patient capacity, when the patient does not want surgery, when it is not technically possible to remove the cyst (unresectable or so much numerous that the entire cystes cannot be removed) and in addition to surgery.

The active ingredients used in medical treatment are albendazole and mebendazole. Albendazole is thought to be superior to mebendazole. (6) Medical treatment may cause cyst rupture. (6)

The main aim of surgery is to remove the parasite and preserve as much lung tissue as possible. While surgical methods were thoracotomy and sternotomy approaches, minimally invasive approaches (VATS) have started to be used today. (5) The surgical method is to remove the entire cyst by preserving the lung tissue as much as possible if the cyst is intact and to prevent spread to the surrounding area, and to combat complications in ruptured cysts. (5) Surgical methods include the removal of the membranes with enucleation or cystotomy

and closure of the cavity in the lung tissue (capitonnage) or lung resection in rare cases. (4) Although lung resection is considered in cases such as treatment-resistant abscesses, large-multiple cysts, bronchiectasis and haemorrhage (6), lung tissue should be preserved as much as possible and large lung resections such as lobectomy should be considered as a last resort. (5) In cases where both lungs are infected, the procedure can be performed in different sessions, sometimes sternotomy can be performed if there are lesions inaccessible areas in both lungs. (5) In complicated cases such as bronchobiliary or pleuropulmonary fistula, methods such as stent, drainage, fistula tract resection can be used. (6) The success rate of surgical treatment is high, although a recurrence rate of 0.6-5% has been reported, postoperative medical treatment is thought to reduce the recurrence rate. (4) In addition, mortality and morbidity of surgical treatment are low. (5,12) The morbidity rate is expected to be higher in ruptured cysts. (10) Post-operative prolonged air leakage, atelectasis, pleural infection, pneumonia, and recurrence are the most common post operative complications (5,10,12). Complications usually respond to conservative approachs (12)

5. Conclusion

Hydatid cyst is a parasitic disease caused by Echinococcus larvae. It is diagnosed mostly by radiological methods, and it has medical and surgical treatment options.

References

1. Sarkar M, Pathania R, Jhobta A, Thakur BR, Chopra R. Cystic pulmonary hydatidosis. Lung India. 2016;33(2):179-191. doi: 10.4103/0970-2113.177449.

2. Lupia T, Corcione S, Guerrera F, Costardi L, Ruffini E, et al. Pulmonary Echinococcosis or Lung Hydatidosis: A Narrative Review. Surgical Infections. 2021;22(5):485-495. doi: 10.1089/sur.2020.197

3. Craig PS, McManus DP, Lightowlers MW, Chabalgoity JA, Garcia HH, et al. Prevention and control of cystic echinococcosis. Lancet Infect Dis. 2007;7: 385–394

4. Demirel A, Cangır AK, Gebitekin C, Gülhan E, Işıtmangil T, et al. (2013) Akciğer Hidatik Kisti. (1st edit). Ankara: Sentez Yayıncılık

5. Yalçınkaya İ (ed). (2016). Hydatid Cyst Of The Lung. (1st edit). Istanbul: Tüsad

6. Esme H, Şahin DA. Treatment Of Pulmonary Hydatıd Cyst: Revıew. Turkiye Klinikleri J Med Sci 2007; 27: 870-875.

7. Santivanez S, Garcia HH. Pulmonary cystic echinococcosis. Curr Opin Pulm Med. 2010;16(3):257-61. doi: 10.1097/MCP.0b013e3283386282.

8. Gottstein B, Reichen J. Hydatid lung disease (echinococcosis/hydatidosis). Clin Chest Med. 2002;23(2):397-408, ix. doi: 10.1016/s0272-5231(02)00007-2.

9. Rawat S, Kumar R, Raja J, Singh RS, Thingnam SKS. Pulmonary hydatid cyst: Review of literature. J Family Med Prim Care. 2019;8(9):2774-2778. doi: 10.4103/jfmpc.jfmpc_624_19.

10. Sivrikoz MC, Boztepe H, Döner E, Durceylan E, Aksu E, et al. Hydatid Cyst of Lung and Surgical Therapy. Solunum 2011; 13(3): 166–169.

11. Işıtmangil T, Görür R, Yiyit N, Erdik O, Yıldızhan A, et al. Evaluation of 308 patients surgically treated for thoracic hydatidosis. Türk Göğüs Kalp Damar Cer Derg 2010;18(1):27-33.

12. Halezeroğlu S, Çelik M, Uysal A, Ürek Ş, Şenol C, et al. Akciğerin Dev Hidatik Kistleri. GKD Cer. Derg. 1995; 3:75-77.

13. Ghoshal AG, Sarkar S, Saha K, Sarkar U, Kundu S, Chatterjee S, Kundu S. Hydatid lung disease: an analysis of five years cumulative data from Kolkata. J Assoc Physicians India. 2012; 60:12-6.

14. Greenber DJ, Canoy J, Pavosevic HT, Epelbaum O. Pulmonary Cystic Echinococcosis. Mayo Clin Proc. 2022;97(4):752-753. doi: 10.1016/j.mayocp.2022.01.034

CHAPTER VI

EMERGENCIES AND DIFFICULTY OF SURGERY IN MENTALLY RETARDED PATIENTS

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1. Introduction

There is a section of society that is dependent on us, cannot easily continue its life without support and care, is pure, clean and does not know evil. Doctors use the term Mental Retarded to identify these people with special conditions. Of course, the scientific terms we use among us are important, but these types of patients are no ordinary case. Although we consider the child not as a small adult but as a separate individual, it is our duty to create special categories for these people. They need to be looked after in a special conditions, approached in a gentle way.

In fact, we think that it is necessary to be better conscious and get special training if necessary in order to establish a dialogue with such patients when these applied to the hospital but the care and surgery of these patients should not frighten us. It may be enough just to get good and comprehensive information about them.

In this study, we wanted to discuss the emergency problems and surgery of mentally retarded patients using both terms "*Intellectual Disability*" and "*Mental Retardation*".

2. Definition of Mental Retardation

The term "*Mental Retardation*" is no longer used in the field of psychology and psychiatry. Instead, the current term used to describe this condition is

"*Intellectual Disability*" or "*Intellectual Developmental Disorder*". Intellectual Disability is a neurodevelopmental disorder characterized by significant limitations in intellectual functioning and adaptive behavior, which includes conceptual, social, and practical skills. These limitations arise during the developmental period and are assessed using standardized tests of intelligence quotient (IQ) and adaptive behavior (1).

According to the The American Psychiatric Association, an individual is diagnosed with intellectual disability if they have:

2.1 Deficits in intellectual functioning, typically assessed by IQ scores that are two or more standard deviations below the mean (approximately an IQ score of 70 or below).

2.2 Significant limitations in adaptive functioning in at least two areas of life skills, such as communication, self-care, home living, social-interpersonal skills, use of community resources, self-direction, functional academic skills, work, leisure, health, and safety.

2.3 Onset of the intellectual and adaptive deficits during the developmental period (before age 18).

It is important to note that a diagnosis of intellectual disability is not based on a specific IQ score alone but also considers an individual's functional abilities and the context in which they live. People with intellectual disability can have a wide range of abilities and challenges, and it is important to provide individualized support and accommodations to help them achieve their full potential (2).

It is difficult to provide an exact rate of mental retardation worldwide, as the prevalence of intellectual disability varies by country and can be affected by a range of factors, including access to healthcare, genetics, and environmental factors. However, the World Health Organization estimates that approximately 1-3% of the global population has an intellectual disability, which translates to between 70 and 200 million people worldwide (3).

3. Approach to the Patient in Mental Retardation

Managing patients with intellectual disability who require surgery in the emergency department can be challenging. Individuals with intellectual disability may have difficulty understanding the need for the procedure, may struggle with communication, and may have difficulty following instructions or complying with medical treatment (4). To ensure the safety and well-being of patients with intellectual disability, it is important to involve their parents, caregivers or family members in the decision-making process and to provide

them with clear and concise information about the surgery and the expected outcomes. It may also be necessary to have a trained interpreter, conscious doctor and experienced nurse maybe with sign language interpreter present to facilitate communication with the patient (5).

In addition, healthcare providers and team should take extra care to ensure the patient is comfortable and calm before and after the surgery. This may involve using specially equipped rooms, calming techniques such as deep breathing exercises, providing sensory distractions such as music or videos, or using sedation if necessary (6). The key to managing patients with intellectual disability who require surgery in the emergency department is to provide individualized care that takes into account their unique needs and abilities. This may require additional time, experience and resources, but it is essential for ensuring the best possible outcome for the patient (7).

4. Diagnosis Difficulty

Patients with intellectual disability may have difficulty communicating their symptoms and medical history, which can make it difficult to obtain accurate information about their condition. May have co-occurring medical conditions or psychiatric disorders that can complicate the diagnosis and treatment of their emergency medical condition. Patients with intellectual disability may have a higher risk of experiencing medical emergencies due to their increased vulnerability to physical illness and injury, and may require more intensive medical interventions(8,9).

Emergency department staff may have limited experience or training in caring for patients with intellectual disability, which can make it difficult to recognize and address their unique needs(10).

To address these challenges, it is important for emergency department staff to have a thorough understanding of the patient's medical history, cognitive and functional abilities, and communication style. Doctors and healthcare providers should also work closely with the patient's caregiver or family members to obtain information about their medical history and any pre-existing medical conditions (11,12).

In addition, doctors may need to adjust their diagnostic and treatment approaches to accommodate the patient's cognitive and functional abilities. This may involve using alternative methods of communication, such as pictures or gestures, and adapting medical procedures to minimize discomfort and distress (13).

Overall, the diagnosis and treatment of medical emergencies in patients with intellectual disability requires a multidisciplinary approach to their care. By working collaboratively and adopting a patient-centered approach, healthcare doctors and providers can work effective and compassionate care to patients with intellectual disability in the emergency department.

5. Examining the Patients with Mental Retardation Coming to the Emergency

Patients with intellectual disability may not be comfortable when they come to the hospital. There may even be panic and fear. These feelings may be excessive or even exaggerated in the emergency room. Doctors and nurses should be patient and kind in this situatio. Examining an injured patient with intellectual disability may require some additional considerations compared to examining a patient without intellectual disability. We can suggest some advice in this regard:

5.1 Creating a Comfortable Environment

It is essential to create a comfortable and non-threatening environment for the patient. Make sure the patient is in a quiet room with no distractions, and that they feel safe and secure. The patient can be taken to another place in or next to the emergency room.

5.2 Communicating Effectively

Do not complicate the dialogue, keep simple sentences and clear language to explain what you are doing and what you need the patient to do. If necessary, use pictures, coloers, gestures, or other visual aids to help the patient understand.

5.3 Taking Time to Build Rapport Unless İt›s an Over Emergency

Building a good rapport with the patient is important. Take time to establish a relationship with the patient before starting the exam. Talk to the patient in a calm and reassuring manner, and avoid rushing the exam. İf not, use sedatives or support from anesthesia.

5.4 Involving Family, Caregiver and Relatives

If possible, involve the parents, caregiver or other family members in the exam process. They can provide important information about the patient's medical history, behavior, and communication style.

5.5 Modifing the Exam as Needed

Inspection is not fixed, target is more important. Depending on the patient's abilities and injuries, you may need to modify the exam. For example, you may need to use nonverbal cues to elicit responses from the patient, or you may need to perform the exam in a different position to accommodate the patient's physical limitations.

5.6. Using Behavioral and Sensory Strategies

Fear can prolong your work. If the patient becomes agitated or uncooperative during the exam, use behavioral and sensory strategies to calm them down. Don't be bothered by being funny or acting childish. You may include playing soothing music, animated emojis or other sensory input, or use distraction techniques.

5.7. Considering Pain Management

People with intellectual disability may have difficulty communicating their pain, so it is important to assess and manage their pain carefully. Use a pain scale that is appropriate for the patient's level of intellectual functioning, and consider using pain medications as needed.

Evaluating an injured patients with intellectual disability requires patience, sensitivity and flexibility. By following these steps, we can ensure a comprehensive examination while ensuring the comfort and safety of the patient (11-14).

6. Additional and Special Examinations to be Made to the Patient in Mental Retardtion

It is difficult to create a comprehensive examination of a patient with intellectual disability. It is important to take into account their unique needs and abilities. In addition to a standard physical exam, other things may be considered. There are no fixed rules on this subject in the literature. The doctor should be thoughtful and creative according to the need. We can list a few tips(15,16).

6.1. Cognitive Assessment

Assess level of consciousness, alertness. A cognitive assessment can help determine the patient's level of intellectual functioning and identify any cognitive deficits or impairments.

6.2. Behavioral Assessment

Be cautious, talk to family. A behavioral assessment can help identify any behavioral difficulties or symptoms the patient may be experiencing, such as aggression, self-harming or repetitive behaviors. A behavioral assessment can help identify any behavioral challenges or symptoms that the patient may be experiencing, such as aggression, self-injury, or repetitive behaviors.

6.3. Sensory Evaluation

Sensory evaluation before starting the exam can help identify sensory processing difficulties that the patient may experience, such as hypersensitivity to touch, sound, or light.

6.4. Communication Assessment

Can the patient speak, is there a common language? Can he explain his problem? Communication assessment can help identify any speech or language impairments the patient may have and determine the best way to communicate with the patient.

6.5. Adaptive Behavior Assessment

Does the patient have withdrawal or social problems or feeling of loneliness? An adaptive behavior assessment can help determine the patient's level of independence and identify any deficits or impairments in areas such as daily living skills, socialization, and leisure activities.

6.6. Psychiatric Evaluation

Are there additional problems such as mania, depression, or schizophrenia? Patients with intellectual disability may be at increased risk for psychiatric disorders such as anxiety, depression, or behavioral problems. A psychiatric evaluation can help identify any mental health concerns and develop an appropriate treatment plan.

6.7. Neurological Evaluation

A neurological evaluation may be necessary to identify any underlying neurological conditions that may be contributing to the patient's symptoms, such as seizures or movement disorders.

It is important to remember that these additional and special examinations may require specialized training and expertise. Healthcare providers should

work closely with a team of professionals, including psychologists, psychiatrists, and other specialists, to ensure that the patient receives comprehensive and individualized care(1,2,11).

7. The diagnosis of Mental Retarder Patient in an Emergency That Requires Surgery

In an emergency situation where a patient with intellectual disability requires surgery, it is important to prioritize the patient's safety and well-being while also ensuring that they receive the necessary medical care. Managing this situation is to obtain a thorough medical history and conduct a physical examination to assess the patient's overall health and any pre-existing medical conditions that may impact the surgical procedure(17).

If the patient is unable to provide information about their medical history in injury and acute situations, most of the work falls to the surgical, emergency team and other healthcare providers. A rapid assessment of the situation and consult with family members or caregivers to obtain this information may be enough(18).

Along with rapid imaging such as FAST USG, interventions such as vascular access can be performed with experienced nurses. The healthcare team should work collaboratively to develop a plan for the surgical procedure that takes into account the patient's unique needs and challenges. This may involve adapting the surgical approach to accommodate the patient's cognitive and functional abilities, ensuring appropriate pain management, and providing additional supportive care throughout the procedure(19).

If they have time to do, is also important to ensure that the patient receives appropriate pre-operative counseling and education about the surgical procedure and peri-operative care, using alternative methods of communication if necessary. Additionally, the healthcare team should work closely with the patient's family members to provide ongoing support and ensure that the patient's pre-operative care needs are met (20).

It's important to remember that in serious illnesses and injuries, these patients may not be aware of the severity of the condition or may not show the usual response. It may not fully reflect pain reactions. The severity of the situation is not visual, but can be revealed with comprehensive assessment.

Making the decision to operate on a patient with intellectual disability requires careful consideration of several factors, including the patient's medical

condition, the potential risks and benefits of the surgical procedure, the patient's cognitive and functional abilities, and their overall quality of life (21).

The healthcare team should carefully evaluate the potential risks and benefits of the surgical procedure for the patient, taking into account their overall health status, the potential impact of anesthesia and surgery on their cognitive and functional abilities, and the potential impact of the surgery on their quality of life. The team should also consider alternative treatment options and the potential consequences of not performing the surgery (22).

8. Surgery of Mental Retarder Patients

It is possible that a surgeon may feel apprehensive or uncertain about operating on a patient with intellectual disability, particularly if they are not experienced in working with this patient population. If a surgeon feels uncertain or uncomfortable about operating on a patient with intellectual disability, they may benefit from additional training or consultation with colleagues who have experience in working with this patient population. However, it is important to note that the decision to operate on a patient with a intellectual disability should be based on the patient's medical condition, the potential risks and benefits of the surgical procedure, rather than the surgeon's personal feelings or biases (23).

Surgeons are trained to provide care to all patients, regardless of their disability status, and should work collaboratively with the patient's healthcare team to develop a plan for the surgical procedure that takes into account the patient's unique needs and challenges. This may involve adapting the surgical approach to accommodate the patient's cognitive and functional abilities, ensuring appropriate pain management, and providing additional supportive care throughout the procedure (23,24).

It is also important for doctors and healthcare providers to recognize that disabled patients have the same right to receive quality medical care as non-disabled patients. Surgeons should strive to provide compassionate and patient-centered care to all patients, including those with disabilities, and work collaboratively with the patient's healthcare team to ensure that their medical needs are met (25). Although immadiate problems and related complications are common in the patients with intellectual disability, they are usually recognized late, may people with mental retardation don´t receive the care or attention they need (26).

9. Surgical Consent

The first step in obtaining consent from a patient with intellectual disability is to ensure that the patient understands the nature of the surgical procedure, the risks and benefits involved, and the expected results. If the patient has a cognitive impairment that affects their ability to understand this information, surgical team and healthcare providers may use alternative communication methods, such as various objects or simplified body language, to help the patient understand (1,2,11,27).

It is also important to involve the patient's guardian, family members in the consent process, as they can provide additional information and support to the patient. The healthcare team may seek approval from the patient's guardian or family members to ensure that the patient's wishes and preferences are taken into account (11,28).

In some cases, it may be necessary to obtain legal consent from a legal guardian or surrogate decision-maker, particularly if the patient is unable to provide informed consent due to their cognitive impairment. In these cases, healthcare providers should work closely with the patient's caregiver or family members to ensure that the legal requirements for obtaining consent are met (27-29).

Except in very urgent situations, decisions should be made by observing legal responsibility and justice. We need to evaluate it not as a baby in need of care, but as an individual who has communication difficulties.

10. Preparing the Patient in Mental Retarder for Surgery

When we reach this stage, we need to keep the patient calm and relaxed as much as possible. Surgery on a patient with intellectual disability may present some unique challenges and difficulties, but with careful planning, communication, and support, the healthcare team can ensure that the patient receives the best possible care and achieves a successful outcome.

10.1. Address the Patient's Fears and Concerns

Patients with intellectual disability may have fears and concerns about the surgical process, such as fear of pain or being separated from their families and caregivers. Healthcare team should address these concerns and provide reassurance and emotional support as needed. While taking off the patient's

clothes or putting on the operating room clothes, people he can trust should be with him during the transport process (30).

10.2. Provide Appropriate Anesthesia and Pain Management

Patients with intellectual disability may have difficulty communicating their pain or discomfort, so it is important to provide appropriate anesthesia and pain management to ensure their comfort and safety during and after the surgical procedure (9,31).

10.3. Difficulty with Anesthesia

Patients with intellectual disability may have difficulty understanding the need for anesthesia, may be unable to follow instructions regarding breathing, or may have difficulty communicating any discomfort or pain they are experiencing. This may require careful monitoring and adjustment of the anesthesia during surgery (31).

10.4. Increased Risk of Medical Complications

Patients with intellectual disability may be at increased risk of certain medical complications during surgery, such as respiratory issues or infections. The healthcare team should closely monitor the patient's vital signs and adjust treatment as needed (32).

10.5. Longer Recovery Time

Patients with intellectual disability may require a longer recovery time after surgery due to a variety of factors, including communication challenges, behavior challenges, and increased risk of medical complications. The healthcare team should provide appropriate support and resources to ensure that the patient has a successful recovery (33).

We generally consider such applications not as patients, but as techniques and cases. However, although the technique is important in such cases, the kind of patient we deal aned treat may be more effective and important.

11. After the Surgery

Patients with intellectual disability may have difficulty communicating their needs, discomfort or pain after the surgery. We need to be prepared for such needs in advance. May be at increased risk of certain medical complications after

surgery, such as infections, respiratory issues, bleeding or blood clots. Our pre-assessment of risks increases the effectiveness of treatment. The healthcare team should closely monitor the patient's vital signs and adjust treatment as needed to minimize these risks (22,32,34). Patients with intellectual disability may exhibit challenging behaviors after surgery, such as fear, agitation, confusion, or restlessness may require a longer hospitalization and recovery time after surgery due to a variety of factors, including communication challenges, behavior challenges, and increased risk of surgical or medical complications (33,34).

Monitoring, nasogastric or foley catheter, even dressing material can cause trauma to them. They may have difficulty understanding or following postoperative instructions, such as medication regimens or activity restrictions. Health team and family members may need to provide additional support in ensuring that the patient follows these instructions. Restricting the patient's movement should be our last option.

12. Conclusion

People with intellectual disability are part of society. Although the surgical procedure to be applied to them is difficult for the surgeon, it is perhaps even more difficult for them. Let's not see them as a forced heavy difficulty. It is not enough for them to be taken care of by the social organs of the state. We expect them to be followed by experienced people who have branched out over time and are specially trained. We wanted to shed some lights on a few points that will help in this regard without discussing the maintenance deficiencies.

References

1. Lee K, Cascella M, Marwaha R. Intellectual Disability. 21. In:StatPearls, 2022. Sep [Internet]. Treasure Island (FL): StatPearls Publishing. 2023 Jan. PMID: 31613434.

2. American Psychiatric Association. Diagnostic and statistical manual of mental disorders (DSM-5 (R))(5th ed.). Washington, DC: American Psychiatric Association Publishing. 2013

3. Jansen S, Vissers LELM, de Vries BBA. The Genetics of Intellectual Disability. Brain Sciences. 2023;13(2):231. Doi: 10.3390/brainsci13020231

4. Hardavella G, Aamli-Gaagnat A, Frille A, Saad N, Niculescu A, Powell P. Top tips to deal with challenging situations: doctor–patient interactions. Breathe. 2017;13(2):129-35. Doi:10.1183/20734735.006616

5. Molina-Mula J, Gallo-Estrada J. Impact of Nurse-Patient Relationship on Quality of Care and Patient Autonomy in Decision-Making. IJERPH. 2020;17(3):835. Doi:10.3390/ijerph17030835

6. Appukuttan D. Strategies to manage patients with dental anxiety and dental phobia: literature review. CCIDE. 2016;35-50. Doi:10.2147/ccide.s63626

7. Ervin DA, Hennen B, Merrick J, Morad M. Healthcare for Persons with Intellectual and Developmental Disability in the Community. Front Public Health. 2014;2:1455. Doi:10.3389/fpubh.2014.00083

8. Edgerton RB, Gaston MA, Kelly H, Ward TW. Health Care for Aging People With Mental Retardation. Mental retardation, 1994:32(2), 146-150.

9. Prater CD, Zylstra RG. Medical care of adults with mental retardation. Am Fam Physician. 2006;73(12):2175-2183.

10. Stefan S. Emergency department treatment of the psychiatric patient: Policy issues and legal requirements. Oxford University Press. 2006

11. Kishore MT, Udipi GA, Seshadri SP. Clinical Practice Guidelines for Assessment and Management of intellectual disability. Indian J Psychiatry. 2019;61(Suppl 2):194-210. Doi: 10.4103/psychiatry.IndianJPsychiatry_507_18

12. Janowsky DS, Davis JM. Diagnosis and treatment of depression in patients with mental retardation. Curr Psychiatry Rep. 2005;7(6):421-428. Doi:10.1007/s11920-005-0062-z

13. Happ MB, Garrett K, Thomas DD, et al. Nurse-patient communication interactions in the intensive care unit. Am J Crit Care. 2011;20(2):e28-e40. Doi:10.4037/ajcc2011433

14. Slayter EM, Garnick DW, Kubisiak JM, Bishop CE, Gilden DM, Hakim RB. Injury prevalence among children and adolescents with mental retardation. Ment Retard. 2006;44(3):212-223. Doi:10.1352/0047-6765(2006)44[212:IPACAA]2.0.CO;2

15. Gordenchuk VD. The organization of work therapy in the comprehensive treatment of severely mentally retarded patients. Soviet Neurology & Psychiatry. 1987

16. Shelton JT. Treatment of the Mentally Retarded. The role of the local physician. Calif med. 1964;101(1):19-22.

17. World Health Organization. Principles for the protection of persons with mental illness and the improvement of mental health care. Geneva: WHO. 17 December 1991. https://www.ohchr.org/en/instruments-mechanisms/instruments/principles-protection-persons-mental-illness-and-improvement

18. Rodziewicz TL, Houseman B, Hipskind JE. Medical Error Reduction and Prevention. In: StatPearls. Treasure Island (FL): StatPearls Publishing; December 4, 2022.

19. Rabinovici R, Frankel H, Kaplan L. Trauma evaluation and resuscitation. Curr Probl Surg. 2003;40(10):599-681. Doi:10.1016/S0011-3840(03)00081-9v

20. Swindale JE. The nurse's role in giving pre-operative information to reduce anxiety in patients admitted to hospital for elective minor surgery. J Adv Nurs. 1989;14(11):899-905. doi:10.1111/j.1365-2648.1989.tb01478.x

21. Daily DK, Ardinger HH, Holmes GE. Identification and evaluation of mental retardation [published correction appears in Am Fam Physician 2000 Sep 1;62(5):961-3]. Am Fam Physician. 2000;61(4):1059-1070.

22. Goldhaber SZ, Reardon FE, Goulart DT, Rubin IL. Cardiac surgery for adults with mental retardation. Dilemmas in management. Am J Med. 1985;79(4):403-406. Doi:10.1016/0002-9343(85)90025-7

23. Stiles CM. Anesthesia for the mentally retarded patient. Orthop Clin North Am. 1981;12(1):45-56.

24. Paransky OI, Zurawin RK. Management of menstrual problems and contraception in adolescents with mental retardation: a medical, legal, and ethical review with new suggested guidelines. J Pediatr Adolesc Gynecol. 2003;16(4):223-235. Doi:10.1016/s1083-3188(03)00125-6

25. Morley L, Cashell A. Collaboration in Health Care. J Med Imaging Radiat Sci. 2017;48(2):207-216. Doi:10.1016/j.jmir.2017.02.071

26. Taha A. Swallowed nail perforating the anterior surface of the stomach and penetrating the abdominal wall from the inside: a case report. PAMJ-CM. 2022;9(11). Doi:10.11604/pamj-cm.2022.9.11.35073

27. Stineman MG, Musick DW. Protection of human subjects with disability: guidelines for research. Arch Phys Med Rehabil. 2001;82(12 Suppl 2):S9-S14.

28. Satyanarayana Rao KH. Informed consent: an ethical obligation or legal compulsion?. J Cutan Aesthet Surg. 2008;1(1):33-35. Doi:10.4103/0974-2077.41159

29. Raina P, O'Donnell M, Rosenbaum P, et al. The health and well-being of caregivers of children with cerebral palsy. Pediatrics. 2005;115(6):e626-e636. Doi:10.1542/peds.2004-1689

30. Katz P. Ritual in the Operating Room. Ethnology. 1981;20(4):335. Doi:10.2307/3773355

31. Chaudhary K, Bagharwal P, Wadhawan S. Anesthesia for intellectually disabled. J Anaesthesiol Clin Pharmacol. 2017;33(4):432-440. doi:10.4103/joacp.JOACP_357_15

32. Bates WJ. Multimodal treatment of mental illness in institutionalized mentally retarded persons. Handbook of mental illness in the mentally retarded, 219-230. Springer 1984

33. Garrard SD. Health services for mentally retarded people in community residences: problems and questions. Am J Public Health. 1982;72(11):1226-1228. Doi:10.2105/ajph.72.11.1226

34. Krahn GL, Hammond L, Turner A. A cascade of disparities: health and health care access for people with intellectual disabilities. Ment Retard Dev Disabil Res Rev. 2006;12(1):70-82. Doi:10.1002/mrdd.20098

CHAPTER VII

CLINICAL BIOCHEMISTRY LABORATORY EXTERNAL QUALITY CONTROL EVALUATION*

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1. Introduction

Quality control applications are error detection procedures that evaluate systematic problems, environmental conditions and personnel performance in addition to precision and accuracy studies in analytical applications and determine how reliable the service is and whether it has predefined features (1).

Quality control practices are an indispensable part of total quality management system control processes. Basically, there are two types of quality

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control practices: internal quality control (IQC) and external quality control (EQC). IQC is the analysis of control samples containing analytes of known concentration to monitor the precision and accuracy of the analytical process over time. EQC involves the analysis and reporting of control samples provided by an external agency at a predefined timeframe, which is monthly in clinical biochemistry laboratories. There is a wide variety of procedures for EQC that differ by program provider and country. Laboratory results directly affect the patient's life. Inaccurate and incomplete results given from the laboratory to the clinician cause prolonged hospital stay of the patients, loss of labor and time in the laboratory, and an increase in laboratory expenses. Countries need to establish their own EQA programs and evaluation systems to avoid this confusion (1).

There are a limited number of national EQC programs implemented by clinical laboratories in the Turkish Health System. The Association of Clinical Biochemistry Specialists External Quality Control Program is one of them, and it monitors and ensures the quality of analytical methods by providing outsourcing services over 114 test parameters with 10 accredited EQC programs (2).

2. Overview of Quality Management and Quality Control in Clinical Biochemistry Laboratories

2.1. Quality Management in Clinical Biochemistry Laboratories

Total quality management (TQM) or quality management system (QMS) principles have become the basis for the management and operation of good clinical laboratories (3). Figure 1. shows the following five components of the QMS:

1. Quality planning begins with conceptualizing and scoping a test for a particular need, only by laying out the measures to ensure that the test meets the user's expectations.

2. The quality process includes analytical processes such as validation or validation of analysis, and general policies, guidelines and procedures that define how work is to be done. This means that test validity is only one component of many components in TQM, and without a fully integrated QMS, the validity of a test cannot guarantee acceptable performance in practice.

3. Quality control (QC) includes different actions and practices to ensure and/or monitor quality, as will be detailed later.

4. Quality assessment (QA), documentation of a facility's organizational structure, personnel qualifications and competence, enrollment testing menu and utility, patient and specimen identification tools, defining turnaround time to meet customer expectations, monitoring laboratory error rates, evaluating customer satisfaction, and It relates to broader measures than QC and tools for monitoring laboratory performance, such as keeping records of documents. From this perspective, quality assessment is the appropriate term, as it has traditionally been called quality assurance, since retrospective measurement of laboratory performance by itself does not detect problems in a timely manner or improve quality (3). From this perspective, quality assessment is the appropriate term, as it has traditionally been called quality assurance, since retrospective measurement of laboratory performance by itself does not detect problems in a timely manner or improve quality (3).

5. Quality improvement (QI) refers to the process of problem solving by identifying the root cause of a problem, finding a remedy, and providing a satisfactory outcome. The new quality planning cycle begins with the proactive implementation of the solution into daily practice to prevent similar errors from occurring in the future. Quality improvement and quality planning for a particular issue can be defined as corrective action-preventive action.

Figure 1. Total quality management (TQM) or quality management system (QMS) principles (4)

1.2. Quality Control in Clinical Biochemistry Laboratories

Total laboratory automation has replaced manual testing of parameters, but laboratory errors are still increasing. The only way to control laboratory errors is to perform quality control at all stages of testing. Quality control in the medical laboratory is a statistical process used to monitor and evaluate the analytical process that produces patient results and the quality of all tests performed in the medical laboratory creates conditions that help clinicians practice good medicine. When a diagnostic test is performed in a medical laboratory, the result of the test is a result. The result can be a patient result or a quality control (QC) result. The result can be quantitative (number) or qualitative (positive or negative) or semi-quantitative (limited to a few different values). The QC results are used to confirm whether the device is working within predefined specifications and the patient test results are concluded to be reliable. Once the test system has been validated, patient results can be used for diagnosis, prognosis, or treatment planning. For most tests, reliability issues can be resolved with regular use of quality control materials and statistical process control (5). QMS in medical laboratories is a multi-layered process that covers all steps before analysis, analysis and post-analysis. The principles of QMS in laboratories have been defined by the World Health Organization (WHO) to monitor the quality of the laboratory and to ensure and implement its processes (Figure 2).

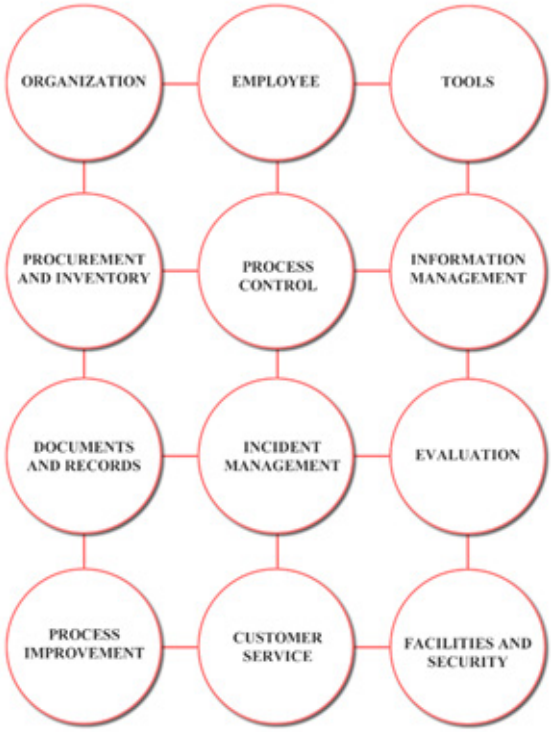

Figure 2. Principles of quality management in laboratories (5)

The QC must go through mainly three parts:

1. Each analytical method should have its own statistical limits of variation.
2. These limits should be used to find the QC data generated for each test type.
3. Elimination of various errors and if found:
   - The cause of the error should be learned.
   - Action should be taken to correct the error.
   - Patient data should be reanalyzed.

The multi-rule procedure includes decision criteria to determine whether an analytical run is under control. It is used to detect random and systemic errors over time and was developed by Westgard and Groth (6).

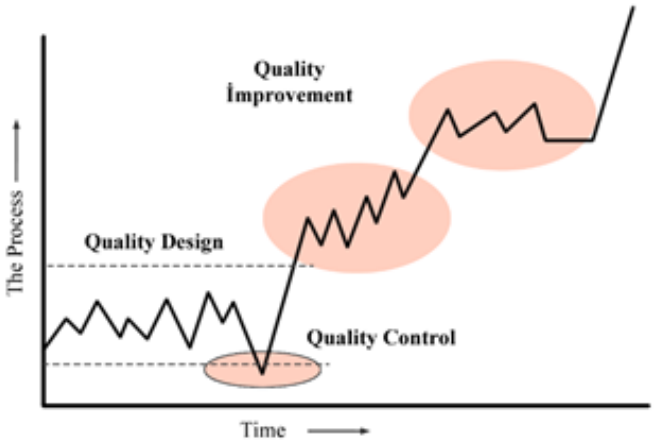

Figure 3. Relationship between various aspects of quality control (6)

Proficiency testing, internal quality control, laboratory reviews, clinical use and quality assurance monitoring play an important role as indicators of analytical performance. Quality management consists of quality design, quality control and quality improvement (6) (Figure 3).

3. Types of Quality Control Programs

QC measurements used to evaluate the analytical stage in a clinical biochemistry laboratory are divided into internal quality control (IQC) and external quality control (EQC). IQC is a sample material whose matrix is the same as the patient sample and has a concentration range available at two or three levels covering medical decision points. It is interpreted using control charts such as the application of Levy Jennings and Westgard rules.

IQC provides continuous monitoring of the analytical system to check whether the results are reliable enough to be disclosed. EQC involves the analysis and reporting of control samples provided by an external agency at a predefined timeframe, which is monthly in clinical biochemistry laboratories. EQC is interpreted with a Z-score or standard deviation index. The Z-score is a calculated value that tells us how many standard deviations a control result deviates from the expected mean value for that material. The exact number of errors made by the laboratory at the analytical stage cannot be evaluated by running internal and external QCs, but can be measured in the laboratory with sigma metrics (7).

In statistics, sigma is used to represent standard deviation, which is an indication of the degree of variation in a set of processes. Sigma measures how much a particular process deviates from perfection. Six Sigma is one of the popular QMS tools used for process improvement (8). The power of Six Sigma is in the measurement of process performance at the "Sigma Scale". The Six Sigma Scale typically runs from 0 to 6, but a process can actually exceed Six Sigma if the variability is low enough to reduce the defect rate. The best or "world-class quality" products have a Six Sigma performance level. Six Sigma methods are generally applied when the outcome of the process can be measured. Poor results are measured in defects per million and expressed on the sigma scale. Sigma metrics are truly the evolution of TQM, with a more quantitative assessment of process performance and clearer goals for process improvement. The exact number of defects or errors made by the laboratory can be measured in the laboratory using sigma metrics (9). The level of sigma metrics and corresponding defects per million tests are shown in Table 1.

Table 1. Level of sigma metrics and corresponding defects per million tests (10)

Six sigma level	Percent accuracy	Defects per million
6	99.9997	3.4
5	99.98	233
4	99.4	6210
3	93.3	66.807
2	69.1	308.537
1	31	698.000

Thus, with the aid of Six Sigma principles and metrics, it is possible to evaluate the quality of laboratory testing processes and the number of QCs necessary to ensure the desired quality is achieved.

3.1. Internal Quality Control

This is based on following the biochemistry testing procedure performed in the laboratory. It includes measurement on specially prepared materials and repeated measurements on routine samples, as well as day-to-day statistical analysis of history from routine testing. Therefore, the reliability of the laboratory's work is constantly evaluated. Therefore, the IQC primarily checks the accuracy of the laboratory work. Each laboratory should establish its own

mean and control limits for daily monitoring of IQC. The mean of each parameter should be calculated considering at least twenty-day values, then the standard deviation and coefficient of variation should be calculated (11).

3.2. External Quality Control

It is a system designed to objectively evaluate the quality of results obtained by laboratories through an external institution. The purpose of the EQC is to provide a measure of individual laboratory quality (12).

In the modern clinical laboratory, quality assurance is demonstrated through the complementary processes of EQA and EQA, also known as proficiency testing. With these processes and accreditation to international (e.g. ISO) standards, the laboratory is able to demonstrate competence to users of its services, wherever they are performed, the scope of service, i.e. clinicians and patients they care for, who have expectations that the results of diagnostic testing and monitoring of treatment are accurate, comparable and fit for purpose.

4. Importance of External Quality Control

Participation in an EQC program provides valuable data and information:

- Allows comparison of performance and results between different test sites.
- Provides early warning of kits or operations.
- Provides objective evidence of test quality.
- Indicates areas that need improvement.
- Determines training needs.

EQC helps customers such as doctors, patients and health authorities ensure that the lab can produce reliable results. Individual laboratories can use EQC to identify problems in laboratory practices and allow appropriate corrective action. EQC involvement will be helpful in assessing the reliability of methods, materials and equipment, and in evaluating and monitoring the impact of training (13).

5. External Quality Control Methods

The term EQC is used to describe a method that allows comparison of a laboratory's tests with a source outside the laboratory. This comparison can be made against the performance of a peer laboratory group or the performance

of a reference laboratory. The term EQC is sometimes used interchangeably with Proficiency testing (PT) ; however, EQC can also be performed using other processes.

Various EQC methods or processes are widely used. These:

1. Proficiency testing - external provider sends unknown samples to a number of laboratories for testing, they are analyzed, compared and reported to the laboratories.

2. Retest and recheck—smears read are rechecked by a reference laboratory; analyzed samples are retested, allowing inter-laboratory comparison.

3. On-site assessment – often done when it is difficult to do traditional PT or use the retest/recheck method.

Another method of interlaboratory comparison is the exchange of samples between a set of laboratories usually reserved for specific tests for which proficiency testing is not available. This method is used by very specialized or complex laboratories (13).

6. Management of External Quality Control in The Clinical Biochemistry Laboratory

1.1. Participation in External Quality Control

All laboratories must participate in EQA programs and include an EQC program for all testing procedures performed. The benefits of this involvement are many, and EQC provides the only way available to ensure that one lab's performance is comparable to that of other labs.

EQC participation is essential for laboratories that are accredited or planning to obtain accreditation. ISO 15189 addresses EQC requirements for laboratories as follows.

- The laboratory is obliged to participate in interlaboratory comparisons.
- Where an established EQC scheme is not in place, an alternative EQC mechanism will need to be considered for interlaboratory comparison, such as sample exchange with other laboratories.
-Laboratory management will monitor the results of the EQC and participate in the implementation of corrective actions (13).

1.2. Management Process

When participating in outsourcing programs, the laboratory needs to develop a process management. The aim is to ensure that all EQC samples are treated in the same way as other samples tested. Procedures should be developed that address the following:

-Processing of samples—They will need to be opened in a timely manner, properly handled, and stored for future use as needed.

-Sample analyzes—Consider whether EQC samples can be tested so that personnel do not perceive them as different from patient samples (blind testing).

-Appropriate record keeping—Records of all EQC test reporting should be retained for a specified period of time so that performance improvement can be measured.

-Investigate for any deficiencies—Any challenge for which performance is not acceptable should be investigated.

-Taking corrective action where performance is not acceptable—The purpose of the EQC is to identify problems in the laboratory and therefore provide an opportunity for improvement.

- To ensure that the results are communicated to the management and all laboratory personnel (13).

7. External Quality Control Program Selection

EQC programs should be selected from national or international programs in accordance with their purpose and legal obligations/requirements and program features should be taken into account. In addition, the program provider must have the ISO 17043 accreditation document. The quantities of samples submitted should be adequate and similar to those routinely studied, and the analyte concentration should be compatible with the range of quantitation and the sensitivity of the method. Apart from these, the results should be confidential, there should be sufficient time for disclosure and the reports should be delivered to the user in a short time. Having reliable technical consultants and the number of participating laboratories are also factors to be considered when choosing an EQC program. Frequency of sending samples and sending sufficient number and variety of samples are also important. In addition, there are programs that include the analytical process as well as pre- and post-analytical processes (14).

EQC program selection is very important. Over the years there has been an increase in EQC participation and there are many program providers. In Turkey, in order to be able to enroll in EQC programs, commercial tests are often added to the tender specifications. For this reason, problems such as interruption of the program over time and enrollment in unwanted programs are encountered during tender times. Therefore, it should be ensured that EQC programs are purchased by institution management in line with laboratory recommendations (14).

Sources of error in clinical laboratories mostly occur in the pre- and post-analysis stages. Therefore, outsourcing program applications have mostly been aimed at measuring analytical performance. The aim should be seen as ensuring patient safety and performing the activities at all stages of the analysis process correctly (15).

8. External Quality Control Programs in Turkey

There are several national outsourcing programs in the Turkish health system. These;

- Association of Clinical Biochemistry Specialists External Quality Control Program (KBUDEK) (2),
- Motakk (Quality Control in Molecular Diagnosis) National Program (16),
- Interlaboratory Comparison Program (LABPT) (17),
- National Antimicrobial Resistance Surveillance System External Quality Assessment Studies (UAMDSS) (18).

9. External Quality Control Programs Used in Turkey

Table 2. External Quality Control Programs Used in Turkey (19)

EQC Program	Country
Association of Clinical Biochemistry Specialists EQC Program	KBUDEK, Turkey
LAB PT Quality Control Program	Turkey
RANDOX International Quality Assessment Program	RIQAS, England
BIORAD External Quality Assurance Services	EQAS, CA, ABD
Bio-development	BIO DEV, Milan, Italy
Bio Group Medical System	Italy
Medical Laboratory Evaluation American Academy of Family Physicians	MLE, ABD
American Academy of Family Physicians-Qualification Test	AAFP-PT, Kansas, ABD
American College of Pathologists, Proficiency Test	CAP, ABD
Labquality EQAS	Finland
INSTAND	Germany
Bioanalytical Reference Institute	RfB, Germany
AccuTest Proficiency Testing Services	ABD
Digital PT-Oneworld Accuracy	Canada
UK National External Quality Assessment Service	UK-NEQAS, UK
Galler External Quality Assurance Program	WEQAS, England
European Association for External Quality Evaluation	ESfEQA, Germany
NOBIS Quality System	Romania

In a study carried out between 2010 and 2017 within the scope of the Ministry of Health's EQC program in Turkey, the accuracy of 24 different clinical biochemistry tests was evaluated in 1941 laboratories. The EQC programs to which the participating laboratories are members are indicated. Table 2 includes 18 different EQC programs, although there are only two EQC programs in Turkey (19).

10. Association of Clinical Biochemistry Specialists External Quality Control Program (KBUDEK)

KBUDEK is designed to compare the performance of participating laboratories with other laboratories in terms of devices, tests and methods. It was

established by the Association of Clinical Biochemistry Specialists (KBUD) in 2005 and was put into service in 2006 with the participation of 75 biochemistry laboratories. It gained an international qualification in 2014. In 2017, 10 laboratories 114 test panels were accredited by the Turkish Accreditation Agency (TÜRKAK) within the scope of TS EN ISO/IEC 17043 (2) (Table 3).

Table 3. KBUDEK EQC Programs (2)

Clinical Biochemistry	**Samples**	**Way of Working**
Alanine Aminotransferase (ALT), Albumin, Alkaline Phosphatase (ALP), Amylase, Aspartate Aminotransferase (AST), Iron (Fe), Iron Binding Capacity (TIBC), Direct Bilirubin, Phosphate (Inorganic) (P), Gama-Glutamil Transferase (GGT), Glucose, HDL Cholesterol, Calcium (Ca), Chloride (Cl), Creatinine, Creatine Kinase (CK), Lactate Dehydrogenase (LDH), Lactate, Lipase, Magnesium (Mg), Potassium (K), Sodium (Na), Total Bilirubin, Total Cholesterol, Total Protein, Triglyceride, Urea, Uric acid, LDL-Cholesterol	12 lyophilized serums	1-year program with 12 samples in one distribution. 1 sample is studied each month.

In the KBUDEK final reports,

- The z-score value, which enables the evaluation of participant performance,
- The total number of participants evaluated and rejected,
- Statistical evaluation of results,
- Graphs and tables where the results are evaluated,
- Information on the device and method used for the test,
- Device performances,
- Comments and recommendations are included (2).

11. Results

EQC is a system for objectively checking the performance of the laboratory using an external institution or facility. EQA is one of the important elements of a laboratory quality management system. For all tests performed whenever possible, laboratories, especially accredited laboratories, should participate in an EQC program.

There are a number of methods for conducting EQC. Traditional PT can be used for many tests, provides useful information and is also cost-effective. When PT does not provide sufficient information, other methods should be used.

Because EQA uses valuable resources, the laboratory should make the most efficient use of its participation in EQA. EQC should not be punitive. It should be seen as educational and used as a tool to directly assist improvement efforts. The various regulations regarding EQC vary by program provider and country. Inaccurate and incomplete results given from the laboratory to the clinician cause prolonged hospital stay of the patients, loss of labor and time in the laboratory, and an increase in laboratory expenses. To avoid this confusion, it is necessary for each country to establish their own EQC programmes.

References

1. Rehnuma B, Ibrahim M, Al Nasir T. Quality Assurance and Quality Control in Clinical Laboratories. Pulse. 2015;8:62-65.

2. https://www.kbudek.com/Content/Documents/Program_Protokolu.PDF. Erişim tarihi 25 Haziran, 2022.

3. Klee GG, Westgard JO. Quality management. In: Burtis CA, Ashwood ER, Bruns DE, editors. Tietz textbook of clinical chemistry and molecular diagnostics. 5th ed. St Louis, MO: Elsevier. 2012;2131-75.

4. Westgard JO, Burnett RW, Bowers GN. Quality management science in clinical chemistry: a dynamic framework for continuous improvement of quality. Clin Chem. 1990;36(10):1712-16.

5. https://www.linkedin.com/pulse/basic-lessons-laboratory-quality-control-asanga-wickremanayaka. Erişim tarihi 14 Mayıs, 2022.

6. Bishop ML, Fody EP, Schoeff LE. Clinical Chemistry: Principles, Techniques, and Correlations. 7th ed. Lippincott Williams and Wilkins. 2013. ISBN-10: 1451118694, ISBN-13: 978-1451118698.

7. Westgard JO. Internal quality control: Planning and implementation strategies. Ann Clin Biochem 2003;40:593611.

8. Coskun A, Inal T, Unsal I, Serteser M. Six Sigma as a Quality Management Tool: Evaluation of Performance in Laboratory Medicine Quality Management and Six Sigma. InTechOpen: Rijeka, Croatia; 2010;24861.

9. Nevalainen D, Berte L, Kraft C, Leigh E, Picaso L, Morgan T, et al. Evaluating laboratory performance on quality indicators with the six sigma scale. Arch Pathol Lab Med. 2000;124:5169.

10. Kumar BV, Mohan T. Sigma metrics as a tool for evaluating the performance of internal quality control in a clinical chemistry laboratory. J Lab Physicians 2018;10:194-9.

11. Kanagasabapathy AS, Swaminathan S, and Selvakumar R. Quality control in Clinical Chemistry. Indian J Clin Biochem. 1996;11: 17.

12. LN Sandle. The management of external quality assurance. J Clin Pathol. 2005;58: 141-4.

13. Martinez A. Evaluation of new external quality assessment guidelines involving random blinded rechecking of acid-fast bacilli smears in a pilot project setting in Mexico. International Journal of Tuberculosis and Lung Diseases. 2005;9(3):301–305.

14. Sayıner A. Laboratuvar İç ve Dış Kalite Kontrol Çalışmalarında Gereklilikler. V.Uluslararası Sağlıkta Performans ve Kalite Kongresi (Konuşma Metinleri), Ankara, Şubat 2015.

15. Lippi G, Becan-McBride K, Behúlová D, Bowen RA et al. Preanalytical quality improvement: in quality we trust. Clin Chem Lab Med. 2013;51:229-41.

16. http://motakk.org/assets/img/yardim/MOTAKK_Katilimci_Bilgilendirme.pdf. Erişim tarihi 25 Haziran, 2022.

17. https://www.labpt.com.tr/content/upfiles/LABPT%20KAL%C4%B0TE%20KONTROL%20PROGRAMI%20YILLIK%20PLAN%2022%20(1).pdf. Erişim tarihi 30 Haziran, 2022.

18. https://hsgm.saglik.gov.tr/tr/uamdss Erişim tarihi 30 Haziran, 2022.

19. Mercan F, Serdar MA, Senes M, Konukoglu D. National External Quality Assess-ment follow-up: 2010–2017 Turkish experience. Turk J Biochem. 2019;44(1):1-8.

CHAPTER VIII

VITAMINS B AND CEREBROCORTICAL NECROSIS

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1. Introduction

Vitamins are complex organic compounds essential for metabolism and found in small amounts in foodstuffs (1).

Vitamins may be categorized into two groups: fat-soluble vitamins and water-soluble vitamins. The fat-soluble vitamins are A, D, E, and K, whereas the water-soluble vitamins are B and C (1). These two vitamin groups differ from one another in terms of fundamental biological features as a result of their different water solubilities. Water-soluble vitamins, with excluding vitamin B_{12}, are not stocked in the body and are eliminated in the urine when consumed in significant amounts (2). Fat-soluble vitamins may be stocked in the body, but when taken in excess, they cause hypervitaminosis (3). A process resembling fat absorption allows for the absorption of fat-soluble vitamins along with dietary fat (2). Fat-soluble vitamins are easier to absorb when there are favorable conditions for fat absorption, such as sufficient bile flow and good micelle production. The absorption of water-soluble vitamins is unaffected by changes in fat absorption because they are not linked to lipids. To prevent deficiencies, water-soluble vitamins must be taken in the diet. Vitamins that are water-soluble

are excreted in the urine, whereas vitamins that are fat-soluble pass through the feces. Although excesses of the fat-soluble vitamins A and D may have catastrophic consequences, water-soluble vitamins are generally non-toxic. Unlike water-soluble vitamins, which can also include nitrogen, hydrogen, sulfur, and cobalt, fat-soluble vitamins exclusively contain carbon, hydrogen, and oxygen (2).

B vitamins are crucial for cellular metabolism and function as co-enzymes for a number of enzymes (4-5). In every step of the catabolic process, B vitamins are involved. This process is negatively affected in vitamin B deficiency (6). The citric acid cycle, adenosine triphosphate synthesis in the electron transport chain, and cellular energy generation all depend on the active forms of thiamine, riboflavin, niacin, and pantothenic acid (7-11). For the mitochondrial metabolism of amino acids, fatty acids, and glucose, respectively, thiamine, biotin, and cobalamin are essential. As a result, they provide substrates for the citric acid cycle (12). B vitamins are also act a part in neurochemical synthesis (13-14).

2. Group B Vitamins

Each step of the catabolic process, which produces energy in cells, involves the involvement of B vitamins (6). Any B vitamin deficiency has a negative effect on this process. B vitamins contribute to the electron transport chain and the citric acid cycle, which is the main catabolic activity in the mitochondria (12). In the mitochondria of eukaryotes, the cycle of citric acid is a chain of chemical reactions that generates ATP as energy. Proteins, lipids, and carbohydrates are first metabolized to acetyl-CoA, generally from pyruvate. The energy produced by the citric acid cycle is then transmitted to the chain of electron transport via eight enzymatic activities that produce NADH and $FADH_2$. FAD (B_2), NAD (B_3), component coenzyme A (B_5), or coenzyme Q10 (B_5) are examples of B vitamins function that play a role as co-factors or enzymes in this procedure. Because they serve as substrates for the synthesis of other substances, such as amino acids and fatty acids, many of the intermediate molecules in the cycle must be filled by anaplerotic synthesis, which takes place outside of the cycle. The methionine cycle's production of ketobutyrate from methionine and the direct synthesis of oxaloacetate from pyruvate are the two most prevalent instances (12).

The vitamin-dependent citric acid cycle serves as a mediator for the production of several important chemicals, such as amino acids, fatty acids, and pyrimidines, in addition to producing energy. Some B vitamins are crucial for the one-carbon metabolism, which arranges the methyl groups required by

molecules as well as functional substances including amino acids, purines, and pyrimidines (8-15-16).

The brain accounts for only 2% of body weight versus 20% of the body's total energy expenditure. In addition, the brain is the most metabolically active organ in the body. As a result, it is believed that B vitamins general metabolic activities and their participation in the manufacture of neurochemicals have a special influence on brain function. The fact that each vitamin is actively carried by particular transport pathways across the blood brain barrier and/or choroid plexus highlights the significance of B vitamins for brain function (13-17).

Absorption, post-absorption transport, metabolic activation, catabolism and excretion mechanisms of B vitamins are similar in ruminant and non-ruminant animals. The synthesis of B vitamins, is possible in ruminants. However, in moderate production in the gastrointestinal tract, ruminants cannot meet their B vitamin needs completely from rumen and intestinal synthesis (18). When B vitamin needs are high, the rumen microbial production is reduced, or the breakdown of B vitamins is elevated in mature ruminants, vitamin B shortages can develop. High-performance dairy cows, beef cattle, lambs, and animals ingesting antivitamin-containing plants have all been documented to be deficient. Additionally, deficiencies can arise in animals consuming diets that encourage the growth of bacteria that break down vitamins in the rumen and in animals consuming some food additives and pharmaceuticals like antibiotics or anthelmintics (19-20). The biochemical or clinical signs of vitamin B inadequacy frequently do not appear until the deficit is severe (21).

B group vitamins are necessary in feed for poultry, pigs, and other monogastric animals. In general, it is believed that ruminants may obtain all of the B vitamins they require from diet or through symbiotic microbes, which produce the vitamins spontaneously. Therefore, in ruminants with fully functional rumens, vitamin B insufficiency is not observed. Additionally, when thiamine, niacin, and the essential building ingredient cobalt are low in ruminant animals, vitamin B_{12} cannot be produced in the rumen due to stress and excessive production (1). In vitamin synthesis, the rumen cannot become functional for a while after birth. In the newborn's first several days, young ruminants need sources of B vitamins in their diets, just like non-ruminants. The development of the rumen flora occurs with the contribution of vitamin B from 8 days to 2 months of age (22). These vitamins are created in the proximal end of the gastrointestinal system and are made accessible to the host when they transit through efficient digestion and absorption zones (2).

2.1. Types of Vitamin B

2.1.1. Riboflavin (Vitamin B2)

Flavin, a yellowish pigment linked to the carbohydrate molecule D-ribitol, is the chemical form of riboflavin. Flavin dinucleotide (FAD) and 5'-phosphate are the two active cofactors that riboflavin is present as (5). Riboflavin regulates the pace at which enzymes enter cellular enzymatic processes and is one of the most significant rate-limiting variables in the majority of cellular enzymatic activities. For instance, synthesis, transformation and recycling of niacin, folate and pyridoxine; essential for the synthesis of all haemoproteins (such as cytochrome, hemoglobin, myoglobin, and catalase), including hemoglobin, P450 enzymes, proteins involved in electron transfer, oxygen transport, and storage, and nitric oxide synthases (9). Flavoproteins (proteins containing a nucleic acid derivative of riboflavin) are also helpful variables affecting the use of iron, the control of thyroid hormones, and the metabolism of important fatty acids in brain lipids (9-23-24). As crucial cofactors in the glutathione redox cycle, riboflavin derivatives also boost endogenous antioxidant status and have direct antioxidant effects (25). In riboflavin deficiency, dermatitis appears with keratosis and atrophy. While vascularization occurs in the tongue of rats in riboflavin deficiency, congenital disorders occur in pregnant rats. Paralysis occurs in chickens. In riboflavin deficiency in dogs, symptoms such as growth retardation, alopecia, and inability to stand are seen. In addition, dermatitis, hypoplastic anemia and bradycardia occur. Growth stops in broilers, paralysis and death occur (26).

2.1.2. Niacin (Nicotinamide, Vitamin B3)

Niacin is pyridine-3-carboxylic acid and is the amide form of nicotinic acid. Niacin is produced in tissues from tryptophan (26). Niacin serves as a precursor for ADP-ribose, which has a variety of cellular activities other than those requiring enzymes (12). Immune cells, adipose tissue, and brain tissue all include peripherally dispersed niacin receptors. Niacin and its derivatives increase high-density lipoprotein (HDL) cholesterol levels while lowering levels of very low-density lipoprotein (VLDL), low-density lipoprotein (LDL), and lipoprotein. (27).

Due to the low tryptophan content of high corn-containing diets, niacin deficiency may occur in corn-fed situations (26). As a result of niacin deficiency, pellagra disease, which affects the nervous system and digestive system, and also

manifests itself with symmetrical wounds on the sun-exposed skin, occurs (28). In pellegra disease, the color of the skin becomes brown and darkens. Pellagra also causes dermatitis, diarrhea and dementia. In niacin deficiency in dogs, digestive system disorder, diarrhea, dehydration, weight loss, loss of appetite and black tongue disease occur in which ulceration of the tongue appears. In addition, excessive intake of thiamine causes toxicity. Toxicity causes vomiting, diarrhea, vascular dilatation, hyperpigmentation and acanthosis (26).

2.1.3. Choline (Vitamin B4)

The precursor of acetylcholine, a crucial neurotransmitter, is vitamin B_4, commonly known as choline. (29). Choline is a vitamin required for the normal function of cells. The primary dietary supply of methyl groups is choline or its metabolites, which also provide the structural integrity and signaling capabilities of cell membranes. One of the metabolites of choline called betaine takes part in the process of methylating homocysteine to produce methionine. Choline directly influences lipid transport, metabolism, and nerve and cell signaling. Choline availability is essential for typical brain development (29).

In rats fed a diet low in choline, levels of carnitine, an amino acid that prevents obesity, were found to be decreased in the liver, heart, and skeletal muscles. After parenchymal cell death in the liver as a result of choline deficiency, an increase in cell proliferation occurs due to regeneration. Cell proliferation and the resulting increased DNA synthesis speed increase susceptibility to chemical carcinogens. As a result of all these, it has been determined that the probability of developing hepatocarcinoma is higher in animals with choline deficiency (30).

2.1.4. Pantothenic Acid (Antidermatitis factor, Vitamin B5)

Pantothenic acid is formed as a result of the peptide bond of pantoic acid with α-γ-dioxy-β-β-dimethyl butyric acid structure and β-alanine. Not resistant to temperature (31). Pantothenic acid serves as an antioxidant and is crucial for the separation of proteins, lipids, and carbohydrates (28). For the production of the common coenzyme A (CoA), this vitamin serves as a substrate. Beyond its role in oxidative metabolism, CoA also contributes to the synthesis of fatty acids, cholesterol, amino acids, and phospholipids, which promote the growth and function of brain cells. Specifically through CoA, pantothenic acid is involved in the production of several neurotransmitters and steroid hormones (10).

In pantothenic acid deficiency, susceptibility to infectious diseases occurs as a result of slowdown in growth and deterioration of the mucosa. In rats,

pantothenic acid deficiency causes regression in growth, reproductive disorders, whitening of black hair and hemorrhagic necrosis. In addition to all these, nervous system disorders and digestive system inflammations occur in animals (26).

2.1.5. Pyridoxine (Vitamin B6)

Vitamin B_6 occurs as three main chemicals, pyridoxal (PL), pyridoxamine (PM), and pyridoxine (14). The chemical name of pyridoxine is 2-methyl-3-hydroxy-4,5-dihydroxy methyl pyridine (32). Pyridoxine is involved in the biosynthesis of structural body protein and amino acid metabolism. While pyridoxine is directly bound to albumin, it is not bound to plasma. Thus, pyridoxine is quickly taken up by erythrocytes and converted to pyridoxal phosphate and pyridoxal (5). The amino acid metabolism of vitamin B_6 is important in the manufacture of neurotransmitters such dopamine, serotonin, -aminobutyric acid (GABA), noradrenaline, and melatonin in addition to its role as a crucial cofactor in the folate cycle. It is a rate-limiting cofactor that also regulates the rate at which different compounds enter the metabolism of amino acids (12).

Pyridoxine, a form of vitamin B_6, is necessary for the production of neurotransmitters. It also prevents the release of neurotoxic glutamate and restores sensory nerve function. (33). The amount of vitamin B_6 present influences how these neurotransmitters are produced in different ways. Even a minor vitamin B_6 shortage causes the synthesis of GABA and serotonin to be downregulated. As a result, GABA's ability to suppress neuronal activity is lost, causing disturbances in sleep, behavior, and cardiovascular function as well as a loss of control over the hypothalamus-pituitary axis' ability to produce hormones (12). Additionally, vitamin B_6 has an impact on immunological response, gene transcription, and brain glucose control. (10-34). In addition, vitamin B_6 balances the nervous metabolism (33). In general, higher levels of pyridoxal-51-phosphate are linked to higher levels of functional indices and inflammatory biomarkers (12).

2.1.6. Biotin (Vitamin B7)

Biotin is formed by the combination of an imidazole and thiophene ring (31). It is a co-enzyme in the structure of enzymes that act as catalysts in carboxylation or decarboxylation events such as acetyl Co-A carboxylase, pyruvate carboxylase, propionyl Co-A carboxylase (5). The modulation of hepatic glucose absorption, gluconeogenesis, lipogenesis, insulin receptor

transcription, and pancreatic beta-cell activity are all important aspects of glucose metabolism and hemostasis that are influenced by biotin (35).

As a result of the experiments, it has been revealed that biotin deficiency occurs in animals fed raw eggs. The avidin in raw egg white is not affected by the complex proteolytic enzymes it forms by combining with biotin, thus preventing resorption. In humans, after consuming diets containing avidin for several weeks, dermatitis, swelling of the skin and mucous membranes, numbness, nausea and muscle pain occur. Generally, anemia and hypercholesterolemia occur. Skin changes occur in chickens, guinea pigs and rabbits with biotin deficiency. Seborrhea and alopecia occur as a result of dermatitis and disruption of fat metabolism; also appear perosis and lameness (26).

2.1.7. Folic Acid (Vitamin B9)

The chemical structure of folic acid consists of the combination of pyteridine, p-aminobenzoic-acid and glutamic acid (36). Folic acid is common in plant and animal tissues. It dissolves easily in alkaline environments. Folic acid itself is not bioactive, but a form of tetrahydrofolic acid called co-enzyme F is active (31). Tetrahydrofolic acid is a cofactor of enzymatic reactions involving a single carbon transfer (5). It also takes part in purine, pyrimidine and thus DNA synthesis (31). Inhibition of neuronal development can result from a real or functional folate deficit, which is accompanied by a decline in purine/pyrimidine production and genomic and non-genomic methylation processes in brain tissue. DNA stability is lowered as a result of repair, gene transcription, and expression. Also folic acid deficiency; impairs the propagation of action potentials, compromising atrophy, demyelination, and membrane phospholipid integrity (6).

In folic acid deficiency; growth retardation, decreases in hemoglobin, leukocyte, erythrocyte and thrombocyte values occur in chickens. Folic acid deficiency in humans and animals causes leukopenia, megablastic anemia, anorexia, weakness, gingivitis and diarrhea (26).

2.1.8. Cobalamin (Antipernicious Factor, Extrinsic Factor, Vitamin B12)

Cobalamins are complex cofactors that are used by a limited number of enzymes and contain natural cobalt in their structure (37). Cobalamins; It is divided into five groups as methylcobalamin, deoxyadenosylcobalamin, hydroxycobalamin, aquacobalamin and cyanocobalamin. Methylcobalamin and deoxyadenosylcobalamin play a role in metabolic functions (38). Vitamin B_{12}

supports the survival and remyelination of nerve cells (33). Because of the crucial roles that cobalamin, pyridoxine, and thiamine play in the nervous system, they are frequently referred to as "neurotropic" vitamins. (39). Cobalamin deficiency causes megaloblastic anemia (40).

The most important finding in humans in cobalamin deficiency is pernicious anemia. In pernicious anemia, poiclocytosis, anisocytosis anemia, atrophy of the gastrointestinal tract mucosa and degenerative disorders in the spinal cord occur. Growth retardation is observed in cobalamin deficiency in chickens, and a decrease in egg production in adults (26).

2.1.9. Thiamine (Antiberiberic Vitamin, Aneurin, Vitamin B1)

The first vitamin ever found, vitamin B_1 is crucial for energy metabolism (41). Thiamine and aneurine are other names for vitamin B_1 (42). It is involved in the intermediate metabolism of carbohydrates in all body tissues. Thiamine stores are very high in the liver, heart and kidneys in the body (28). Thiamine is synthesized in prokaryotes, yeasts, and plants in intricate ways. Specific high-affinity transporters that are necessary for thiamine homeostasis allow animal cells to take up the vitamin. Thiamine is phosphorylated to higher phosphate derivatives inside of cells. Thiazole and pyrimidine rings are joined by a methylene bridge to form the chemical structure of the compound (31). There is a very active N in the thiazole ring and an NH_2 (amino group) in the pyrimidine ring. The methylene bridge connecting the thiazole and pyrimidine rings is very weak. Especially if it is heated in alkaline solution, this bridge is broken. With the rupture of this bridge, the vitamin loses its properties (43).

2.1.9.1. Thiamine Derivatives

Thiamine is naturally found in the free form and in the form of thiamine monophosphate (ThMP), thiamine diphosphate (ThDP) (or thiamine pyrophosphate), thiamine triphosphate (ThTP). Finally, another derivative called thiamine adenosine thiamine triphosphate was discovered. Recently found in *E. coli*, adenosine thiamine triphosphate builds up in situations when there is a lack of carbon and could function as an intracellular signal (44). In animal tissues, it is found in the form of thiamine pyrophosphate (45). All forms of thiamine (except adenosine triphosphate) are found in plant and animal tissues. However, plant tissues contain more free thiamine form than animal tissues. ThMP is a by product of the enzymatic hydrolysis of ThDP in animal tissues, and it has no recognized physiological fuction (46). The existence of thiamine triphosphate

in eukaryotic cells was initially hypothesized in the 1950s, but research in this field has been impeded by the absence of analytical techniques to measure the tiny ThTP rings that are present in most cells. With the development of HPLC (High performance liquid chromatography) techniques in the late 1970s and early 1980s, progress in this field proceeded (47). Most of these approaches derivatize thiamine to produce intensely fluorescent thiochrome derivatives, which boosts sensitive and selective. According to later studies, ThTP has been identified in most of the cells analyzed so far, from bacterial to people (48). The process of ThTP production is still unknown, though being partly described in *E. coli*. It is a soluble enzyme that causes ThDP, ADP, or ATP to be converted into adenosine thiamine triphosphate (46).

It is possible for bacteria, certain protozoa, plants, and fungus to synthesize thiamine (49-50). Thiamine-phosphate synthase joins the independently produced thiazole and pyrimidine moieties to create ThMP. Thiamine is active in metabolism as thiamine pyrophosphate. Thiamine pyrophosphate is essential for normal carbohydrate, nucleic acid and amino acid metabolism and plays a central role in metabolism. It is one of the key cofactors used by the pyruvate dehydrogenase complex in the decarboxylation of alpha-keto acids in the TCA cycle, which produces acetyl coenzyme A from pyruvate. Thiamine pyrophosphate and other thiamine phosphate esters are involved in the transmission of nerve impulses (51). Various forms of thiamine are involved in the production of adenosine triphosphate and the oxidation of macronutrients as a coenzyme (52). Neurotransmitter biosynthesis and production depend on thiamine-dependent enzymes (42). These enzymes, which are involved in carbohydrate catabolism and exhibit decreased activity when thiamine is deficient, include mitochondrial pyruvate dehydrogenase, -ketoglutarate dehydrogenase complexes, and cytosolic transketolase (42).

2.1.9.2. Properties of Thiamine

Thiamine is one of the water-soluble vitamins with the lowest stability at neutral pH. It is not stable at alkaline pH values. While thiamine has maximum stability between pH 2.0 and 4.0, losses may occur after heat treatment in foods with low acidity (51-53). Thiamine is also available in commercial forms such as hydrochloride and mononitrate salts, which are commonly used in food fortification and nutritional supplements (53).

Thiamine; It is abundant in grains, peas, beans, nuts, brown rice and red meat. Cooking can cause some thiamine in food to be lost. Because it is a water-

soluble vitamin, extra amounts are eliminated in the urine rather than being retained by the body. Severe exhaustion becomes evident within 18 days in individuals fed a thiamine-deficient diet (54).

2.1.9.3. Thiamine Degrading Enzymes

Thiaminases are enzymes that can break down thiamine (55). Thiaminase I is a pyrimidine transferase with a wide range of receptors that it may employ. Shellfish, some freshwater fish internal organs, fern species (*Pteridium aquilinum*), and some bacteria (*Bacillus thiaminolyticus*) all contain thiaminase I. Although its physiological relevance is unknown, this enzyme causes poisoning in both humans and animals. Thiaminase II, on the other hand, is a hydrolase that breaks down thiamine at its thiazole and pyrimidine moieties and is frequently connected to rumen acidosis. Found in microorganisms such as *Bacillus subtilis* (56-57).

2.1.9.4. Biochemical Function of Thiamine

An essential enzyme for the Krebs cycle is pyruvate dehydrogenase. As pyruvate enters the Krebs cycle to produce acetyl-coenzyme A, this enzyme plays role in the oxidative decarboxylation of pyruvate. The Krebs cycle enzyme -ketoglutarate dehydrogenase is responsible for carrying out the oxidative decarboxylation of -ketoglutarate to succinyl-CoA, and it is this enzyme that regulates the reaction's rate. An additional mechanism for the oxidation of glucose is provided by transketolase, which works in the pentose phosphate pathway. The activity of these enzymes is reduced by thiamine shortage. This decrease varies depending on the enzyme and is strongly cell type dependent (58).

These enzymes become less active when there is a thiamine deficiency. There is a significant cell type dependence in this decrease, which occurs at various amounts for various enzymes (59). Lactic acid concentrations rise as a result of decreased pyruvate input into the Krebs cycle. This condition is associated with localized acidosis in damaged areas of the brain (59-61). Death of vital cells has been linked to mitochondrial dysfunction, acidosis and apoptosis (62-64). Acetylcholine is a crucial neurotransmitter chemical in the nervous system and its synthesis is lost when acetyl-CoA production declines (65-66). When thiamine levels are low, it is believed that the loss of -ketoglutarate dehydrogenase activity affects the intracellular and extracellular levels of a number of neurotransmitters, such as glutamate, aspartate, and g-aminobutyric

acid (67-68). Neuronal losses occur as a result of excessive secretion of these neurotransmitters (69).

2.1.9.5. Effect of Thiamine on the Nervous System

In the metabolism of the brain, thiamine is crucial. Thiamine, pyridoxine and cobalamin protect nerves against harmful environmental effects and support new cell growth (33). Thiamine has a structural role in addition to its coenzymatic role in metabolism (16-70). It has a significant impact on the axoplasmic, mitochondrial, and synaptosomal membranes' structure and operation (71-72). It contributes to myelinogenesis, synapse development, axonal growth, and cellular differentiation (73-74). Thiamine and other B vitamins control brain growth in fetuses and the first few months after birth (75-76).

2.1.9.6. Transport of Thiamine

Thiamine is taken up by the small intestines through a transport system. Thiamine is received by the liver, heart, and other blood cells across the blood-brain barrier in addition to the neuronal tissue where it is transferred from the blood to the cerebrospinal fluid. Following entry into the cell, transport proceeds across the mitochondrial and nuclear membranes. Thiamine-sensitive megaloblastic anemia is caused by the absence of a functioning transporter (77-78). Mutations in the SLC19A2 thiamine transporter protein gene have been linked to thiamine-sensitive megaloblastic anemia in patients (79). Thiamine is quickly transformed from its inactive form (thiamine) to its active form (thiamine pyrophosphate) in the brain and liver after consumption thanks to a specialized enzyme called thiamine diphosphokinase (42).

2.1.9.7. Thiamine Deficiency

Daily thiamine requirement is directly proportional to calorie intake. For healthy people, it varies from 1.0 to 1.5 mg per day (80). In cases where the carbohydrate content of the diet is high, the amount of thiamine needed in the diet increases (81). Heat sensitivity and water solubility are both characteristics of thiamine. Therefore, some rice loses its vitamins when washed in water or cooked in water. Thiaminases and thiamine antagonists are anti-thiamine factors found in a variety of dietary items that render thiamine inactive. Fish, shellfish, certain microorganisms, and foods that are raw or fermented contain heat-sensitive thiaminase. When ingested without being heated, these foods contain

anti-thiamine properties. There are thermostable thiamine antagonists in a few plants, such as tea and fern. Additionally, they involve polyphenols that can also be contained in blueberries, red currants, red beets, brussels sprouts, red cabbage, and coffee. It reacts with thiamine to produce non-absorbable thiamine disulfide. Ascorbic acid, tartaric acid, and citric acid, which are present in many vegetables and fruits, might neutralize the anti-thiamine action of tannins, an essential component of fermented tea leaves (82).

Thiamine deficiency is primarily due to inadequate thiamine intake. Due to increasing needs in circumstances like hyperthyroidism, pregnancy, and lactation, secondary thiamine insufficiency develops. In addition, taking anti-thiamine factors into the body can also cause thiamine deficiency. In addition, thiamine deficiency can happen due to impaired absorption in long-term diarrhea and decreased thiamine utilization in severe liver diseases (42). Elderly patients are more susceptible to thiamine deficiency (83). Thiamine deficiencies may also happen in offspring if the mother receives insufficient amounts of thiamine (84).

The body can only store a little amount of thiamine. Adults have subjective symptoms such as heart disease, stroke, and motor nervous system problem 2 to 3 weeks after eating a diet low in thiamine (26-82).

2.1.9.7.1. Clinical Symptoms

First to show up in thiamine shortage are non-specific symptoms including weariness, lack of appetite, and constipation (85). In the first or early phase, urinary thiamine excretion is significantly decreased due to insufficient thiamine intake brought on by malnutrition, malabsorption, or aberrant metabolism. In the second stage, or biochemical stage, sees a considerable reduction in transketolase activity. Diverse general symptoms, such as decreased appetite, sleeplessness, aggression, and weakness, start to show up in the third stage, often known as the physiological stage. At the fourth stage, known as the clinical stage, a number of symptoms that are often present in thiamine deficient disorder, such as ophthalmoplegia, bradycardia, peripheral edema, and intermittent claudication, may appear. At the fifth or anatomical stage, histopathological alterations brought on by cell structure damage can be evident in the form of heart enlargement, cerebellar granular layer degeneration, and swollen microglia in the brain. It is interesting that there are no distinct deficiency symptoms in the first three phases (42). In thiamine deficiency, a nervous system disorder called beriberi, which

causes swelling and pain in the joints and balance disorders, occurs in humans and causes encephalopathy (5-26).

Thiamine deficiency in the rumen can dramatically limit microbial protein production in animals with thiamine shortage, which can result in poor growth (21). In thiamine deficiency, disorders occur in the nervous system and cardiovascular system. In dogs, thiamine deficiency causes muscle weakness, growth retardation, and polyneuritis. Opistotonus is seen in pigeons in thiamine deficiency (26). Cerebrocortical necrosis occurs in ruminants with thiamine deficiency. In foxes and minks, Chastek paralysis with ataxia, incoordination, pupillary dilatation, weakening of the papillary reflexes, convulsions and a ventroflexion posture of the head occurs in thiamine deficiency (86).

3. Cerebrocortical Necrosis (Polioencephalomalasi)

Cerebrocortical necrosis, also called polioencephalomalacia; It is a nervous system disease that develops as a result of thiamine deficiency due to various reasons, can be seen in all ruminants, especially in young cattle, and is characterized by focal necrotic lesions in the cerebral cortex (87-88). It is usually a disease of young cattle, the incidence of age varies depending on population exposure. The incidence is also seen in sheep in the age group from weaning to 18 months. Merino sheep are thought to be much more resistant than other breeds (87). Cerebrocortical necrosis usually occurs in ruminants, as energy generation from glucose in the brain of ruminants occurs via the pentose phosphate cycle (89).

3.1. Causes of Occurrence

The main causes of cerebrocortical necrosis in ruminants are feeding animals with carbohydrate-rich feeds, consuming water and feed containing high sulfate or sulfite compounds, and high doses and long-term use of thiamine antagonist drugs (90). High sulfate levels in the diet can affect plasma thiamine levels and decrease duodenal thiamine absorption (91). In addition, the fern plant contains thiaminase and thiamine deficiency may occur in animals fed this plant (92). During the first several weeks of life, calves are fully dependent on thiamine-containing feed (93). Milk and colostrum contain large amounts of thiamine. Deficiency may occur if sufficient amount of thiamine is not taken from milk and colostrum (94). The requirement for thiamine in ruminants is determined by factors such as the animal's age, nutrition, feed quality and

type, housing conditions, and the presence of infection or parasite infestation. A deficiency of nutrients or the presence of a thiamine antagonist both cause thiamine insufficiency (95). Another cause of cerebrocortical necrosis is the decreased thiamine production in the rumen, which is caused by digestive disorders in the fore stomach. In addition, it is thought that this disease is formed as a result of increased ruminal thiaminase and thiamine deficiency in the rumen and other tissues (96-97). Cerebrocortical necrosis is often associated with rumen acidosis (98-99). Rumen acidosis caused by excessive grain production can cause cerebrocortical necrosis by lowering the rumen pH to a value close to the optimum for thiaminase activity (100). Thiamine deficiency is also thought to aggravate the symptoms of lactic acidosis. Because thiamine pyrophosphate is a coenzyme required for pyruvic and lactic acid metabolism (101). An increase in pyruvic acid concentration occurs in thiamine deficiency, and it is stated that this may be toxic to neurons (102).

Cerebrocortical necrosis may be experimentally triggered by thiamine antagonists like pyrithiamine, oxythiamine, and amprolium (103). Pyritamine is a thiamine pyrophosphate inhibitor and interferes with thiamine phosphorylation. Oxythiamine is also known to be a potent inhibitor of several enzyme systems linked to thiamine pyrophosphate (104). Experiments in poultry show that Amprolium inhibits intestinal absorption of thiamine (105). Another experiment shows that low Amprolium levels in the rat cerebral cortex also inhibit thiamine uptake (106). Thiamine absorbed from the intestines in calves treated with Amprolium can be excreted directly in the urine without a metabolic process (107). Since glucose oxidation in the brain of ruminants occurs via the pentose phosphate cycle, polyocephalomalacia usually occurs in ruminants (89).The occurrence of cerebrocortical necrosis in calves given parenterally with Amprolium indicates that it can act on the cerebral cortex after absorption (104). Oxythiamine is easily phosphorylated, and the resulting oxythiamine pyrophosphate is a powerful antagonist of numerous thiamine-dependent enzymes. It has been demonstrated that pyrithiamine inhibits thiamine phosphorylation and causes thiamine accumulation in the brain (108). A study was reported that when mice and rats were given oxythiamine and pyrithiamine, their urine elimination of thiamine increased (109). In calves receiving oxythiamine and pyrithiamine, a significant reduction in thiamine concentration in the cerebrum was noted, although there was no evidence of a reduction of thiamine in the stomach contents and liver (110).

Although a study did not prove that cobalt deficiency causes thiamine deficiency, it has been reported that cobalt fortification is beneficial in avoiding

the occurrence of favorable conditions for the formation of cerebrocortical necrosis under certain conditions (111).

Cerebrocortical necrosis in animals and Wernicke's syndrome in humans are associated with thiamine deficiency and the resulting encephalopathies are similar (112).

3.2. Pathogenesis

The brain first uses glucose for energy production. Glucose enters the brain by diffusion after crossing the blood-brain barrier (113). During the Krebs cycle, 30 percent of the glucose taken in by the brain is completely oxidized (114). Thiamine pyrophosphate, which makes up 80 percent of the thiamine in nerve tissues and is employed as a cofactor, is a thiamine-dependent enzyme such as pyruvate dehydrogenase and α-ketoglutarate dehydrogenase that is required for the cerebral metabolism of glucose (115). Because of thiamine enters the structure of transketolase, which has a significant function in the metabolism of carbohydrates, glucose catabolism of brain glia cells is impaired in its deficiency. Therefore, the nutrition of the neurons is disrupted and necrosis occurs in the neurons. Glucose accumulates in the blood as a consequence of the nervous system's inability to use it, and hyperglycemia may happen (116).

Transketolase is abundant in RBC and neurons, and pentose phosphate pathway activity decreases in transketolase deficiency, and pyruvate metabolism is impaired. The pentose phosphate pathway is very important for glucose metabolism. ATP-dependent Na-K metabolism of nerve cells is impaired as a result of enzymatic changes due to thiamine deficiency. Water and Na enter the neurons, resulting in intraneuronal edema. Intracranial pressure increase occurs as a result of intraneuronal edema. Infiltration of macrophages into the cerebral cortex occurs and proliferation occurs in the capillary endothelium. As a result of all these, laminar cortical necrosis and polioencephalomalacia are formed (57).

3.3. Clinical Symptoms

Affected animals develop immediate clinical signs such as anorexia, depression, incoordination, grinding of teeth, opistotonus, erect ears, inability to stand, decreased vision, blindness, intermittent convulsions, side-lying, and the animal may go into a coma before dying (90-117). If untreated, these animals die within 2 to 6 days after developing clinical signs (118).

3.4. Diagnosis

Since standard laboratory testing for thiamine deficiency are not available, the diagnosis is primarily clinical, and waiting for test results may cause the diagnosis to be delayed. Thiamine's urinary excretion is not a highly reliable method for measuring tissue reserves. The thiamine pyrophosphate impact assay's measurement of transketolase activity is the most credible way to determine the functional status of thiamine. The activity of erythrocyte transketolase is a reliable indication of tissue reserves. In erythrocytes, which do not contain mitochondrial, the only way used to produce NADPH is the pentose phosphate way. Furthermore, NADPH is essential for the preserve structure of red blood cells as well as the reduction of iron, hemoglobin, and glutathione. Transketolase is an enzyme that catalyzes reactions in the pathway of pentose phosphate and requires thiamine pyrophosphate. Red blood cell transketolase activity is thus regarded as a reliability diagnotic marker of thiamine status. For the erythrocyte transketolase test, the haemolysed blood sample should be incubated with high amounts of ribose 5-phosphate while thiamine pyrophosphate is added. Following the incubation period, the quantity of product generated and that remaining substrate are measured. The lack of an increase in enzyme activity as a result of the addition of thiamine pyrophosphate shows that the sample was initially lacking in thiamine. The level of thiamine deficiency is represented as a percent of warning above the control level. A fifteen percent rise or greater in enzyme activity is a sure evidence of lacking (42).

Although high blood pyruvate and lactate levels are beneficial, numerous tests are false-positive, making diagnosis challenging. For instance, thiamine deficiency might be mistaken for conditions such as sepsis, cardiogenic shock, and meningitis. Yet, this condition is linked to lactic acidosis. But the persistence of lactic acidosis and its rise following glucose load significantly support the diagnosis. Clinical response to thiamine treatment is the most practical indication for diagnosis. It can be assumed that a thiamine shortage is the cause of the disease if the patient reacts to treatment. Besides, even at high doses, thiamine is not harmful (42).

Erythrocytes do not have the active transport function that hepatocytes and intestinal absorption cells have, and the thiamine reserve in the blood is only 0.8% of the whole body. Therefore, thiamine deficiency in the body may not affect the amount of thiamine in the blood. Thus, it is believed that plasma thiamine levels are not an accurate marker of thiamine shortage and cerebrocortical necrosis (107). Histological evaluation of brain tissues and other biochemical evidence

of a lack of thiamine, such as reduced tissue levels of the vitamin, low levels of blood ketoacids, raised blood levels of pyruvate kinase, and abnormalities in red blood cells transketolase, can be used to confirm the diagnosis of cerebrocortical necrosis (118). It is known that an increase in blood pyruvate level is an indicator of thiamine deficiency, but does not show a pathognomy on its own (119). In addition, the brain of ruminants affected by cerebrocortical necrosis shows autofluorescent spots in the cerebral cortex under ultraviolet light (120). The main cause of brain fluorescence is increased thiamine deficiency and is thought to be a characteristic condition for cerebrocortical necrosis (112). In a study in which cerebrocortical necrosis was experimentally induced in lambs with Amprolium, it was reported that serum AST, ALT, CPK and glucose levels increased, while thiamine levels decreased significantly (121).

3.5. Necropsy Findings

Necropsy shows congestion and scattered petechiae in the leptomeninges. An increase in the amount of cerebrospinal fluid can be detected (117). In acute events or in young animals, only cerebral edema and swelling can be seen as a result of circulatory disorders. In advanced cases, softening occurs in the brain consistency (122). A yellowish discoloration is seen in the cerebral cortex (123). Lesions usually occur in the dorso-medial, dorso-lateral, and dorsal gyri. In cerebrocortical necrosis, changes occur mostly in gray matter. In the gray matter of the cortex, first focal, bilateral, symmetrical, then diffuse necrosis and malasia occur. In severe effects, the gray matter is completely destroyed and internal hydrocephalus may develop (122). The lateral and third ventricles are enlarged and the brain appears smaller than normal. Enlargement of the subarachnoid space occurs (87).

In animals that survived the disease, the areas of lesions are sunken in appearance and glious scarring areas are formed. In addition, cysts may form under the cerebral cortex (122).

3.6. Microscopic Findings

In cerebrocortical necrosis, the lesions are mostly in the brain or spinal cord's gray matter. This indicates that cerebrocortical necrosis lesions have affinity for gray matter compared to white matter (124). Several studies have reported that pathological lesions occur as a result of neuronal damage due to astrocytic edema and loss of cell volume control (59-125). Histopathologically, multiple laminar and focal spongy areas are observed in the cerebral cortex

in cerebrocortical necrosis (117). The leptomeninges are thickened, and the neurons in the impacted regions are small and acidophilic (87). Necrosis of cerebrocortical neurons is seen with perineuronal and peri-capillary edema. It has been found that edema is formed in addition to neuronal damage and is a feature of cerebrocortical necrosis (104-126). Edema sometimes causes a severe increase in intracranial pressure that forces the posterior part of the cerebellum to enter the foramen magnum (127). It is thought that necrosis is formed due to intracellular fluid accumulation in astrocytes, narrowing of capillaries and related oxygen deficiency (122). Hemorrhagic areas in the brain, degeneration of Purkinje cells and neuronal necrosis in the corpus striatum may occur. Often the thalamus and cerebellum are also affected (127).

Cerebrocortical necrosis has been accepted histopathologically as an anoxic change (88-126). Since the neuronal lesion of cerebrocortical necrosis is anoxic type, ischemia can be considered as a possible cause of the disease (128). In an experimental study in goats, hemorrhagic foci were found in the brain. This state is thought to be caused by low oxygen levels and raised cranial pressure (124). Since the brain depends on glucose for energy via the pathway of pentose phosphate, hypoxia can arise from inadequate energy utilization. Thiamine is required for transketolase in this pathway (124).

3.7. Treatment

Thiaminase is thought to need to react with a certain common substrate such as a picoline-like base to produce a product of the reaction that may function as a thiamine inhibitor. According to numerous research, co-substrates like certain anthelmintics and tranquilizers employed in the treatment of animals may activate thiaminase in vitro. Yet it is considered that thiaminase function in ruminants can lead to drug inactivation, decrease drug efficiency, and potentially cause the drug to undergo a more toxic form of a reaction (129). Finally, in vitro research has demonstrated that thiaminase only rapidly degrades thiamine when a secondary substrate (co-substrate) is present. Because nicotinic acid is a co-substrate, it is possible that thiaminase will cause these two types of vitamins to become depleted. Therefore, multivitamin supplements are recommended for the treatment of cerebrocortical necrosis cases (130). It is claimed that yeast (*Saccharomyces cerevisia*) has a stimulating effect on the gastrointestinal system due to the enzyme and thiamine it contains and has a substantial influence on the rumen ecology and population of bacteria (131). As a result, yeast raises the population of anaerobic and cellulotic microorganisms and may help in the

digestion of cellulose-rich feeds (132). They reported that yeast can prevent metabolic acidosis by inhibiting lactate production in acute ruminal acidosis (133).

Thiamine deficiency is frequently coupled with other B-complex deficits, and therapy with a variety of water-soluble vitamins is recommended. For several weeks, five to ten times the normal daily dosage is frequently advised. Magnesium, a transketolase cofactor, for the treatment of thiamine resistance and the commonly accompanied hypomagnesemia, magnesium sulfate (one to two ml of a fifty percent solution intramuscularly) should be given with thiamine. Some congenital metabolic diseases respond to thiamine therapeutic dosages (five to twenty mg/day). Thiamine is a reliable and affordable therapeutic agent that can stop irreversible harm, thus even if the diagnosis is uncertain, it is still advised that the patient be treated with it (42).

4. Result

The complex necessary for the metabolism of vitamins has been observed. Thiamine, one of the first vitamins to be discovered, is a water-soluble vitamin. B vitamins plays an important role in energy metabolism and acts as a coenzyme. B vitamins are involved in every process of the catabolic process and this process is negatively affected in their deficiency. In particular, thiamine is a coenzyme to various enzymes in cellular energy production, the citric acid cycle, and the phosphate-pentose pathway. In thiamine deficiency, the activity of the enzymes it functions as a coenzyme decreases and therefore sufficient energy cannot be produced. Thiamine deficiency can be caused by antithiamine factors, rumen acidosis, and thiamine antagonists. Thiamine is important for both human and animal health and its deficiency causes nervous system and cardiovascular system disorders. In thiamine deficiency, especially in young ruminants, cerebrocortical necrosis occurs and the animal shows nervous symptoms. While economic losses are shaped due to anorexia due to mild thiamine deficiency, economic losses due to death occur in cases of excessive thiamine deficiency. Correct diagnosis and treatment of thiamine deficiency helps to prevent economic losses.

References

1. McDowell LR. *Vitamins in animal and human nutrition*. John Wiley & Sons; 2000.

2. McDowell LR. *Vitamins in animal nutrition: comparative aspects to human nutrition*. Elsevier; 2012.

3. Friedrich W. *Vitamins*. Walter de Gruyter; 2013.

4. Berg JM, Tymoczko JL, Stryer L. Biochemistry. 5th editio. *New York: NCBI*. 2002.

5. Agyemang-Yeboah F, Oppong SY. B-vitamins role in cellular metabolism and clinical nutrition. *Tropical Series in Health Science*. 2013;1:39-49.

6. Bailey LB, Zempleni J, Rucker R. Folic acid. *Handbook of vitamins*. 2007:385-412.

7. Depeint F, Bruce WR, Shangari N, Mehta R, O'Brien PJ. Mitochondrial function and toxicity: role of B vitamins on the one-carbon transfer pathways. *Chemico-biological interactions*. 2006;163(1-2):113-132.

8. Huskisson E, Maggini S, Ruf M. The role of vitamins and minerals in energy metabolism and well-being. *Journal of international medical research*. 2007;35(3):277-289.

9. Rucker RB, Zempleni J, Suttie JW, McCormick DB. *Handbook of vitamins*. Crc Press; 2007.

10. Zempleni J, Suttie JW, Gregory III JF, Stover PJ. *Handbook of vitamins*. CRC Press; 2013.

11. Kerns JC, Arundel C, Chawla LS. Thiamin deficiency in people with obesity. *Advances in nutrition*. 2015;6(2):147-153.

12. Kennedy DO. B vitamins and the brain: mechanisms, dose and efficacy—a review. *Nutrients*. 2016;8(2):68.

13. Spector R, Johanson CE. Vitamin transport and homeostasis in mammalian brain: focus on Vitamins B and E. *Journal of neurochemistry*. 2007;103(2):425-438.

14. Srivastava M, Rani P, Singh N, Yadav R. Experimental and theoretical studies of vibrational spectrum and molecular structure and related properties of pyridoxine (vitamin B6). *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*. 2014;120:274-286.

15. Depeint F, Bruce WR, Shangari N, Mehta R, O'Brien PJ. Mitochondrial function and toxicity: role of the B vitamin family on mitochondrial energy metabolism. *Chemico-biological interactions*. 2006;163(1-2):94-112.

16. Bâ A. Metabolic and structural role of thiamine in nervous tissues. *Cellular and Molecular Neurobiology*. 2008;28:923-931.

17. Spector R. Vitamin transport diseases of brain: focus on folates, thiamine and riboflavin. *Brain Disord Ther*. 2014;3(02):1-6.

18. Virtanen A. Produktion der Kuhmilch ohne Protein mit Harnstoff und Ammoniumsalzen als Stickstoffquelle und gereinigten Kohlenhydraten als Energiequelle. *Biochemische Zeitschrift*. 1963;338:443-&.

19. Miller B, Meiske J, Goodrich R. Effects of dietary additives on B-vitamin production and absorption in steers. *Journal of Animal Science*. 1986;62(2):484-496.

20. Harmeyer J, Kollenkirchen U. Thiamin and niacin in ruminant nutrition. *Nutrition research reviews*. 1989;2(1):201-225.

21. Candeau M, Kone L. In vitro effect of thiamin on rumen microbial metabolism [sheep]. *Reproduction Nutrition Developpement*. 1980.

22. Smith R, Marston H. Some metabolic aspects of vitamin B12 deficiency in sheep. *British Journal of Nutrition*. 1970;24(4):879-891.

23. Powers HJ, Hill MH, Mushtaq S, Dainty JR, Majsak-Newman G, Williams EA. Correcting a marginal riboflavin deficiency improves hematologic status in young women in the United Kingdom (RIBOFEM). *The American journal of clinical nutrition*. 2011;93(6):1274-1284.

24. Sinigaglia-Coimbra R, Lopes AC, Coimbra CG. Riboflavin deficiency, brain function, and health. In Handbook of Behavior, Food and Nutrition; Springer: Berlin, Germany pp. 2427–2449. 2011.

25. Ashoori M, Saedisomeolia A. Riboflavin (vitamin B2) and oxidative stress: a review. *British journal of nutrition*. 2014;111(11):1985-1991.

26. Kalaycıoğlu L, Serpek B, Nizamlıoğlu M, Başpınar N, Tiftik AM. *Biyokimya*. Nobel Yayın; 2006.

27. Öz Ş, Kılıçarslan A. Vitaminlerin Yaşamımızdaki Yeri Nedir, Ne Olmalıdır? İç Hastalıkları Dergisi. 2012;19:139-43.

28. Gerald F, Combs J. The Vitamins Fundamental Aspects in Nutrition and Health. 3rd editio. Elsevier Academic Press. 3rd editio. Elsevier Academic Press; 2008.

29. Zeisel SH. Choline: needed for normal development of memory. *Journal of the American College of Nutrition*. 2000;19(sup5):528S-531S.

30. Zeisel SH. Choline deficiency. *The Journal of nutritional biochemistry*. 1990;1(7):332-349.

31. Ası T. *Tablolarla Biyokimya (Cilt 2), Ankara, 234s.* 1999.

32. Ezer N. *B vitamini ve türevlerinin model membranlar ile etkileşimlerinin spektroskopik ve kalorimetrik teknikler ile incelenmesi*. Fen Bilimleri Enstitüsü; 2017.

33. Baltrusch S. The role of neurotropic B vitamins in nerve regeneration. *BioMed research international*. 2021;2021:1-9.

34. Anitha M, Abraham PM, Paulose C. Striatal dopamine receptors modulate the expression of insulin receptor, IGF-1 and GLUT-3 in diabetic rats:

effect of pyridoxine treatment. *European journal of pharmacology*. 2012;696(1-3):54-61.

35. Via M. The malnutrition of obesity: micronutrient deficiencies that promote diabetes. *International Scholarly Research Notices*. 2012;2012.

36. Bingöl G. Vitaminler ve enzimler. Ankara: Ankara Üniversitesi Eczacılık Fakültesi; 1977.

37. Selva Kumar S, Chouhan RS, Thakur MS. Trends in analysis of vitamin B12. *Analytical biochemistry*. 2010;398:139-149.

38. Doğan M, Demirci S. Vitamin B12 ve nörolojik hastalıklardaki etkisi. *SD Ü Sağlık Bilimleri Enstitüsü Dergisi*. 2010;1(1):23-29.

39. Reyes-García G, Medina-Santillán R, Flores-Murrieta FJ, Caram-Salas NL, Granados-Soto V. Analgesic effects of B vitamins: a review. *Current Topics in Pharmacology*. 2006;10(1):1-31.

40. Murray RK, Mayes, P.A., Granner, D.K. and Rodwell, V.W. . Harper'ın Biyokimyası, (Çev. G. Menteş, B. Ersöz), Barış Kitapevi, İstanbul, 913s. 1993.

41. Engelking L. *Textbook of veterinary physiological chemistry, updated 2/e*. Academic Press; 2010.

42. Fattal-Valevski A. Thiamine (vitamin B1). *Journal of Evidence-Based Complementary & Alternative Medicine*. 2011;16(1):12-20.

43. Çelik S. Gıda Kimyası Ders Notları (basılmamış), Hacettepe Üniversitesi, Ankara. 2006.

44. Bettendorff L, Wirtzfeld B, Makarchikov AF, et al. Discovery of a natural thiamine adenine nucleotide. *Nature chemical biology*. 2007;3(4):211-212.

45. Ergün A, Tuncer Ş, Çolpan İ, et al. *Hayvan besleme ve beslenme hastalıkları*. Vitamins. 2020:123-124.

46. Bettendorff L, Wins P. Thiamin diphosphate in biological chemistry: new aspects of thiamin metabolism, especially triphosphate derivatives acting other than as cofactors. *The FEBS journal*. 2009;276(11):2917-2925.

47. Kawasaki T. Vitamin B1: thiamin. In Modern Chromatographic Analysis of Vitamins (De Leenheer AP, Lambert WE & Nelis HJ, eds),pp. 319–354. 1992.

48. Makarchikov AF, Lakaye B, Gulyai I, et al. Thiamine triphosphate and thiamine triphosphatase activities: from bacteria to mammals. *Cellular and Molecular Life Sciences CMLS*. 2003;60:1477-1488.

49. Webb ME, Marquet A, Mendel RR, Rébeillé F, Smith AG. Elucidating biosynthetic pathways for vitamins and cofactors. *Natural product reports*. 2007;24(5):988-1008.

50. Begley TP, Chatterjee A, Hanes JW, Hazra A, Ealick SE. Cofactor biosynthesis—still yielding fascinating new biological chemistry. *Current opinion in chemical biology*. 2008;12(2):118-125.

51. Eitenmiller RR, Landen Jr W, Ye L. *Vitamin analysis for the health and food sciences*. CRC press; 2016.

52. Hanninen SA, Darling PB, Sole MJ, Barr A, Keith ME. The prevalence of thiamin deficiency in hospitalized patients with congestive heart failure. *Journal of the American College of Cardiology*. 2006;47(2):354-361.

53. Gregory J. Vitamins in food chemistry. *Food chemistry*. 1996;3:532-590.

54. World Health Organization. *Thiamine deficiency and its prevention and control in major emergencies*. 1999.

55. Murata K. Actions of two types of thiaminase on thiamin and its analogues. *Annals of the New York Academy of Sciences*. 1982;378(1):146-156.

56. Jenkins AH, Schyns G, Potot S, Sun G, Begley TP. A new thiamin salvage pathway. *Nature chemical biology*. 2007;3(8):492-497.

57. Sezgin Ş. *Buzağıların İç Hastalıkları (Olgu Tartışmalı)*. Buzağıların Sinir Sistemini Etkileyen Önemli Hastalıklar. 2019:419-423.

58. Butterworth RF, Kril JJ, Harper CG. Thiamine-dependent enzyme changes in the brains of alcoholics: relationship to the Wernicke-Korsakoff syndrome. *Alcoholism: Clinical and Experimental Research*. 1993;17(5):1084-1088.

59. Aikawa H, Watanabe IS, Furuse T, et al. Low energy levels in thiamine-deficient encephalopathy. *Journal of Neuropathology & Experimental Neurology*. 1984;43(3):276-287.

60. McCandless DW, Schenker S. Encephalopathy of thiamine deficiency: studies of intracerebral mechanisms. *The Journal of clinical investigation*. 1968;47(10):2268-2280.

61. Hakim A. The induction and reversibility of cerebral acidosis in thiamine deficiency. *Annals of Neurology: Official Journal of the American Neurological Association and the Child Neurology Society*. 1984;16(6):673-679.

62. Bettendorff L, Mastrogiacomo F, Kish SJ, Grisar T. Thiamine, thiamine phosphates, and their metabolizing enzymes in human brain. *Journal of neurochemistry*. 1996;66(1):250-258.

63. Pannunzio P, Hazell AS, Pannunzio M, Rao KR, Butterworth RF. Thiamine deficiency results in metabolic acidosis and energy failure in cerebellar granule cells: an in vitro model for the study of cell death mechanisms in

Wernicke's encephalopathy. *Journal of neuroscience research*. 2000;62(2):286-292.

64. Wang JJ-L, Hua Z, Fentress HM, Singleton CK. JNK1 is inactivated during thiamine deficiency-induced apoptosis in human neuroblastoma cells. *The Journal of Nutritional Biochemistry*. 2000;11(4):208-215.

65. Heinrich C, Stadler H, Weiser H. The effect of thiamine deficiency on the acetylcoenzymeA and acetylcholine levels in the rat brain. *Journal of neurochemistry*. 1973;21(5):1273-1281.

66. Jankowska-Kulawy A, Bielarczyk H, Pawełczyk T, Wróblewska M, Szutowicz A. Acetyl-CoA and acetylcholine metabolism in nerve terminal compartment of thiamine deficient rat brain. *Journal of neurochemistry*. 2010;115(2):333-342.

67. Gaitonde M, Fayein N, Johnson A. Decreased metabolism in vivo of glucose into amino acids of the brain of thiamine-deficient rats after treatment with pyrithiamine. *Journal of neurochemistry*. 1975;24(6):1215-1223.

68. Langlais PJ, Zhang SX. Extracellular glutamate is increased in thalamus during thiamine deficiency-induced lesions and is blocked by MK-801. *Journal of neurochemistry*. 1993;61(6):2175-2182.

69. Todd K, Butterworth R. Evaluation of the role of NMDA-mediated excitotoxicity in the selective neuronal loss in experimental Wernicke encephalopathy. *Experimental Neurology*. 1998;149(1):130-138.

70. Haas RH. Thiamin and the brain. *Annual review of nutrition*. 1988;8(1):483-515.

71. Matsuda T, Cooper JR. Thiamine as an integral component of brain synaptosomal membranes. *Proceedings of the National Academy of Sciences*. 1981;78(9):5886-5889.

72. Itokawa Y, Schulz R, Cooper J. Thiamine in nerve membranes. *Biochimica et Biophysica Acta (BBA)-Biomembranes*. 1972;266(1):293-299.

73. Goldberg DJ, Cooper JR. Effects of thiamine antagonists on nerve conduction. I. Actions of antimetabolites and fern extract on propagated action potentials. *Journal of neurobiology*. 1975;6(5):435-452.

74. Reddy TS, Ramakrishnan C. Effect of thiamine deficiency and undernutrition on the lipid composition of rat spinal cord at 21 days of age. *Journal of Neuroscience Research*. 1983;9(2):111-114.

75. Ramakrishna T. Vitamins and brain development. *Physiological Research*. 1999;48:175-188.

76. Georgieff MK. Nutrition and the developing brain: nutrient priorities and measurement. *The American journal of clinical nutrition*. 2007;85(2):614S-620S.

77. Martin P. Molecular mechanisms of thiamine utilization. *Current molecular medicine*. 2001;1(2):197-207.

78. Rindi G, Casirola D, Poggi V, De Vizia B, Patrini C, Laforenza U. Thiamine transport by erythrocytes and ghosts in thiamine-responsive megaloblastic anaemia. *Journal of inherited metabolic disease*. 1992;15(2):231-242.

79. Labay V, Raz T, Baron D, et al. Mutations in SLC19A2 cause thiamine-responsive megaloblastic anaemia associated with diabetes mellitus and deafness. *Nature genetics*. 1999;22(3):300-304.

80. Electrolytes IoMPoDRIf, Water. *DRI, dietary reference intakes for water, potassium, sodium, chloride, and sulfate*. National Academy Press; 2004.

81. Watson A, Walker J, Tomkin G, Finn M, Keogh J. Acute Wernickes encephalopathy precipitated by glucose loading. *Irish journal of medical science*. 1981;150:301-303.

82. Organization WH. *Thiamine deficiency and its prevention and control in major emergencies*. 1999.

83. Lee DC, Chu J, Satz W, Silbergleit R. Low plasma thiamine levels in elder patients admitted through the emergency department. *Academic Emergency Medicine*. 2000;7(10):1156-1159.

84. Seear M, Lockitch G, Jacobson B, Quigley G, MacNab A. Thiamine, riboflavin, and pyridoxine deficiencies in a population of critically ill children. *The Journal of pediatrics*. 1992;121(4):533-538.

85. Brin M. Erythrocyte as a biopsy tissue for functional evaluation of thiamine adequacy. *JAMA*. 1964;187(10):762-766.

86. Milli ÜH, Hazıroğlu R. *Veteriner Patoloji 1.Cilt*. Sinir Sistemi Dejenerasyonları. 2000.

87. Jubb, Kennedy and, Palmer's. Pathology of Domestic Animals. Volume 1. USA: ELSEVER; 2015. p. 309.

88. Terlecki S. Cerebrocortical necrosis in cattle and sheep. *Vet Rec*. 1961;73:23-27.

89. Yarsan E. *Koyun ve Keçi Hekimliği*. Sinir Sistemi Hastalıkları. 2017:455-456.

90. Ok M, Irmak K, Ortatlı M, et al. Kuzularda deneysel oluşturulan serebrokortikal nekrozun profilaksi ve tedavisinde mayanın etkisi. *Veteriner Bilimleri Dergisi Eurasian Journal of Veterinary Sciences*. 1998;14(1):75-85.

91. Goetsch A, Owens F. Effects of Supplement Sulfate (DynamateR) and Thiamin-HCI on Passage of Thiamin to the Duodenum and Site of Digestion in Steers. *Archives of Animal Nutrition*. 1987;37(12):1075-1083.

92. Evans W, Evans IA, Humphreys D, Lewin B, Davies W, Axford R. Induction of thiamine deficiency in sheep, with lesions similar to those of cerebrocortical necrosis. *Journal of Comparative pathology*. 1975;85(2):253-267.

93. Draper HH, Johnson BC. Thiamine Deficiency in the Lamb: Three Figures. *The Journal of Nutrition*. 1951;43(3):413-422.

94. Zintzen H. Vitamin Bl (Thiamin) in der Erniihrung des Wiederkiiuers. Obers. Tieremiihrg. 1, : 273-323. 1973.

95. Edwin E, Markson L, Shreeve J, Jackman R, Carroll P. Diagnostic aspects of cerebrocortical necrosis. *The Veterinary Record*. 1979;104(1):4-8.

96. Edwin E, Jackman R. Ruminal thiaminase and tissue thiamine in cerebrocortical necrosis. *The Veterinary Record*. 1973;92(24):640-641.

97. Piii A. Evidelice of tlıiamine deficieıncy in calves affected ıvitlı cerebrocortical ıiecrosis. *Yet Ree*. 1967;81:178-181.

98. Flachowsky G, Lohnert H, Hennig A. Einige Aspekte zur Vitamin-B1-Versorgung von Lammern na ch provozierter Pansenazidose. *Archiv fur experimentelle Veterinarmedizin*. 1974.

99. Bouda J, Jagoš P, Dvořák V, Pivník L. Syndrome of Cerebrocortical Necrosis Experimentally Induced in Calves by Administration of Amprolium. *Acta Veterinaria Brno*. 1982;51(1-4):109-116.

100. Brent B. Relationship of acidosis to other feedlot ailments. *Journal of Animal science*. 1976;43(4):930-935.

101. Boyd J, Walton J. Cerebrocortical necrosis in ruminants: An attempt to identify the source of thiaminase in affected animals. *Journal of Comparative Pathology*. 1977;87(4):581-589.

102. Robins S, Cotran R, Kumar V. Pathologic basis of diseases 3rd ed. Philadelphia: WB Saunders Co. p; 1984.

103. Little PB. Biochemical and pathological studies of thiamine deficiency and polioencephalomalacia in cattle, Ph. D. Thesis, University of Minnesota USA. 1969.

104. Markson L, Lewis G, Terlecki S, Edwin E, Ford J. The aetiology of cerebrocortical necrosis: The effects of administering antimetabolites of thiamine to preruminant calves. *British Veterinary Journal*. 1972;128(10):488-499.

105. Rogers EF. Ann. Nor. Acad. Sci. gS, 412. 1962.

106. Sharma SK, Quastel J. Transport and metabolism of thiamine in rat brain cortex in vitro. *Biochemical Journal*. 1965;94(3):790.

107. Kasahara T, Ichijo S, Osame S, Sarashina T. Clinical and biochemical findings in bovine cerebrocortical necrosis produced by oral administration of amprolium. 1988;

108. Rindi G, Perri V. Separation and determination of thiamine and pyrithiamine in biological materials by chromatography on polyethylene powder. *Analytical Biochemistry*. 1963;5(3):179-186.

109. Peterson J, Gubler C, Kuby S. Partial purification and properties of thiamine pyrophosphokinase from pig brain. *Biochimica et Biophysica Acta (BBA)-Enzymology*. 1975;397(2):377-394.

110. Edwin E, Lewis G. Section E. Diseases of dairy cattle. Thiamine deficiency, with particular reference to cerebrocortical necrosis. *Journal of Dairy Research*. 1971;38(1):79-90.

111. MacPherson A, Moon F, Voss R. Biochemical aspects of cobalt deficiency in sheep with special reference to vitamin status and a possible involvement in the aetiology of cerebrocortical necrosis. *British Veterinary Journal*. 1976;132(3):294-308.

112. Edwin E, Jackman R. Nature of the autofluorescent material in cerebrocortical necrosis. *Journal of Neurochemistry*. 1981;37(4):1054-1056.

113. Rao J, Oz G, Seaquist ER. Regulation of cerebral glucose metabolism. *Minerva endocrinologica*. 2006;31(2):149.

114. Siebert G, Gessner B, Klasser M. Energy supply of the central nervous system. *Nutrition and neurobiology*. 1986;38:1-26.

115. Ishii K, Sarai K, Sanemori H, Kawasaki T. Analysis of thiamine and its phosphate esters by high-performamze liquid chromatography. *Analytical Biochemistry*. 1979;97(1):191-195.

116. Kaneko JJ, Harvey JW, Bruss ML. *Clinical biochemistry of domestic animals*. Academic press; 2008.

117. Yukio Seimiya HI, and Kan-ichi Ohsima. A Case of Cerebrocortical Necrosis in a Sheep. 1989.

118. Edwin E, Jackman R. A rapid radioactive method for determination of thiaminase activity and its use in the diagnosis of cerebrocortical necrosis in sheep and cattle. *Journal of the Science of Food and Agriculture*. 1974;25(4):357-368.

119. Steyn-Parve EP. In Ciba Foundation Study Groups. No. 28, p. 26. Ed. Wolstenholme, G.E.W., & O ‘Connor, M. London:j. & A. Churchill. 1967.

120. Edwin E, Markson L, Jackman R. The aetiology of cerebrocortical necrosis: the role of thiamine deficiency and of deltapyrrolinium. *British Veterinary Journal*. 1982;138(4):337-349.

121. Ok M, Aslan V, Çiftçi MK, Maden M, Sevinç M, Yener Z. Clinical, Laboratory and Pathological Findings in Cerebrocortical Necrosis (CCN) Induced Experimentally in the Lambs. *Eurasian Journal of Veterinary Sciences*. 1995;11(1):5-11.

122. Çiftçi M, Ortatatlı M, Erer H, Hatipoğlu F, Özdemir Ö. Veteriner Sistemik Patoloji, 2. *Konya, Selçuk Üniversitesi Basımevi*. 2017;2:24,25.

123. Thornber E, Dunlop R, Gawthorne J, Huxtable C. Polioencephalomalacia (cerebrocortical necrosis) induced by experimental thiamine deficiency in lambs. *Research in Veterinary Science*. 1979;26(3):378-380.

124. Lonkar P, Prasad M. Pathology of amprolium-induced cerebrocortical necrosis in goats. *Small Ruminant Research*. 1994;13(1):85-92.

125. Askaa JaM, T. Ultrastructural study of cerebrocortical necrosis in calves. Nor. Vet. Med., 30:126-131 (Vet. Bull., 48: 6169). 1978.

126. Terlecki S, Baldwin B, Bell F. Experimental cerebral ischaemia in sheep: neuropathology and clinical effects. *Acta neuropathologica*. 1967;7:185-200.

127. Markson L, Terlecki S. The aetiology of cerebrocortical necrosis. *British Veterinary Journal*. 1968;124(7):309-315.

128. King J, Roth L, Haschek W. Myocardial necrosis secondary to neural lesions in domestic animals. *Journal of the American Veterinary Medical Association*. 1982;180(2):144-148.

129. Edwin E, Lewis G, Allcroft R. Cerebrocortical Necrosis-A hypothesis for possible role of Thiaminases in its pathogenesiss. *Veterinary Record*. 1968;83(7):176-&.

130. Roberts G, Boyd J. Cerebrocortical necrosis in ruminants: Occurrence of thiaminase in the gut of normal and affected animals and its effect on thiamine status. *Journal of Comparative Pathology*. 1974;84(3):365-374.

131. Broberg G. *Acute overeating with cereals in ruminants*. Ab. Lovisa Nya Tryckeri; 1960.

132. Walli T. Role of yeast culture in rumen ecosystem and animal performance. *International Journal of Animal Sciences*. 1994;9:117-117.

133. Höltershisken. M. P, D., Figueride, M., Feldman, M., Scholz, H. Invetstigalion for using different yeast against acidolic conditions in rumen fluid (in vitro). XVIII World Buiatrics Congres, Agust 29- September 2, Bologno, Italy. . 1994.

CHAPTER IX

SARCOPENIA

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1. Introduction

Sarcopenia, is a combination of the words "sarx" meaning muscle in Greek and "penia" meaning loss, is a syndrome characterized by progressive generalized loss of muscle mass and muscle strength was first described by Irwin Rosenberg in 1988(1). Subsequently several strategies for assessment of body composition were proposed, with particular interest in direct and indirect methods of measuring and estimating muscle mass. In 1998, Baumgartner introduced the definition of sarcopenia into clinical use. At the Initial descriptions of sarcopenia focused on loss of muscle mass and did not consider inclusion of muscle strength or physical impairment as part of the disease process. In the last three decades with increasing interest various definitions have been proposed and many consensuses published definitions by a range of expert groups from around the world. The most widely cited definition nowadays is that proposed by the European Working Group on Sarcopenia in Older People (EWGSOP). EWGSOP revised the definition and diagnostic criteria of sarcopenia in 2019. It showed that muscle strength is the primary determinant in diagnosis, occurs earlier than the loss of muscle mass, and has more substantial effects on quality of life and mortality (2). Sarcopenia is a geriatric syndrome associated with adverse effects on function, quality of life, and survival.

Muscle mass, assessment of muscle strength, and function are decisive in diagnosing and classification of sarcopenia (Table1). Sarcopenia primarily develops with advancing age, and its incidence increases over the age of 65. Physiologically, muscle losses begin in our body from the age oVAf 40, and this loss reaches 8% per decade until the age of 70 and 15% per decade over the

age of 70 (Primary Sarcopenia) (3). However, chronic diseases (diabetes, heart failure, chronic liver diseases, chronic kidney failure, obstructive pulmonary disease, etc.), malnutrition, physical inactivity, smoking, and alcohol use lead to the development of sarcopenia at an earlier age (Secondary Sarcopenia) (4). The prevalence of sarcopenia varies based on factors such as age, gender, race, and residence location (home, hospital, nursing home). In addition to the etiological classification of sarcopenia, according to The European Working Group on Sarcopenia in Older People (EWGSOP) also classifies it based on the duration of development as acute and chronic sarcopenia. Sarcopenia that develops in less than 6 months is considered acute, while sarcopenia that develops over 6 months or longer is classified as chronic sarcopenia. Acute sarcopenia is typically associated with acute illness, surgery or trauma, while chronic sarcopenia is likely associated with chronic and progressive conditions and increases the risk of death. This distinction aims to emphasize the need for periodic sarcopenia assessments in individuals at risk to determine how rapidly the condition is developing or worsening.

Sarcopenia is a significant cause of morbidity (disability, falls, fractures, increase of hospitalization, longer hospital stay etc.) and mortality in both the elderly population and individuals with chronic diseases. Therefore, early diagnosis of sarcopenia, determination of underlying predisposing causes, and elimination of correctable factors are essential.

Table1. Sarcopenia according to 2019 Revised EWGSOP Criteria

(Possible sarcopenia with the presence of criterion 1, The diagnosis becomes definite with the addition of criterion 2, The addition of criterion 3 indicates that sarcopenia is severe.
1. Low muscle strength 2. Low muscle mass and quality 3. Low physical performance

2. Pathogenesis

The development of sarcopenia is multifactorial and distinguishing between advanced age and secondary causes creates a roadmap for patient management. Proinflammatory cytokines (such as tumour necrosis factor alpha (TNF-α) and interleukin-6 (IL-6)) mediators increase with advancing age in pathogenesis and trigger basal inflammation, and this phenomenon

is called “inflammaging”. These immune system changes are thought to be related to muscle loss and functional capacity. With increasing basal inflammation, alpha motor dysfunction, mitochondrial dysfunction, increased oxidative stress, and increased apoptosis, decreasing anabolic hormones (GH, IGF-1, Estrogen, testosterone) with age, malnutrition (decreased calorie and protein intake), and immobility can cause sarcopenia. In sarcopenia is accompanied by a loss of innervation and adaptive changes in the proportions of slow and fast motor units, also as well as in the cross-sectional area of muscle fibers. (5,6). Ageing appears to result in an imbalance between muscle protein anabolic and catabolic pathways, leading to overall loss of skeletal muscle. Cellular changes in sarcopenic muscle include a reduction in the size and number of myofibres, which particularly affects type II fibres. This is partly due to transition of muscle fibres from type II to type I with age, together with intramuscular and intermuscular fat infiltration (myosteatosis), and a decreased type II fibre satellite cells and strength with aging (7). The development of sarcopenia in the course of chronic diseases increases the mortality and complications of the underlying disease and significantly reduces patients’ quality of life. Therefore, controlling the underlying disease and early recognition and control of sarcopenia enhancing the quality of life and survival times.

3. New Topics in Sarcopenia

3.1. Sarcopenic Obesity, refers to the coexistence of sarcopenia clinical, and functional impairments in individuals with a high body mass index (BMI) of 30 kg/m2 or higher. The combination of these two conditions can synergistically exacerbate their negative effects. Sarcopenic obese individuals have been shown to have a higher risk of metabolic disease, cardiovasculer risk factors, and mortality compared to individuals who are solely sarcopenic or obese. This increased risk is attributed to factors such as infiltration of fat into muscle tissue, increased insülin resistance, pro-inflammatory cytokines, oxidative stress and reduced physical activity (8,9).

3.2. Osteosarcopenia, is a geriatric syndrom that describes the coexistence of two chronic musculoskletal disorders associated with aging: osteoporosis and sarcopenia. This combination of conditions can have a synergistic effect, leading to greater impairment in physical function, increased risk of falls and fractures, higher morbidity and mortality rates, and reduced quality of life (10).

4. Case Finding

Sarcopenia is a largely overlooked clinical condition whose usually undiagnosed. Therefore, in practise, case finding approaching is recommended. This approach involves looking for sarcopenia when relevant symptoms are reported. Such as falling, weakness, slowness, self-reported muscle wasting, or difficulties carrying out daily life activities.

In nursing homes, rehabilitation settings, or geriatric inpatient services, is a higher prevalence of sarcopenia. Case finding should be done, especially here. A wide variety of tests and screening tools are now available to characterize sarcopenia in practice and research. The SARC-F (Table 2) questionnaire is a screening tool clinicians can rapidly implement to identify probable sarcopenic patients. SARC-F is an inexpensive and convenient method for sarcopenia risk screening (11). There are Five SARC- F components: Strength, Assistance with walking, rising from a chair, Climb stairs, and falling. SARC- F questionnaire is self-administered by patients as a screen for sarcopenia risk. SARC-F scale scores range from 0 to 10. If the SARC-F score equals or exceeds 4, it best predicts the need for a further, more comprehensive evaluation.

Table 2. SARC-F Questionnaire

Component	Question	Scoring
Strength	How much difficulty do you have in lifting and carrying 10 pounds?	None:0 Some:1 A lot or unable:2
Assistance in walking	How much difficulty do you have walking across a room	None:0 Some:1 A lot, use aids, or unable:2
Rise from a chair	How much difficulty do you have transferring from a chair or bed	None:0 Some:1 A lot or unable without help:2
Climb stairs	How much difficulty do you have climbing a flight of 10 stairs	None:0 Some:1 A lot or unable:2
Falls	How many times have you fallen in the past year	None:0, 1-2 falls:1 4 or more falls:2

5. Diagnosis

According to the EWGSOP 2019 criteria, sarcopenia was defined as low muscle strength combined with low muscle mass either or low physical

performance. Low muscle strength (hand grip strength) is the primary parameter of the diagnosis.

In case of clinical suspicion of sarcopenia, performing the SARC-F questionnaire assessment muscle strength then measuring muscle mass and quality are essential. If the values are below the limits, it clarifies the diagnosis of sarcopenia. The state of accompanying physical performance correlates with the severity of sarcopenia.

Table 3. Methods for measurement of muscle mass, muscle strength, and physical performance

Muscle Mass	Muscle Strength	Physical Performance
Anthropometry	Handgrip strength	Short physical performance battery
Computerize Tomography/ Manyetic Resonance	Chair- Rise Test	Usual gait speed
Dual-Energy/X-Ray absorptiometry		Timed Up and Go Test
Bioimpedance analysis		

5.1. Muscle Strength

The most commonly used methods to measure muscle strength are as follows:

5.1.1. Handgrip Strength

The hand grip dynamometer measures upper extremity muscle strength. It is an easy, inexpensive, and noninvasive method. Low grip strength indicates poor patient outcomes, such as longer hospital stays, increased functional limitations, poor health-related quality of life, and even death. If hand grip strength is below the reference values or thresholds defined by gender or social norms, suspicion of sarcopenia should arise (12,13).

5.1.2. Chair-Rise Test

(also known as the sit-to stand test); widely used to measure the strenght of leg muscles (quadriceps muscle group). It measures the time required for a

patient to rise from a seated position without using their arms, repeated five times. Since the sit-to-stand test requires both strenght and endurance, it is aqualified and suitable measure of strength (11).

5.2. Muscle Quantity/Mass:

Bioimpedance analysis (BIA), Dual -Energy/X-Ray absorptiometry (DXA), Computerize Tomography (CT), Magnetic Reosnance (MRI) are some common methods for estimated measure muscle mass. BIA is the most commonly used technique in clinical practise, being a low-cost, non invasive, and widely spread method. It is useful as a bedside test due to portability. With bioelectrical impedance analysis, is possible to obtain information about fat- free mass and total body water. However,in patients with fluid and electrolyte abnormalities, BIA may not yield accurate results. It may provide accurate results in patients with conditions such as congestive heart failure or kidney failure etc., where there is volume overload or in patients with dehydration (14).

Dual-Energy/X-Ray Absorptiometry (DXA is a fast, easy, and non- invasive technique that measures lean body mass, fat mass, and bone mineral density. It can be used for whole-body measurements or segmental measurements. However, it can not directly measure muscle mass. Despite this, DXA mesurements Show a very good correlation with body composition measurements obtained using other techniques such as CT and MRI.

Magnetic Resonance Imaging (MRI) and Computerized Tomography (CT) are considered gold standard methods for non-invasive assessment of muscle quantity and muscle mass. These imaging techniques can also measure intramusculer adipose tissue (IMAT), which serves as an indicator of muscle quality. The effects of IMAT on muscle strength and mass are associated with moleculer mechanisms such as lipotoxic and inflammatory effects, as well as mechanical factors such as decreased strength and muscle mass loss (15,16). These effects can lead to mobility limitations in individuals with sarcopenia. However, despite their effectiveness, MRI and CT scans are not commonly used in primary care due to high equipment costs, and the need for highly qualified personnel to operate the equipment. Additionally, there are no established absolute cutoff values for muscle quantity/mass assesment using CT or MRI. Limitations such as radiation exposure in CT screening and long examination times in MRI screening make routine screening for muscle mas impractical(17).

Anthropometric measurements such as upper- middle arm circumference, skinfold thickness, or calf circumference (<31 cm) were previously used in the

diagnosis of sarcopenia. However, due to the high margin of error associated with anthropometric measurements, they are not routinely recommended for the diagnosis of sarcopenia (1,18).

5.3. Physical Performance:

Physical performance is associated with the severity of sarcopenia. Physical performance can be assessed using the Short Physical Performance Battery (SPPN), usual gait speed, and time up-and-go test. Physical performance is an objective measurement of movement-related body functions, encompassing muscle and peripheral and central nervous system functions. In the assessment of physical performance, walking speed, the "short physical performance battery," the "timed up and go test," and the "400-meter walk test" can be used. The most commonly used test for evaluating walking speed is the "4-meter usual gait speed test". The EWGSOP recommends this test for assessing physical performance, and a cutoff value of ≤0.8 meters/second indicates severe sarcopenia (11). The short physical performance battery evaluates walking speed, balance, and chair standing together, and a score of ≤8 indicates poor physical performance. The timed up-and-go test considers the time it takes for a person to rise from a chair, walk 3 meters, and return to the chair. In the 400-meter walk test, the person is instructed to walk a 20-meter distance as fast as possible, 20 times. Although these tests are determinants of mortality and outcomes related to sarcopenia, they may not be feasible for elderly individuals with dementia or walking and balance problems (1,19).

6. Treatment

Early recognition and intervention are crucial for improving outcomes in patients with sarcopenia. It is more effective to prevent the progressive loss of skeletal muscle mass, strength, and function rather than attempting to restore them in older age. Therefore, preventive strategies should be implemented along with treatment interventions and initiated as early as possible, before the onset of skeletal muscle mass, strength, and function loss (20).

6.1. Non-Pharmacologic Treatment

Exercise interventions and nutritional approaches play a significant role in management of sarcopenia.

Exercise is considered the primary treatment for sarcopenia as it can effectively increase both muscle strength and muscle mass (21).

Resistance exercise involves performing movements against a force or weight, while aerobic exercise involves rhytmic movements of large muscle groups for a spesific duration of time. Among various types of exercise, resistance exercise is recognized as the safest and most effective method to improve both muscle mass and muscle function. Robust evidence suggests that resistance training, such as weightlifting, promotes muscle protein synthesis, increases muscle mass, and enhances strenght, even in fail older adults (22).

6.1.2. Nutrition

Nutrition plays a significant role in the prevention, delay, or management of sarcopenia. Malnutrition contributes to poor muscle protein synthesis and is thus involved in the pathogenesis of sarcopenia.

Paying attention to energy balance is essential for maintaining healthy body composition. The recommended value for energy intake in older individuals is 30 kcal/kg/day. However, this value should be individually adjusted based on nutritional status, physical activity level, and underlying disease condition. Therefore energy intake should be in line with energy expenditure. Being overweight or obese can increase the risk of sarcopenia, while energy deficit (insufficient energy intake) can trigger muscle loss (23,24).

6.1.2.1. Protein

In the case of sarcopenia, adequate protein intake is essential for preserving muscle mass and supporting muscle function. Generally, protein requirements can vary from person to person and depend on age, gender, activity level, and overall health status. The recommended protein intake for individuals with sarcopenia is typically 1-1.2 g/kg/day for healthy older individuals. However, the recommendation may increase to 1.2-1.5 g/kg/day when older individuals have acute or chronic illnesses. In severe illness, injury, or malnutrition, the recommended protein intake may be higher, up to 2.0 g/kg/day (23,25).

6.1.2.2. Vitamin D

Vitamin D deficiency is a widespread health problem. Its impact on the musculoskeletal system has been extensively researched, demonstrating that this hormone can stimulate the proliferation and differentiation of skeletal muscle fibers, as well as maintain and enhance muscle strength and physical performance. Older individuals are at a higher risk of vitamin D deficiency due to factors such as inadequate nutrition, reduced sun exposure, and underlying

chronic conditions such as chronic kidney disease, malabsorbtion. Therefore, older individuals with vitamin D deficiency may be at risk of sarcopenia (26).

In clinical practice, vitamin D level is assessed by measuring the inactive form, 25-hydroxyvitamin D ((OH)D), in the blood serum. For adults, optimal levels are considered above 30 ng/mL, while it is suggested that levels above 40 ng/mL may be more appropriate for older individuals (27,28).

6.1.2.3. Omega 3

Polyunsaturated fatty acids (alpha-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHE) have anti-inflammatory properties. Some studies demonstrate their anti-inflammatory activity and ability to increase muscle mass, strength, and physical performance. EPA prevents muscle atrophy by enhanced protein synthesis, while DHA regulates the Ubiquitin-Proteaosome and Autophagy-Lysosome systems, thereby reducing proteolysis and inflammation (29,30,31). Addition omega-3 supplements combined with resistance training significantly enhance the benefits of exercise intervention. Although studies show the positive effects of PUFA treatments, there is currently no pharmacological intervention method with a clear underlying molecular mechanism to prevent or treat sarcopenia.

6.2. Therapeutic Intervention

6.2.1. Testosterone

The decrease in testosterone levels with advancing age is one-factor affecting muscle mass and strength reduction. While studies have shown improvements in lean tissue mass and muscle strength with testosterone supplementation in individuals with low testosterone levels (32), the current evidence does not provide conclusive assurance of the effectiveness of testosterone supplementation regimens in older adults with sarcopenia. Moreover, serious side effects can occur (such as fluid retention, gynecomastia, worsening of sleep apnea, polycythemia, prostate enlargement, and acceleration of prostate tumors) (33).

6.2.2. Anabolic Steroids and Selective Androgen Receptor Modulators (SARMs)

As an alternative to androgen replacement therapy, SARMs were created to provide a targeted therapeutic effect through the androgen receptors in different

tissues. While the binding of SARMS to the androgen receptor in the prostate and seminal vesicles is partially agonistic, with the receptor in muscle and bone is fully agonistic (34). This limits adverse effects such as prostate growth or androgenization. Although SARMs have shown benefits in body composition, such as increased muscle mass and decreased fat mass, the effects on muscle strength and function, as well as their long-term efficacy, are still not definitely known (35).

6.2.3. Growth hormone (GH)/ Insulin-like growth factor (IGF-1)

Growth hormone (GH) is essential for developing muscle and bone mass. GH exerts its anabolic effects by secretion of insulin-like growth factor 1 (IGF-1) from the liver, which is then released into the systemic circulation. IGF-1, in turn, promotes the production of contractile proteins in muscles by increasing satellite cell proliferation. GH also improves muscle mitochondrial functions, increasing oxidative enzymes and fatigue resistance (36). With aging, both GH and IGF-1 significantly decrease in pulsatile frequency and amplitude (37). While it has been hypothesized that the combination of GH replacement and exercise could synergistically improve muscle function, empirical studies have indicated that the supplementary benefits of GH replacement on top of exercise-induced improvements are constrained or negligible (38).

6.3. New Approaches in Treatment

Myostatin (MSTN) is a member of the transforming growth factor-beta (TGF-β) superfamily and is a potent negative regulator of muscle fiber growth and differentiation. Inhibition of the MSTN pathway has become an attractive therapeutic strategy for stimulating muscle growth and/or preventing muscle wasting in many muscle-wasting diseases, including sarcopenia. Therefore, strategies and studies for designing monoclonal antibodies against MSTN are currently being discussed.

It accurately states that Landogrozumab, a humanized monoclonal antibody targeting myostatin, has been shown to significantly increase muscle mass and partially improve muscle function and mobility in older individuals aged 75 years or over. Additionally, Trevogrumab, another monoclonal anti-myostatin antibody, has been found to effectively increase muscle mass and improve isometric force production in a study conducted on a mouse model. Although the phase II clinical trials of these studies have been completed., data evaluation is still underway. Also several other MSTN inhibitors have progressed to clinical trials for muscle-wasting diseases. (39).

7. Conclusion

Despite numerous studies and promising treatments, there are no pharmacological approaches that provide definitive evidence in the ability to prevent the decline in physical function and sarcopenia. Current and future pharmacological and clinical trials and epidemiological studies could radically change our therapeutic approach to understanding and treating mobility disability in elderly.

REFERENCES

1. Cruz-Jentoft AJ, Baeyens JP, Bauer JM, Boirie Y, Cederholm T, Landi F, et al. Sarcopenia: European consensus on definition and diagnosis: Report of the European Working Group on Sarcopenia in Older People. Age Ageing. 2010;39(4):412-23.

2. Cruz-Jentoft AJ, Bahat G, Bauer J, Boirie Y, Bruyere O, Cederholm T, et al. Sarcopenia: revised European consensus on definition and diagnosis. Age Ageing. 2019;48(1):16-31.

3. Grimby G, Saltin B. The ageing muscle. Clin Physiol. 1983;3(3):209-18.

4. Marzetti E, Lees HA, Wohlgemuth SE, Leeuwenburgh C. Sarcopenia of aging: underlying cellular mechanisms and protection by calorie restriction. Biofactors. 2009;35(1):28-35.

5. Salvioli S, Capri M, Valensin S, Tieri P, Monti D, Ottaviani E, et al. Inflamm-aging, cytokines and aging: state of the art, new hypotheses on the role of mitochondria and new perspectives from systems biology. Curr Pharm Des. 2006;12(24):3161-71.

6. Prior SJ, Ryan AS, Blumenthal JB, Watson JM, Katzel LI, Goldberg AP. Sarcopenia Is Associated With Lower Skeletal Muscle Capillarization and Exercise Capacity in Older Adults. J Gerontol A Biol Sci Med Sci. 2016;71(8):1096-101.

7. Walrand S, Guillet C, Salles J, Cano N, Boirie Y. Physiopathological mechanism of sarcopenia. Clin Geriatr Med. 2011;27(3):365-85.

8. Atkins J, Wannamathee SG. Sarcopenic obesity in ageing: cardiovascular outcomes and mortality. Br Nutr. 2020 Nov;124(10):1102-13.

9. Wannamathee SG, Atkins J. Muscle loss and obesity: the health implications of sarcopenia and sarcopenic obesity. Proc.Nutr.Soc. 2015;74: 405-412

10. J. Il Yoo, H. Kim, Y.C. Ha, H. Bin Kwon, K.H. Koo, Osteosarcopenia in patients with hip fracture is related with high mortality, J. Korean Med. Sci. 33 (2018)

11. Malmstrom TK, Morley JE. SARC-F: a simple questionnaire to rapidly diagnose sarcopenia. *J Am Med Dir Assoc*.2013 Aug;14(8):531–2.

12. Ibrahim K, May C, Patel HP et al. A feasibility study of implementing grip strength measurement into routine hospital practice (GRImP): study protocol. Pilot Feasibility Stud 2016; 2: 27.

13. Lauretani F, Russo CR, Bandinelli S, Bartali B, Cavazzini C, Di Iorio A, et al. Age-associated changes in skeletal muscles and their effect on mobility: an operational diagnosis of sarcopenia. J Appl Physiol. 2003;95(5):1851-60.

14. Houtkooper LB, Lohman TG, Going SB, Howell WH. Why bioelectrical impedance analysis should be used for estimating adiposity. Am J Clin Nutr. 1996;64(3 Suppl):436S-48S.

15. M. J. Delmonico, T. B. Harris, M. Visser et al., "Longitudinal study of muscle strength, quality, and adipose tissue infiltration," American Journal of Clinical Nutrition, vol. 90, no. 6, pp. 1579–1585, 2009

16. Correa-de-Araujo R, Addison O, Miljkovic I, et al. Myosteatosis in the context of skeletal muscle function deficit: An interdisciplinary workshop at the national institute on aging. *Front Physiol.* 2020;11:963.

17. Malafarina V, Uriz-Otano F, Iniesta R, Gil-Guerrero L. Sarcopenia in the elderly: diagnosis, physiopathology and treatment. Maturitas. 2012;71(2):109-14.

18. Rolland Y, Lauwers-Cances V, Cournot M, Nourhashémi F, Reynish W, Rivière D, et al. Sarcopenia, calf circumference, and physical function of elderly women: a cross-sectional study. J Am Geriatr Soc. 2003;51(8):1120-4.

19. Dent E, Morley JE, Cruz-Jentoft AJ, Arai H, Kritchevsky SB, Guralnik J, et al. International Clinical Practice Guidelines for Sarcopenia (ICFSR): Screening, diagnosis and management. J Nutr Health Aging 2018;22:1148-61.

20. Sayer AA, Robinson SM, Patel HP, Shavlakadze T, Cooper C, Grounds MD. New horizons in the pathogenesis, diagnosis and management of sarcopenia. Age Ageing 2013; 42: 1145–50.

21. Peterson MD, Rhea MR, Sen A, Gordon PM. Resistance exercise for muscular strength in older adults: a meta-analysis. Ageing Res Rev.2010; 9: 226–37.

22. Montero-Fernández N, Serra-Rexach JA. Role of exercise on sarcopenia in the elderly.European Journal of Physical and Rehabilitation Medicine, 01 Feb 2013, 49(1):131-143

23. Volkert D, Beck AM, Cederhom AL, et al. ESPEN guideline on clinical nutrition and hydration in geriatrics. Clin Nutr. 2019 Feb;38(1):10-47.

24. Gaillard C, Alix E, Salle A, Berrut G, Ritz P. Energy requirements in frail elderly people: a review of the literature. Clin Nutr 2007;26(1):16e24

25. Bauer J, Biolo G, Cederholm T, Cesari M, Cruz-Jentoft AJ, Morley JE, et al. Evidence-based recommendations for optimal dietary protein intake in older people: a position paper from the PROT-AGE study group. J Am Med Dir Assoc.2013;14:542–559

26. Remelli F, Vitali A, Zurlo A, Volpato S. Vitamin D Deficiency and Sarcopenia in Older Persons.Nutrients.2019 Dec; 11(12): 2861.

27. Bischoff-Ferrari H.A. Optimal serum 25-hydroxyvitamin D levels for multiple health outcomes. *Adv. Exp. Med. Biol.* 2014;810:500–525

28. Anagnostis P, Dimopoulou C, Karras S, Lambrinoudaki I, Goulis DG. Sarcopenia in post-menopausal women: Is there any role for vitamin D? Maturitas. 2015;82(1):56–64.

29. Frison E, Boirie Y, Peuchant E et al (2017) Plasma fatty acid biomarkers are associated with gait speed in community-dwelling older adults: the Three-City-Bordeaux study. Clin Nutr 36: 416–422.

30. Smith, G.I.; Julliand, S.; Reeds, D.N.; Sinacore, D.R.; Klein, S.; Mittendorfer, B. Fish oil-derived n-3 PUFA therapy increases muscle mass and function in healthy older adults. Am. J. Clin. Nutr. 2015, 102, 115–122.

31. JH Lee, JH Jeon, MJ Lee. Docosahexaenoic Acid, a Potential Treatment for Sarcopenia, Modulates the Ubiquitin–Proteasome and the Autophagy–Lysosome Systems. Nutrients, 12 (9) (2020), p. 2597

32. Isidori A M, Giannetta E, Greco E A, Gianfrilli D, Bonifacio V, Isidori A, Lenzit A, Fabbri A. Effects of testosterone on body composition, bone metabolism and serum lipid profile in middle-aged men: a meta-analysis. Clinical Endocrinology. 2005; 63, 280–93

33.Huang LT, Wang JH. The Therapeutic Intervention of Sex Steroid Hormones for Sarcopenia. Front Med (Lausenne) 2021; 8: 739251.

34. Fonseca GWPD, Dworatzek E, Ebner N, Von Haehling S. Selective androgen receptor modulators (SARMs) as pharmacological treatment for muscle wasting in ongoing clinical trials. Expert Opin Investig Drugs. (2020) 29:881– 91.

35. Papanicolaou DA, Ather SN, Zhu H, Zhou Y, Lutkiewicz J, Scott BB, et al. A phase IIA randomized, placebo-controlled clinical trial to study the efficacy and safety of the selective androgen receptor modulator (SARM), MK0773 in female participants with sarcopenia. J Nutr Health Aging. (2013) 17: 533–43

36. Goya RG, Brown OA, Bolognani F. The thymus-pituitary axis and its changes during aging. Neuroimmunomodulation. 1999;6(1-2):137-42.

37. Rudman D, Kutner MH, Rogers CM, Lubin MF, Fleming GA, Bain RP. Impaired growth hormone secretion in the adult population: relation to age and adiposity. J Clin Invest. 1981;67(5):1361-9.

38. Lange KH, Andersen JL, Beyer N, Isaksson F, Larsson B, Rasmussen MH, et al. GH administration changes myosin heavy chain isoforms in skeletal muscle but does not augment muscle strength or hypertrophy, either alone or combined with resistance exercise training in healthy elderly men. J Clin Endocrinol Metab. 2002;87(2):513-23.

39. Y Feike, L Zhijie, C Wei. Advances in research on pharmacotherapy of sarcopenia. Aging Medicine. 2021;00: 1-13

CHAPTER X

PROTEOMIX APPLICATIONS IN INFERTILITY*

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1. Introduction

Proteins, which are functional molecules in the organism, are encoded by genes. There are thousands of genes in the cell nucleus, proteins are formed as the products of these genes and each protein shows unique functions. In the last 20 years, gene structures in different organisms have been intensively studied and especially within the scope of the "Human Genome Project" initiated in 1990, the gene structures of human and many other organisms have been largely elucidated [1]. After the completion of the human genome project and the disclosure of the data, it has been accepted that genetic information alone cannot be sufficient unless the knowledge of proteins, which

* Part of this review was presented as a doctoral seminar

are the functional products of genes, increases. Proteins provide more precise information about the dynamic state of the cell, tissue or organism. Following the genome project, the number of genes in the human genome was calculated as 25000 +/- 5000. Relatively few of the proteins produced by these identified genes are known and this number is estimated to be around 200,000. If the 1990s can be called the age of genetics, the first decade of the new century can be called the age of proteomics. For this reason, following the Human Genome Project, studies on the Human Proteome Project, which covers protein identification and expression analyses of human genes, have also been initiated. For this purpose, the Human Proteome Organization (HUPO) in Canada initiated the Human Proteome Project in 2010 [1].

2. Proteom and Proteomics

The word 'proteome', which was first proposed and accepted by Mark Wilkins at the bidirectional electrophoresis meeting held in Siena, Italy in 1994, was defined as all proteins expressed by the genome of a cell, organ, or organism [1].

Protein + Genome = Proteome

'Proteomics', which can be defined as the analysis of the proteome by large-scale protein separation and identification techniques, is a dynamic term and includes quantitative and qualitative analyses of proteins in cells, tissues, organs or body fluids exposed to different conditions. The initial definition of this term was simply 'the whole of the studies carried out towards the identification and characterisation of all proteins synthesised by an organism or tissue'. For example, the term 'characterisation' included determination of protein function, positioning and post-translational modifications. Subsequent studies have shown that even this definition is insufficient and the investigation of environmental, pharmacological, genetic and pathological factors affecting the proteins within the scope of the subject has been included in the definition [1].

Proteome is the sum of all the different proteins that an organism possesses and expresses at a given time and condition. The word proteomics aims to identify each protein in a biological system under a certain condition and at a certain time, to reveal its gene expression profile, to determine its intracellular localisation, activity, control mechanisms (regulation), post-transcriptional regulation, molecular interactions, three-dimensional structure and function [2].

In recent years, although the function of many genes has been defined by genomic and functional genomic methods, researchers have focused on

proteins directly involved in these processes for biological processes that have not been fully elucidated. These developments have led to a systems biology approach in the post-genomic era, including proteomics, transcriptomics, metabolomics and mathematical modelling systems. Genome research, in other words the most detailed elucidation of an organism's genome, provides only limited information about the organism's true potential. Identification of genes by DNA sequence analysis does not show the rate at which the organism uses these genes and which ones it will respond to the situations it will encounter [3]. Post-genomic tools should be used to determine gene expression profiles, modifications of gene products, their location in the cell and their relative amounts. Transcriptional changes do not reflect changes at the protein level due to post translational modifications and differences in the stability of proteins. Moreover, a gene encodes different proteins with different biological functions and these proteins undergo post-translational modifications. In most cases, post translational modifications are directly related to the function of the protein and it is not possible to access this information with genomic/functional genomic approaches [4,5].

A protein, the basic unit of the proteome, is composed of amino acids and its active form in the cell is three-dimensional. Although the amino acid sequences of proteins are determined by the appropriate genes, genetic information does not provide all information about a protein. One gene one protein correlation is a concept that has lost its meaning today. Because unlike the one-dimensional genomic information determined by the combination of four nucleotides, the information encoded in proteins is not limited to amino acid sequences. Proteins are subjected to many regulations by cells. Regulations such as the formation of alternative forms of mRNA (splicing), fusions and post-translational modifications contribute to the tightly controlled dynamic structure, multidimensionality and variability of proteomics. This explains how a large number of unique protein molecules are formed [6]. The structure of the proteome depends on internal and external factors such as environmental factors, age, gender and diseases, physiological states (such as cell cycle, apoptosis). This situation reflects the opposite of the nature of the genome. The necessity of proteomics studies arises from the fact that mRNA expression levels cannot be correlated with protein expression levels, mRNA levels do not reflect the activity of the encoded protein, information on post-translational modifications of proteins cannot be obtained at the mRNA level, and therefore the genome and proteome cannot provide complementary data [6].

Before proteomics, the studies mainly shed light on the changes at the RNA level under the name of transcriptomics. However, changes at the RNA level are not seen exactly at the protein level. In a recent study (2009), both RNA and protein expression levels were analysed in 23 different human cell lines. It was observed that protein expression levels matched RNA expression levels only 33% of the time. Therefore, the best approach is to follow the changes in protein expression rather than determining the changes in RNA expression levels [7].

2.1. Clinical Proteomics

Proteomic methods are mostly used in the field of medicine, also called 'clinical proteomics'. Proteomics is a science that requires multidisciplinary technology that works on the determination of proteins and their interactions involved in the response to many stimuli such as specific diseases, toxic agents or infections.

Today, proteomics is widely used in the understanding of disease pathogenesis, especially in the discovery of disease-related biomarkers and personalised treatment approaches. In addition to genetic techniques, which are widely used in the identification of diseases and treatment development, proteomics techniques have been a great hope for researchers [1,8-9].

The most important advantages of proteomics studies in addition to gene studies are the identification of diagnostic and prognostic disease markers in body fluids, cells and tissue biopsies and the identification of new treatment strategies. In addition, the identification of genetically predisposed diseases requires accurate detection of the phenotype as well as an accurate genetic test. This is because the proteome in an organism is more dynamic than the genome and can change depending on the condition of the organism (e.g. pathological condition) and time. Currently, it is not known where and under what conditions many molecules are synthesised in the cell and the roles of protein molecules that reflect the whole natural reality cannot be explained [9,10].

Drug design and development have recently gained importance in proteome applications commercially.

Proteomics has been applied in many diseases such as Alzheimer's, Parkinson's, Huntington's, Down's syndromes and cancer. The development of cancer and tumour markers is one of the most studied topics. So far, bladder, kidney, prostate, leukaemia, pancreas, stomach, lung, colon, thyroid, brain and

hepatocellular carcinoma have been widely studied. Significant progress has been made in the identification of disease-specific proteins. In addition, the proteomes of microorganisms involved in some diseases have also been studied [11].

Recently, proteomic applications have taken place in the field of infertility, which is an important health problem.

3. Infertility

Infertility is the inability to achieve pregnancy. Infertility affects 10-15 per cent of couples of reproductive age. Although the causes of infertility vary between societies and age groups, it can be thought that ovulation disorders are responsible for approximately 20-25% of infertile couples, tubal problems in 20-30% and male factor in 40%. More than one cause is present in 40% of couples and no cause can be identified in 15-25% of couples.

The probability of a normal young couple achieving pregnancy at the end of 1 month is 25 per cent, 70 per cent at 6 months and 90 per cent at the end of the first year. Approximately 50-80 million people in the world are thought to be infertile. In Turkey, this rate is estimated as 1.5-2 million according to the sources.

As can be seen, infertility can occur due to reasons related to only the woman or the man or both [12]. Infertility researches are aimed at both spouses.

Research in the field of molecular biology of male and female reproductive functions has the advantage of helping us to understand the causes, normal and pathological complexities of infertility. To understand these biological complexities in humans, it is necessary to carefully define the physiological functions of each protein in tissues.

Researchers working on reproduction identify problems and pathologies related to infertility through proteomics. It reveals the profiles of various tissues important for reproductive functions (e.g. testis, ovary, placenta, epididymis and endometrium) and allows the specific functions of tissues to be determined. It also contributes to our understanding of the pathology of various reproductive diseases, such as azoospermia (no sperm in semen), oligozoospermia (low sperm count) in men and endometriosis (presence of endometrial cells outside the uterus) and polycystic ovary syndrome (a disease characterised by the formation of many small benign cysts in the ovaries) in women [13].

4. The Role of Proteomics in Male Infertility

4.1. Sperm Proteomics

With the development of proteomics techniques, researchers have performed proteomics comparisons of sperm samples of different quality to identify new biomarkers in male infertility.

Among these studies, the pilot study was conducted by Thacker et al. Thacker et al. compared sperm proteins in 1 fertile and 3 infertile men by proteomics analyses and identified 4 specific proteins (semenogelin II precursor, prolactin-induced protein, clusterin isoform 1 and prostate-specific antigen isoform 1 preproprotein) in fertile men. However, they found that two of these 4 specific proteins (semenogelin II precursor and clusterin isoform 1) were not found in infertile men [15].

In another study, Zhao et al. (2007) compared the sperm protein profiles of infertile men with asthenozoospermia with the sperm proteins of fertile normozoospermia men and determined the expression of 10 different proteins in the asthenozoospermia group [16].

Similarly, Martinez et al. (2008) compared infertile men with asthenozoospermia and fertile men and found 10 upregulated (increased expression) and 7 downregulated (decreased expression) proteins in the asthenozoospermia group [17].

Until 2014, a total of 109 differentially expressed proteins were identified in infertile patients with asthenospermia compared to fertile patients in different comparative studies [18].

4.2. Seminal Plasma Proteomics

Human seminal plasma ranges between 35-55 g/l. The protein content of seminal plasma is potentially of great importance in determining the fertilisation capacity of sperm.

Proteomics may be potential markers for spermatogenesis in spermatogenesis in which seminal plasma proteins are impaired. With the use of proteomics, proteins in seminal plasma can be identified, thus facilitating the identification of new proteins related to spermatogenesis and sperm function [19].

Ayyagari et al. (2007) determined the presence of a group of proteins in seminal plasma of infertile men with oligospermia, which are not found in normal spermatozoa and azoospermia [20].

In another study, Wang et al. (2009) compared the seminal plasma protein profiles of the asthenospermic infertile group with the control group with normal sperm and found that 45 proteins were 3-fold down-regulated (decreased expression (release)) and 56 proteins were 3-fold up-regulated (increased expression) in the asthenospermic infertile group [21].

Sharma et al. (2013) identified 7 different seminal proteins in men with abnormal sperm quantity and/or morphology compared to men with normal sperm [22].

4.3. DNA breaks / Oxidative Stress Proteomics

The association of DNA breaks with infertility has also been investigated by proteomics analyses. De Mateo et al. (2007) identified 8 different proteins associated with DNA breaks in infertile men compared to fertile men [23].

Instasqui et. al. (2013) determined that 23 proteins were highly expressed in males with high DNA fragmentation rate and 71 proteins were highly expressed in males with low DNA fragmentation [24].

Proteomics analyses were also used to determine the relationship between oxidative stress and infertility. Sharma et. al. (2013) compared the expression of proteins in human sperm with low and high ROS (reactive oxygen species) levels. As a result, they determined that 10 of the proteins were overproduced and 5 of them were underproduced in sperm with high ROS level. As a result, it was suggested that these proteins may be marker proteins in sperm in oxidative stress production [25].

4.4. In Vitro Fertilization (IVF) Proteomics

One area of infertility research using proteomics techniques has been failed In Vitro Fertilization (IVF; the process of fertilisation of an egg by sperm under laboratory conditions) trials. For example, Pixton et al. (2004) compared men with fertilisation failure in IVF treatment with fertile men and found that 20 proteins were expressed at different levels in infertile men compared to fertile men [26].

A similar study was conducted by Fraqsauce et al. (2009) and when they compared the sperm proteins of men who failed IVF treatment and men who succeeded in treatment, they identified 14 different proteins in the group that failed IVF treatment [27].

Zhu et. al. (2014) compared the protein profiles of sperm samples of 6 men who achieved clinical pregnancy with Assisted Reproductive Techniques (ART)

and 6 men who failed to achieve clinical pregnancy and found that 21 different proteins were expressed in the group with clinical pregnancy compared to the group with pregnancy failure [28].

4.5. Testicular Tissue Proteomics

The testis is the organ where sperm, the male reproductive cell, is produced. Recent 2DE (2-dimensional polyacrylamide gel electrophoresis) studies have identified a protein source map containing 1908 proteins in the testis [34]. In addition, the combined variant and phosphorylated forms of proteins have been reported, allowing us to understand the underlying heterogeneity and regulatory mechanism of human testicular proteins [29].

In studies on testicular tissue, 10 different protein expressions have been shown in normal and pathological testicular tissue samples in relation to spermatogenesis. Four of them are phospholipid hydroperoxide glutathione peroxidase (GPX4), peroxiredoxin 4 (Prx4), heat shock protein beta-1 (HSP 27), cathepsin D (CTSD) proteins [30]. These proteins have been reported to play important roles in various events in the spermatogenetic process such as spermiation and movement of the male germ cell. If any of these proteins are identified as molecular markers, a new dimension will be added to the perspectives on the regulation of fertility and birth control methods [30].

5. The Role of Proteomics in Female Infertility

5.1. Endometrium and Uterine Proteomics

The endometrium is a membrane covering the lining of the uterus. With the sequential action of various steroid hormones, it fulfils the functions necessary for the initiation and maintenance of pregnancy.

The molecular content of the receptive endometrium is a strong subject of research in order to discover the underlying causes of the low success rate of IVF (in vitro fertilisation) and Embryo Transfer (ET) used as infertility treatment in women [14].

Before the genomic era, research on endometrial receptivity was carried out with hypotheses, specific probes and limited experimental tools. However, with the advent of proteomics techniques, it has been possible to analyse the endometrial profile with protein levels at different stages of the menstrual cycle [14].

Dominguez et al. (2009) found two differentially regulated proteins (Annexin A-2 and Stathmin-1) when they compared day 2 (non-receptive) or

day 7 (receptive) of menstrual cycles of fertile women after luteinising hormone (LH) secretion [31].

In addition to endometrial tissue, uterine fluid or samples aspirated from the endometrium can also be analysed using proteomics tools at different stages of the menstrual cycle. Proteomics studies using mass spectrometry have shown that proteins found in the uterine luminal fluid of fertile women are altered compared to those found in infertile women. In this way, mapping the endometrial endometrium during the receptive phase of the menstrual cycle will help us to understand the causes of implantation failures in the natural cycle and the causes of low success in pregnancies attempted to be achieved using assisted reproductive techniques [32].

5.2. Endometriosis Proteomics

Endometriosis is an increasingly worrying disease that affects between 2-48% of women of reproductive age. EOS is a chronic disease caused by the presence of endometrial tissue cells outside the uterus and causes dysmenorrhoea, chronic pelvic pain and infertility [33].

Zhang et. al. (2006) compared the protein expressions of eutopic endometrium of women with and without endometriosis and found 13 and 11 different proteins in serum and endometrium, respectively [34].

In endometriosis (EOS) research, chaperones such as secretory proteins (e.g. apoliprotein A2) and redox regulators (e.g. peroxiredoxin-2) have been identified using 2-DE techniques including DIGE. These proteins are involved in DNA metabolism and catabolism in the eutopic endometrium of women with and without EOS. These proteins have a wide range of functions including regulation of cell growth, signalling and adhesion, and any defect in their expression can have a significant impact on the behaviour of endometrial cells [35].

In all these studies, it has been proven that there are intrinsic differences between women with endometriosis and eutopic endometrium and normal control group women [36, 37].

5.3. Ovary Proteomics

Research on the protein profile in the ovary has become a focal point for diseases such as PCOS (polycystic ovary syndrome) and ovarian cancer.

5.4. Polycystic Ovary Syndrome (PCOS)

Infertility is due to ovulation irregularity. It may be the result of abnormalities in women's ovaries or other hormonal causes. This disorder is

characterised by hyperandrogenism, insulin resistance, obesity, elevated LH hormone in serum and polycystic ovaries that can be measured on ultrasound.

Abdominal adiposity and consequently obesity is observed in 50% of patients with PCOS. This metabolic syndrome leads to a variety of pathological conditions including cardiovascular disease and diabetes [36]. Pcos is often difficult to diagnose due to the diversity in the clinical picture of the disease. Proteomics technologies are used to identify therapeutic targets and markers for Pcos.

Proteomics studies on Pcos have been conducted with different body fluids, ovarian tissue, serum, plasma, visceral adipose tissue and ovarian granulosa cells [36].

Carton et al. (2007) identified differential expression of 15 proteins between normal and Pcos patients [37]. Ma et al. (2007) investigated protein expression between normal and Pcos ovaries and found that 69 of the 110 proteins identified were differentially expressed between the groups [38].

Dai and Lu (2012) identified 20 different protein expressions in patients with Pcos when they compared normal patients with Pcos. These proteins involve glucose and lipoprotein mechanisms, cell proliferation, apoptosis and insulin resistance [39].

In another study by Gupta et al (2014), investigations were performed with different test tools and as a result, different protein expressions were detected in patients with PCOS and normal patients [40].

5.5. Oocyte Proteomics

The oocyte proteome represents factors that directly determine the phenotype of the oocyte. The protein structure of the oocyte is crucial for fertilisation and pregnancy outcome.

Meng et al. (2007) investigated the mature mouse coocyte- oocyte complex (CoCs) and as a result, they identified 259 proteins in total, 156 of which were unique [41]. The biological classification of these proteins is as follows: 12% in cell signalling-communication, 7% in cell division, 31% in gene/protein expression, 24% in cell metabolism, 10% in cell structure and motility, 12% in cell/organism defence and 4% are unknown.

In another study, Katz-Jaffe et al. (2005) determined that mature (MII) mouse oocytes contain 3699 proteins. It was stated that 28 of these proteins were present in the proteomes of undifferentiated mouse stem cells [42].

Again, these proteins are associated with nuclear reprogramming of RNA, lipids, small molecules and proteins. Therefore, oocyte proteomics will provide

us with very important information about oocyte maturation, fertilisation and embryo development in humans and animals.

5.6. Embryo Proteomics

The zygote is a very unique combination of two sex cells, the sperm and the oocyte, and it is the totipotent cell with the most competent differentiation capacity. As a result of division and differentiation, an embryo is formed. The embryo, in turn, undergoes further growth and differentiation to form the organism. The cellular compositions of embryo development express the transcriptome and proteome of the embryo [14].

With the advances in methods such as mass spectrometry and SELDI-TOF MS (Surface enhanced laser desorption/ionisation mass spectrometry), pre-implantation studies have been conducted with a limited number of human and mouse embryos, especially blastocysts [42].

Katz-Jaffe et. al. (2006) analysed the proteomics of early, enlarged and degenerated blastocysts using SELDI-TOF/ MS technique. When they compared early and enlarged blastocysts, they detected two negatively charged proteins/ biomarkers of approximately 5 kDa and 6.3 kDa with increased expression in the enlarged blastocyst and two negatively charged proteins/ biomarkers of approximately 3.8 kDa with decreased expression in early and enlarged blastocysts [50]. According to this study, all human blastocysts with similar morphology have the same protein profiles. In addition, protein expression profiles were analysed according to blastocyst developmental stages, and it was found that negatively charged up- and down-regulated proteins were different when enlarged and early blastocysts were compared [43].

Proteome analyses of embryos at different developmental stages provide insight into the molecular mechanisms underlying embryo development, which can help to identify markers that are specific for the embryo stage. These markers can help to assess embryo quality and viability. As a result, clinical IVF with limited number of embryo transfers may lead to an increase in pregnancy/ live birth rates [43].

Since proteomics analysis requires cell lysis, it is not possible to apply it to embryos before transfer. Therefore, proteins secreted into IVF culture media (secretomics) have been analysed. In this way, the secretory profile of any given embryo can be correlated with its reproductive success rate. For example, by analysing the profiles of embryos that successfully implant or fail to implant, biomarkers for the optimum embryo to be transferred can be found. Even the

genetic profiles of the embryos to be transferred can be analysed non-invasively (without physical damage).

As shown by McReynolds et al. (2011), lipocalin-1 is a secretome of blastocysts and is associated with aneuploidies (44).

Katz-Jaffe et al (2006) identified different secretomic profiles at different developmental stages of the embryo and found ubiquitin as a protein biomarker associated with blastocyst development [43].

Advances in proteomics have deepened our understanding of the growth environment and secretions of normal versus abnormal embryos. The results of these studies are expected to improve the selection of the 'best' embryo for transfer and increase the success rate of IVF.

Increased precision in embryo selection will result in the transfer of fewer embryos with high implantation potential, thereby increasing both maternal and foetal treatment safety [14, 42].

5. Conclusion

Without proteomics, DNA sequence data alone would not answer any question for expressing protein levels. Proteomics requires a comprehensive study of proteins in structural and functional aspects. The information obtained will form an important database.

Proteomics can provide new avenues in reproductive biology for expected reproductive outcomes in general and with new targets. It is certain that proteomics approaches will help to develop new tools in the understanding and diagnosis of infertility.

Proteomics has provided a better understanding of protein functions in infertility, from the normal state to the abnormal state. With large-scale proteomics studies, new biomarkers for many proteins can be developed. Protein biomarkers may also be helpful for better diagnosis and drug development for infertility causes. With the help of these, fertility diagnosis can be made.

The place and importance of proteomics studies in infertility continues to increase day by day.

REFERENCES

1. Demiralp D, İğci N, Peker S, Ayhan B. Basic Proteomic Strategies. Türkiye: Ankara Unv. Biotechn. Enst. Publications; 2013

2. Maeda K, Hägglund P, Finnie C, Svensson B. Proteomics of Disulphide and Cysteine Oxidoreduction. Oxford: Blackwell Publishing Ltd; 2006.

3. Kav V, Srivastava S, Yajima W, Sharma N. Application of proteomics to investigate plant-microbe interactions. Curr. Proteomics. 2007; 4: 28-43.

4. Jansen C, Nap P, Mlynarova L. Errors in genomics and proteomics, Nat Biotechnol. 2002; 20: 19.

5. Başaran E, Aras S, Cansaran-Duman D. Overview of genomics, proteomics, metabolomics concepts and application areas. Turkish Journal of Hygiene and Experimental Biology. 2010; 67(2): 85-96.

6. Rappsilber M. What Does it Mean to Identify a Protein in Proteomics. Trends Biochem. Sci. Feb. 2002;1:27(2): 74-78.

7. Gry M, Rimini R, Strömberg S, et.al. Correlations between RNA and protein expression profiles in 23 human cell lines. BMC Genomics. 2009; 10: 365.

8. Makro-Varga G. Proteomics Principles and Challenges. Pure Appl. Chem. 2004; 76(4): 829-837.

9. Liu Y, Yang L, Chen J, et.al. Comparative proteomics and correlated signalling network of rat hippocampus in the pilocarpine model of temporal lobe epilepsy. Proteomics. 2008; 8(3): 582-603.

10. Giorgianni B. Proteome Analysis by Two-Dimensional Gel Electrophoresis and Mass Spectrometry: Strengths and Limitations. Trends in Analytical Chemistry 2003; 22(5): 273-281.

11. Zabel C. Proteins Are Specifically Altered in Disease--Really? Expert Rev Proteomics. 2006; 3(5): 469-70.

12. Esenler I. Reproductive Endrocrinology. Türkiye: Güneş Kitabevi; 2006.

13. Çiçek N. Gynaecological Endocrinology and Infertility Handbook. Türkiye: Modern Medicine Publications; 2013.

14. Upadhyay D, Balasinor H, Kumar V, Sachdeva G, Parte P, Dumasia K. Proteomics in reproductive biology: Beacon for unraveling the molecular complexities. Bioch. et Biophysica Acta. 2013; 1834: 8-15. Thacker S, Yadav P, Sharma K, et al. Evaluation of sperm proteins in infertile men: a proteomic approach. Fertil. Steril. 2011; 95: 2745-2748.

16. Zhao C, Huo R, Wang Q, et al. Identification of several proteins involved in regulation of sperm motility by proteomic analysis. Fertil. Steril. 2007; 87: 436-438.

17. Martınez-Heredia J, de Mateo S, Vidal-Taboada M, et al. Identification of proteomic differences in asthenozoospermic sperm samples. Hum. Reprod. 2008; 23: 783-791.

18. Amaral A, Castillo J, Ramalho-Santos J, et al. The combined human sperm proteome: cellular pathways and implications for basic and clinical science. Hum. Reprod. Update. 2014; 20: 40-62.

19. Camargo M, Intasqui P, Del Giudice T, et.al. Human Reproduction. 2013; 28(1): 33-46.

20. Ayyagari R, Fazleabas T, Dawood Y. Seminal plasma proteins of fertile and infertile men analysed by two-dimensional electrophoresis. Am. J. Obstet. Gynecol. 2005; 157: 1528-1533.

21. Wang J, Wang J, Zhang R, et al. Proteomic analysis of seminal plasma from asthenozoospermia patients reveals proteins that affect oxidative stress responses and semen quality. Asian. J. Androl. 2009; 11: 484-491.

22. Sharma R, Agarwal A, Mohanty G, et al. Functional proteomic analysis of seminal plasma proteins in men with various semen parameters. Reprod. Biol. Endocrinol 2013; 11: 38.

23. De Mateo S, Martınez-Heredia J, Estanyol M, et al. Marked correlations in protein expression identified by proteomic analysis of human spermatozoa. Proteomics. 2007; 7: 4264-4277.

24. Intasqui P, Camargo M, Del Giudice T, et al. Unraveling the sperm proteome and post-genomic pathways associated with sperm nuclear DNA fragmentation. J. Assist. Reprod. Genet. 2013; 30: 1187- 1202.

25. Sharma R, Agarwal A, Mohanty G, et al. Proteomic analysis of human spermatozoa proteins with oxidative stress. Reprod. Biol. Endocrinol. 2013; 11: 48.

26. Pixton L, Deeks D, Flesch M, et al. Sperm proteome mapping of a patient who experienced failed fertilisation at IVF reveals altered expression of at least 20 proteins compared with fertile donors: case report. Hum. Reprod. 2004; 19: 1438-1447.

27. Frapsauce C, Pionneau C, Bouley J, et al. Unexpected in vitro fertilisation failure in patients with normal sperm: a proteomic analysis. Gynecol. Obstet. Fertil. 2009; 37: 796-802.

28. Zhu Y, Wu Y, Jin K, et al. Differential proteomic profiling in human spermatozoa that did or did not result in pregnancy via IVF and AID [e-pub ahead of print], Proteomics Clin Appl. doi: 10.1002/prca.201200078, 2014.

29. Guo,X, Zhao C, Wang F, et.al. Investigation of human testis protein heterogeneity using 2-dimensional electrophoresis. J. Androl. 2010; 31: 419-429.

30. Huo R, He Y, Zhao C, Guo J, Lin M, Sha H, et.al. Identification of human spermatogenesis-related proteins by comparative proteomic analysis: a preliminary study. Fertil. Steril. 2008; 90: 1109-1118.

31. Domínguez F, Garrido-Gómez T, López A, et.al. Proteomic analysis of the human receptive versus non-receptive endometrium using differential in-gel electrophoresis and MALDI-MS unveils stathmin 1 and annexin A2 as differentially regulated. Hum. Reprod. 2009; 24: 2607-2617.

32. Spencer E. Biological roles of uterine glands in pregnancy. Semin. Reprod. Med. 2014; 32(5): 346- 57.

33. Have T, Fraser I, Markham R, Lam A, Matsumoto I. Proteomic analysis of protein expression in the eutopic endometrium of women with endometriosis. Proteomics. Clin. Appl. 2007; 1: 1243-1251.

34. Zhang H, Niu Y, Feng J, Guo H, Ye X, Cui H. Use of proteomic analysis of endometriosis to identify different protein expression in patients with endometriosis versus normal controls. Fertil. Steril. 2006; 86: 274-282.

35. Scotchie G, Fritz A, Mocanu M, Lessey A, Young L. Proteomic analysis of the luteal endometrial secretome. Reprod. Sci. 2009; 16: 883-893.

36. Dunaif A. Insulin resistance and the polycystic ovary syndrome: mechanism and implications for pathogenesis. Endocr. Rev. 1997; 18: 774-800.

37. Corton M, Botella-Carretero I, Benguria A, et.al. Differential gene expression profile in omental adipose tissue in women with polycystic ovary syndrome. J. Clin. Endocrinol. Metab. 2007; 92: 328-337.

38. Ma X, Fan L, Meng Y, et.al. Proteomic analysis of human ovaries from normal and polycystic ovarian syndrome. Mol. Hum. Reprod. 2007; 13: 527-535.

39. Dai G, Lu G. Different protein expression patterns associated with polycystic ovary syndrome in human follicular fluid during controlled ovarian hyperstimulation. Reproduction, Fertility and Development. 2012; 24 (7): 893-904.

40. Gupta S, Ghulmiyyah J, Sharma R, Halabi J, Agarwal A. Power of Proteomics in Linking Oxidative Stress and Female Infertility. Hindawi Publishing Corporation BioMed Research International. 2014. Article ID 916212, 26.

41. Meng Y, Xiao-hui L, Shen Y, et.al. The protein profile of Mouse mature cumulus-oocyte complex. Biochim. Biophys, Acta. 2007; 1774: 1477-1490.

42. Katz-Jaffe G, Linck W, Schoolcraft B, Gardner K. A proteomic analysis of mammalian preimplantation embryonic development. Reproduction. 2005; 130: 899-905.

43. Katz-Jaffe G, Schoolcraft B., Gardner K. Analysis of protein expression (secretome) by human and mouse preimplantation embryos. Fertil. Steril. 2006; 86: 678-685.

44. McReynolds S, Vanderlinden L, Stveens J, Hansen K, Schoolcraft B, Katz-Jaffe G. Lipocalin-1: a potential marker for noninvasive aneuploidy screening. Fertility and Sterility. 2011; 95 (8): 2631-2633.

CHAPTER XI

LEADERSHIP IN THE HEALTHCARE FIELD

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1. Introduction

Leadership is an important concept in management that helps maximize efficiency and achieve organizational goals. Leaders are significant figures who communicate policies and plans to subordinates, clearly explain tasks, and provide effective guidance to reach objectives (36).

Leadership has been defined in various ways by numerous foreign and domestic researchers. Bennis and Nanus (9) defined leadership as influencing and directing, where it involves influencing opinions, actions, and tendencies. Similarly, Bass and Stogdill (8) defined leadership as facilitating interactions among group members (17). The term "leader" refers to someone with power and influence in management, as well as a guide or chief. Another definition describes a leader as a person responsible for the top-level management of a party or organization. In the dictionary, leadership can also refer to being in the forefront in a competition or being part of the leading team (57).

Eren (22) defines leadership as the combination of gathering a group of people around specific goals and motivating them to take action to achieve these goals, while Koçel (36) describes leadership as the process of influencing and directing the activities of others to achieve specific personal or group goals under certain conditions. Leadership theories often address the characteristics of leaders, their behaviors, and the environmental conditions in which leadership is

discussed and debated (16). The prominence of topics such as managers/leaders and organizational behavior has led to the need for different types of leadership and hindered a singular understanding of the concept of leadership. This need has necessitated the development of unique understandings of leadership in various fields (sociology, medicine, nursing, psychology, political science, management science) and has led to the emergence of different leadership theories. Researchers such as Akçakaya (1), Komives and Dugan (37), Nikezić et al. (46), and Scott (51) have emphasized this need. Additionally, other researchers such as Bass (8), Rost (48), Astin et al. (3), Eraslan (21), Avolio et al. (7), Tabak et al. (56), and Posner (47) have developed different leadership theories to address the specific requirements of their fields.

In this context, the concept of leadership has been approached from an interdisciplinary perspective and acquired different meanings in each field. In organizations, leadership is considered as a process that enables managers to make effective decisions, guide employees, and achieve organizational goals. In the healthcare field, leadership plays a significant role in the management and coordination of healthcare professionals to enhance the quality and effectiveness of healthcare services. The concept of leadership carries different meanings in various fields and disciplines. Different leadership theories have been developed to cater to the needs of each field and customize the universal principles of leadership. Therefore, adopting an interdisciplinary approach is crucial in leadership research.

In this context, theories and approaches related to leadership are classified in different ways. These classifications are often made as follows:

- Approaches focusing on the characteristics of leaders (17).
- Approaches examining leader behaviors (1950s-1960s).
- Situational approaches (early 1950s-1980s).
- Modern leadership approaches (mid-1920s, from 1978 to the present) (38).

Leadership is the ability to identify opportunities that others may not see and utilize these opportunities for the benefit of the organization. Leaders influence others within a specific vision framework, motivating them to work willingly (2). When examining definitions related to leadership in general, concepts such as influence, vision, mobilization, process, guidance, and mission are seen as common elements (64). If a manager is unable to influence the individuals they work with, certain problems within the organization are inevitable.

Leadership behaviors displayed by managers under the conditions they are in are referred to as leadership styles. The personal characteristics of managers, the nature of the target audience, the leadership power possessed by the leader, and the position of the leader within the organization all influence the emergence of leadership styles (Yılmaz & Kantek, 2016). Considering the concepts of power and influence, two types of leadership become apparent. A formal leader influences the group through their authority, while an informal leader integrates with the group they lead through their authority. As these statements indicate, leadership is based more on personality traits than on status. Therefore, the leader's behaviors have an impact on the development of organizations. This is because a leader is necessary for the existence of an organization and should be an effective element for the continuity of the organization (64). However, leadership approaches will vary depending on the conditions in which the organization is located and the organizational and management theories. In this context, leadership style will influence the perception of the leader's effectiveness by shaping the culture of the institution and being perceived by other individuals and organizations.

The concept of leadership can be defined in different ways in various fields and contexts. For example, in the business field, leadership is seen as an ability that is effective in achieving an organization's goals. Yukl (66) defined leadership as the "process of influencing the behaviors of a group." However, it can also be said that leadership plays an important role in the political field. Avolio and Gardner (5) defined political leadership as the "process of making and implementing decisions that affect society."

In the field of education, leadership is a factor that influences relationships between schools, teachers, and students. Leithwood et al. (41) described educational leadership as a "process that promotes transformation in education." In the sports field, leadership is a factor that influences teams' performance and directs them toward success. Chelladurai (13) defined sports leadership as a "behavioral process that is effective in helping a team achieve its goals." In the healthcare field, leadership plays a significant role in the management of healthcare services and the effective coordination of healthcare professionals. Wong and Cummings (61) defined health leadership as a "process that promotes collaboration and innovation in healthcare systems and among healthcare professionals." These examples demonstrate that leadership operates in different fields based on similar principles. Leadership is a universal concept that involves influencing a group of people, achieving goals, and promoting change.

2. Leadership in the Healthcare Field

The rapid changes in healthcare services, efforts to build an advanced society, the use of complex technologies, increasing competition, and the involvement of intense human relationships are significant factors that require managers in healthcare institutions to exhibit leadership behaviors. The concept of leadership is defined as the knowledge and ability to bring a group of people together around specific goals and mobilize them towards those goals (65).

Management in the healthcare sector is achieved through the effective management of healthcare institutions of various sizes and complexities. These institutions take on the task of preserving and improving people's health, thus playing an important role in determining health policies (12). The increasing share of the service sector globally and in the Turkish economy, the extension of life expectancy, and the continuous technological development and economic growth within the healthcare sector increase the importance of the quality of healthcare services provided (63). Hospitals require the presence of effective, creative, visionary, motivating, knowledgeable, and principled leaders for continuous improvement. Therefore, it is important to identify the qualities that can contribute to the development of effective leadership skills for hospital managers (34).

In today's healthcare system, significant changes are taking place with the goals of increasing cost-effectiveness, improving quality, and ensuring accessibility. Therefore, healthcare service organizations acknowledge the necessity of an effective management approach in the face of changing consumer demands and the work environment. The rapid change and development in the healthcare sector increase the importance of developing managerial and leadership skills for healthcare managers. Managers and leaders need to integrate management and leadership to maximize their potential (44). Gardner (24) identified some characteristics that express integrated manager-leader qualities, including long-term thinking, having a broad perspective, influencing others, emphasizing vision, values, and motivation, being politically competent, and focusing on change and innovation.

Current studies in the healthcare field are evaluating the effects of transformational and transactional leadership concepts in healthcare institutions. For example, Lombardi (43) demonstrated the positive effects of transformational leadership on employee performance, job satisfaction, and service quality in healthcare organizations. Similarly, Gedikoğlu (27) emphasized the significance of transformational leadership as a prominent leadership style in the healthcare field. Particularly during periods of rapid

social change and in crisis environments, transformational leadership has been found to be effective. Positive leadership behaviors exhibited by managers in healthcare institutions have positive effects on employee performance, job satisfaction, and service quality. Transformational and transactional leadership are among the prominent leadership types in healthcare institutions and play an important role in improving the quality of healthcare services.

Research conducted on leadership in the healthcare sector emphasizes the importance of leadership skills for healthcare service managers. These studies serve as a fundamental resource for applying leadership theories and approaches to leadership effectiveness in the healthcare sector and improving the quality of healthcare services (29). Particularly, it is emphasized that healthcare leaders need to possess skills such as motivation, communication, teamwork, and adaptability to change (33).

Leadership in the healthcare sector is a process where healthcare managers aim to influence other healthcare professionals to achieve specific goals. This leadership process plays a significant role in providing care and treatment to both patients and healthy individuals. Clinical leadership, on the other hand, refers to the efforts of clinicians working in a specific department or unit of a healthcare institution to provide direct care to patients and strive to improve the quality of this service, as well as having the power and ability to change policies, procedures, and systems through these efforts (11).

Smith et al. (53) examined how leadership skills affect performance in healthcare services. Their studies demonstrate that the skills of healthcare leaders, such as communication, vision building, and motivation, enhance employee satisfaction and improve service quality. Additionally, Johnson et al. (32) emphasize the importance of transformational leadership in the healthcare sector. Transformational leadership focuses on leaders' abilities to motivate team members, create a vision, and encourage innovative thinking. This study reveals that transformational leadership enhances employee commitment and improves their performance in healthcare services. Furthermore, Walker and Avolio (59) highlight the significance of ethical leadership skills among healthcare leaders. Ethical leadership involves making value-based decisions, embracing ethical values like honesty and fairness. The study shows that the ethical leadership skills of healthcare leaders increase employee morale and positively impact organizational performance.

In today's healthcare institutions, leaders with the ability to demonstrate leadership behaviors are expected due to factors such as competition and

privatization in the healthcare field. The leadership qualities of healthcare managers have a significant impact on individual and organizational success. These qualities require managers to enhance their ability to effectively lead and develop leadership behaviors (19).

In this context, interactional leadership and transformative leadership concepts play a significant role in healthcare studies and have been shown to be effective in improving quality in healthcare institutions (43).

It is known that positive leadership behaviors exhibited by managers in healthcare institutions have positive effects on employees' performance, job satisfaction, and service quality (43). The managerial processes and policies implemented by managers along with successful leadership behaviors bring a new perspective to the healthcare environment, improve quality in healthcare institutions, and create beneficial environments for both patients and employees. In this context, various types of leadership can be considered. A prominent leadership type in studies related to healthcare leadership is transformative leadership. Especially during periods of rapid change in society and increasing socio-economic problems in crisis environments, transformative leadership is considered the most effective form of leadership (27). During these periods, there is a need for transformative leadership to meet external demands and capture positive changes occurring in other organizations. Additionally, interactive leadership is also important in this context. Interactive leadership refers to a leadership style in which the leader interacts with employees and encourages their participation. This leadership style aims to increase employee motivation and commitment by emphasizing communication skills, empathy, teamwork, and support. Interactive leadership in healthcare institutions has positive effects on employee job satisfaction, motivation, and performance.

Hospital services are an area where complex technology and intensive human relationships converge, leading to significant managerial challenges. As the responsibilities and scope of hospital services expand, management difficulties also increase (50).

The healthcare sector is under pressure to increase quality and accessibility, along with efforts to achieve cost-effectiveness (40). Therefore, healthcare service organizations prioritize employees having better leadership skills to survive with a good management understanding (28). Leadership plays an important role in healthcare service management. The influence of managers on their subordinates and their impact on achieving goals are integral parts of leadership qualities (30).

In the healthcare field, leadership is defined as a process in which healthcare managers influence other healthcare workers to achieve specific objectives (11). Leadership also plays a significant role in clinical settings and involves clinicians' efforts to improve service delivery to patients (10). However, there are some differences between the concepts of manager and leader. While a manager is defined as a person who plans, implements, and controls tasks to achieve predetermined objectives, a leader emerges as.

The relationship between leadership and management is important for success in contemporary organizations. Possessing leadership skills as a manager encompasses qualities such as following changes, creativity, guidance, and influence (22). Managers often exercise authority, while leaders exert influence and derive their power from their personal characteristics and leadership positions (49).

The differences between the concepts of manager and leader are becoming clearer. A manager is typically responsible for serving the goals determined by others, whereas a leader acts in line with self-determined goals (49). Managers are often appointed from outside, while leaders emerge from within a group and guide individuals within the same group.

Gardner and Schermerhorn (24) state that they represent some researchers' perspectives on leadership and management (55).

Leadership = Management Perspective: According to this perspective, leadership is the process of selecting talented employees, providing them with purpose and direction, and gaining the trust of employees. Management, on the other hand, consists of critical components such as planning, organizing, and controlling.

Leadership and Management as Separate but Complementary Processes Perspective: According to this perspective, leadership and management are separate but complementary. While leadership functions in areas such as structuring, producing, or adapting to change, the primary function of management is to make an organization successful in terms of budgeting and timing. Both are necessary for success.

Leadership ≠ Management Perspective: According to this perspective, leadership and management are completely different. Managers perceive their role as fulfilling a process in a regular and stable environment. Leaders, on the other hand, focus on creativity and change. They can take risks, work in chaotic and uncertain environments, and seek opportunities for change.

Leadership is an important concept from an organizational perspective due to its power to influence individual and group behaviors (49). Some

managers may struggle to apply leadership skills because they have a direct coercive understanding of subordinates' behaviors (14). In this context, the concept of leadership holds separate importance from management. Leadership encompasses the power to influence group members' behaviors by directing them towards self-determined goals. These leadership skills are necessary for effectively managing organizations.

In summary, the concepts of leadership and management are complementary but not synonymous terms. Leadership plays a significant role in the healthcare sector and clinical settings. It is important for successful managers to enhance their leadership skills in order to manage transformations in healthcare services and increase their ability to influence employees.

Research on leadership in healthcare indicates that various leadership models and dimensions have positive effects on performance and effectiveness in healthcare institutions (60). For example, the "Leadership Model in Healthcare" proposed by the UK's National Health Service Leadership Academy aims to encourage healthcare professionals to become better leaders (45). This model includes nine leadership dimensions such as leading with care, evaluating information, connecting with service, sharing a vision, engaging the team, holding to account, developing capability, and achieving results.

Kumar and Khiljee (39) contributed to the explanation of healthcare leadership dimensions in their study. Leading with care means recognizing and addressing the needs and behaviors of team members and expanding the care environment. Sharing a vision involves establishing a clear direction for long-term goals through trustworthy and secure communication. Engaging the team emphasizes encouraging creative participation in a trust-based environment. Achieving results includes contributing to others and working collaboratively. Evaluating information supports extensive knowledge exchange and creative thinking. Inspiring a shared purpose entails taking personal risks and being bold while adhering to NHS principles. Connecting with service means considering relationships within the system and embracing external approaches. Developing capability involves creating opportunities that support individuals' and teams' long-term skill development. Holding to account aims to have clear expectations, promote continuous improvement, and foster a mindset for innovative change.

These studies demonstrate that leadership dimensions play a crucial role in improving performance and adapting to change in healthcare services. Leaders in the healthcare sector need to understand that the leadership process is

dynamic, emphasizing it as a fundamental element for successful management and improvement efforts (30).

This leadership model aims to enhance the influence of leaders by emphasizing the importance of their role in improving healthcare services, complying with current standards, and resolving organizational alignment issues.

3. The Sustainability of Leadership

The healthcare sector operates in a complex and rapidly changing environment, where leadership plays a crucial role in successful management and service delivery. However, leadership should aim not only to achieve short-term goals but also to target long-term sustainability and success. The sustainability of leadership in the healthcare field can be achieved through leaders developing future-oriented strategies, motivating employees, and embracing organizational values.

The sustainability of leadership in the healthcare domain has been addressed and discussed by numerous researchers. Some studies in this area emphasize the importance of leaders' competencies and leadership behaviors concerning the sustainability of healthcare leadership (4; 25). Characteristics such as effective communication skills, promoting teamwork and a culture of collaboration, creating a vision, and adapting to change are seen as critical factors for the sustainability of leadership in the healthcare sector (6; 15). Another significant factor is leaders' self-management skills. Developing skills such as self-management, stress coping, emotional intelligence, and self-awareness is crucial for sustainable healthcare leadership (42; 54). Additionally, leaders need to focus on long-term goals and adopt sustainability strategies for the sustainability of healthcare leadership (20; 31). Making decisions aligned with organizational values, adhering to ethical standards, and considering the interests of future generations are fundamental elements of sustainable leadership (6; 58).

The sustainability of leadership in the healthcare field can be achieved by focusing on leaders' competencies, leadership behaviors, self-management skills, and sustainability strategies. Research conducted in this regard guides healthcare leaders in embracing the required qualities and approaches for sustainable leadership.

Leadership plays an important role in ensuring the sustainability of healthcare by effectively managing and directing healthcare organizations

towards their sustainability goals. Leaders need to possess skills such as vision-setting, strategic management, promoting a culture of collaboration, teamwork, communication, and change management (4; 25). These skills enable healthcare leaders to influence and motivate employees to achieve organizational goals.

Another crucial factor for the sustainability of healthcare leadership is leaders' self-management skills. Skills such as self-management, stress coping, emotional intelligence, and self-awareness empower leaders to achieve long-term success in healthcare organizations (42; 54). Leaders can embrace the concept of sustainable leadership by focusing on personal development and continuously improving their leadership skills.

Furthermore, it is essential for leaders to focus on long-term goals and embrace sustainability strategies for the sustainability of healthcare leadership. Leaders need to make decisions while considering environmental sustainability, alignment with ethical values, social responsibility, and stakeholder relationships (20; 31). In this way, leaders contribute not only to the short-term success of healthcare organizations but also to long-term sustainability goals.

The sustainability of leadership brings various benefits in the healthcare domain. Healthcare institutions led by effective leadership enhance employee satisfaction, increase productivity, and foster collaboration. Additionally, organizations that achieve leadership sustainability deliver better services to patients, maintain quality standards, and enhance their reliability. As a result, healthcare organizations become more competitive and achieve long-term success.

The sustainability of leadership in healthcare has been extensively studied and researched in academic studies. These studies highlight the importance of leadership skills, self-management abilities, focusing on long-term goals, and embracing sustainability strategies in shaping the success of leadership in the healthcare field. Considering these factors will assist healthcare leaders in achieving the sustainability goals of.

4. Conclusion

In conclusion, leadership is of critical importance for successful management in the healthcare sector. In the management of healthcare services, leadership plays a role in influencing subordinates and enabling them to achieve goals. Research on leadership in the healthcare sector emphasizes the significance of applying leadership theories and approaches in healthcare

settings. These studies provide guidance for the development of healthcare leaders and contribute to the improvement of healthcare services.

The role of leadership in the management of healthcare services is highly significant due to its contribution to directing subordinates towards the achievement of goals. Leadership and management are seen as inseparable concepts, and challenges such as managing change and ensuring cohesion among professionals are encountered in healthcare services as well. In this regard, the transformational leadership approach is considered an effective leadership model for facilitating the work of healthcare professionals.

The role of leaders in the healthcare sector holds great importance in areas such as improving healthcare services, complying with current standards, and resolving internal organizational issues. Leaders understanding the leadership process as a dynamic one and realizing its essential aspect of adapting to daily requirements is crucial for their success. Additionally, it is important for leaders to continuously enhance their leadership skills and evaluate their leadership approaches. Leadership in the healthcare sector is a key factor in successfully managing healthcare services. Therefore, further research on the role and effects of leadership on management in the healthcare sector should be conducted in academic studies.

References

1. Akçakaya, M. (2010). Örgütlerde Uygulanan Personel Güçlendirme Yöntemleri: Türk Kamu Yönetiminde Personel Güçlendirme. *Karadeniz Araştırmaları. 25*:145-174.

2. Ardahan M. & Konal E. (2017). Hemşirelikte yöneticilik ve liderlik. *Gümüşhane Üniversitesi Sağlık Bilimleri Dergisi. 6*(1), 140-147.

3. Astin, H. S., Astin, H., Boatsman, K., Bonous-Hammarth, M., Chambers, T., ve Goldberg, S. (1996). *A social change model of leadership development: Guidebook (Version III)*. Higher Education Research Institute, University of California, Los Angeles.

4. Avey, J. B., Wernsing, T. S., & Palanski, M. E. (2011). Exploring the process of ethical leadership: The mediating role of employee voice and psychological ownership. *Journal of Business Ethics, 96*(4), 543-554.

5. Avolio, B. J., & Gardner, W. L. (2005). Authentic Leadership Development: Getting to the Root of Positive Forms of Leadership. *The Leadership Quarterly, 16*(3), 315-338.

6. Avolio, B. J., Gardner, W. L., Walumbwa, F. O., Luthans, F., & May, D. R. (2010). Unlocking the mask: A look at the process by which authentic leaders impact follower attitudes and behaviors. *Leadersh. Q* (16), 315-338.

7. Avolio, B. J., Walumbwa, F. O. ve Weber, T. J. (2009). Leadership: Current theories, research, and future directions. *Annual review of psychology, 60*, 421-449.

8. Bass, B. M. (1990). From transactional to transformational leadership: Learning to share the vision. *Organizational dynamics, 18*(3), 19-31.

9. Bennis, W., & Nanus, B. (1985). *Leaders: Strategies for taking charge*. Harper & Row.

10. Bilir, N. (2007). Health services and leadership. *Uludağ University Journal of Faculty of Medicine, 33*(2), 97-101.

11. Budak, F. & Özer, Ö. (2017). Kültürlerarası hemşirelik bağlamında klinik liderliğin önemi. 16.05.2023 tarihinde https://www.researchgate.net/publication/321371470_Kulturlerarasi_Hemsirelik_Baglaminda_Klinik_Liderligin_Onemi adresinden erişilmiştir.

12. Bulut, D. & İşman, Ç. (2004). Muğla İli Sağlık Yöneticilerinde Kişisel Değer Analizi, *Hacettepe Sağlık İdaresi Dergisi,, 7(*3): 277–281.

13. Chelladurai, P. (2007). *Leadership in Sports.* In G. Tenenbaum & R. C. Eklund (Eds.), Handbook of Sport Psychology (3rd ed., pp. 113-135). John Wiley & Sons.

14. Conger, J. A. (1985). *Charismatic leadership in business: An exploratory study. Unpublished Doctoral Dissertatio*n, School of Business Administration, Harvard University.

15. Cummings, G. G., MacGregor, T., Davey, M., Lee, H., Wong, C. A., Lo, E., ... & Stafford, E. (2010). Leadership styles and outcome patterns for the nursing workforce and work environment: A systematic review. *International Journal of Nursing Studies, 47*(3), 363-385.

16. Çekmecelioğlu, H. G. (2014). Göreve ve İnsana Yönelik Liderlik Tarzlarının Örgütsel Bağlılık, İş Performansı ve İşten Ayrılma Niyeti Üzerindeki Etkileri. *Kocaeli Üniversitesi Sosyal Bilimler Dergisi, (28*), 21-34.

17. Dalgın, T. (2015). *Yöneticilerin Liderlik Davranışlarının Örgütsel Vatandaşlık Davranışı Üzerindeki Etkisi ve Örgütsel Adalet Algısının Düzenleyici Rolü: Muğla Konaklama Sektörü Örneği.* Doktora Tezi. Muğla Sıtkı Koçman Üniversitesi, Muğla.

18. Davis, K. (1984). *İşletmelerde İnsan Davranışı*. Çeviri: K. Tosun ve diğerleri. İstanbul Üniversitesi Yayınları No:302, 2.baskı, İstanbul.

19. Duygulu, S. & Kubilay, G. (2008). Yönetici hemşirelerin ve birlikte çalıştıkları hemşirelerin liderliğie ilişkin değerlendirmeleri ve yönetici hemşirelerin sahip oldukları liderlik özellikleri. *Sağlık Bilimleri Fakültesi Hemşirelik Dergisi. 18*(5), 1-15.

20. Eisenbeiss S. A., Van Knippenberg D., Boerner S. (2008). Transformational leadership and team innovation: Integrating team climate principles. *Journal of Applied Psychology, 93*, 1438-1446.

21. Eraslan, L. (2004). Liderlikte post-modern bir paradigma: dönüşümcü liderlik. *Journal of Human Sciences, 1(*1), 1-30.

22. Erçetin, S. (1998). *Lider Sarmalında Vizyon*, Ankara: Öder Matbaacılık.

23. Eren, E. (2013). *Stratejik Yönetim ve İşletme Politikası (*9 b.). Beta Basım Yayın Dağıtım, İstanbul, 461-477.

24. Gardner, W. L., & Schermerhorn, J. R. (1992). Organizational leadership: Relationships between management and leadership. *Journal of Management, 18*(3), 535-554.

25. Gardner, W. L., Avolio, B. J., Luthans, F., May, D. R., & Walumbwa, F. (2005). Can you see the real me?"A self-based model of authentic leader and follower development. *The Leadership Quarterly, 16*(3), 343-372.

26. Gardner, W. L., Avolio, B. J., Luthans, F., May, D. R., & Walumbwa, F. (2005). Can you see the real me? A self-based model of authentic leader and follower development. *The Leadership Quarterly, 16*(3), 343-372.

27. Gedikoğlu, T. (2015). *Liderlik ve okul yönetimi*. Ankara: Anı Yayıncılık.

28) Gilmartin, M., D'Aunno, T., & Jiang, L. (2012). An examination of the effects of external quality management practices and transformational leadership on job satisfaction and organizational commitment. *Journal of Healthcare Management, 57*(6), 445-461.

29. Gök, R., (2019). Türk Eğitim Sisteminde Liyakat (Meritokrasi) Esaslı Eğitimyöneticiliği. *Mehmet Akif Ersoy Üniversitesi Eğitim Fakültesi Dergisi (52*) 39-64.

30. Gupta, M., Rao, S., Kumar, P., & Pasha, S. (2019). Transformational leadership: A boon for healthcare delivery. Indian Journal of Public Health, 63(3), 235-238.

31. Hargis, M. B., Watt, J. D., & Piotrowski, C. (2011). Sustainable leadership practices: An empirical investigation. *Journal of Leadership, Accountability and Ethics, 8*(5), 32-47.

32. Johnson, S., Adams, A., Brown, D., & Smith, R. (2019). Transformational leadership in healthcare: A systematic review. *Journal of Advanced Nursing, 75*(10), 2184-2195.

33. Jones, T. L., Hamilton, P., & Murry, N. (2017). Unpacking the black box: A case study approach to understanding clinical leadership. Journal of Health Organization and Management, 31(1), 112-127.

34. Karahan, A. (2008). *Türkiye'deki Devlet ve Üniversite Hastanelerinde Yönetici-Hekimlerin Liderlik Davranışlarının Belirlenmesi*. Yayınlanmamış Doktora Tezi, Gazi Üniversitesi, Ankara.

35. Koçak, T. (2006). *Okul Yöneticilerinin Dönüşümcü Liderlik Stilleri İle Öğretmenlerin İş Doyumları Arasındaki İlişki*, Yayınlanmamış Yüksek Lisans Tezi, Gazi Üniversitesi, Ankara.

36. Koçel, T. (2015). *İşletme Yöneticiliği* (16 b.). İstanbul, Beta.

37. Komives, S. R. & Dugan, J. P. (2010). Contemporary leadership theories. Political and civic leadership: *A reference handbook, (1),* 111-120.

38. Komives, S.., Lucas, N., McMahon, T. R. (2009). *Exploring Leadership: For College Students Who Want To Make A Difference*. John Wiley ve Sons.

39. Kumar, R. D., Khiljee, N. (2016). Leadership in healthcare. *Anaesthesia ve Intensive Care Medicine, 17*:1, 63-65. https://doi.org/10.1016/j.mpaic.2015.10.012

40. Lega, F., De Pietro, C., & Morandi, F. (2018). Cost containment-oriented leadership and employee job satisfaction: A moderated mediation model. *BMC Health Services Research, 18*(1), 954.

41. Leithwood, K., Louis, K. S., Anderson, S., & Wahlstrom, K. (2008). *How Leadership Influences Student Learning*. New York: The Wallace Foundation.

42. Leroy, H., Anseel, F., Gardner, W. L., & Sels, L. (2012). Authentic leadership, authentic followership, basic need satisfaction, and work role performance: A cross-level study. *Journal of Management, 38*(2), 483-510.

43. Lombardi, J. (2017). The road to leadership in SPN. *Journal of Pediatric Nursing, 34*, 96-97.

44. Marquis, B. L., & Huston, C. J. (2009). *Leadership roles and management functions in nursing: Theory and application*. Philadelphia: Lippincott Williams & Wilkins.

45. NHS Leadership Academy (2013) Healthcare Leadership Model: The Nine Dimensions of Leadership Behaviour. NHS Leadership Academy, Leeds, 16.

46. Nikezić, S., Purić, S. & Purić, J. (2012). Transactional and transformational leadership: Development through changes. *International Journal for Quality Research, 1*(6), 285-296.

47. Posner, B. Z. (2012). Effectively measuring student leadership. *Administrative sciences, 2*(4), 221234.

48. Rost, J. C. (1993). *Leadership for the twenty-first century*. USA:Greenwood Publishing Group.

49. Sabuncuoğlu, Z. & Tüz, M. (2008). *Örgütsel Psikoloji*, Furkan Ofset, Bursa.

50. Sayın, N. (2008). *Ortaöğretim Kurumlarında Kriz Yönetimi Stratejisinin İncelenmesi.* Yayınlanmamış Doktora Tezi. Marmara Üniversitesi, İstanbul.

51. Scott, D. (2014). *Contemporary leadership in sport organizations.* USA: Human Kinetics.

52. Serinkan C. & İpekçi İ. (2005). Yönetici hemşirelerde liderlik ve liderlik özelliklerine ilişkin bir araştırma. *Süleyman Demirel Üniversitesi İktisadi ve İdari Bilimler Fakültesi Dergisi, 10*, 281-294.

53. Smith, J., Doe, A., Johnson, B., & Williams, C. (2018). The impact of leadership skills on performance in healthcare services. *Journal of Healthcare Management, 45*(2), 87-94.

54. Sutcliffe, K. M., Vogus, T. J., & Dane, E. (2015). Mindfulness in organizations: A cross-level review. *Annual Review of Organizational Psychology and Organizational Behavior, 2*, 1-25.

55. Şahin, S. (2003). *Okul Müdürlerinin Liderlik Stilleri ile Okul Kültürü Arasındaki İlişkiler*, Yayımlanmamış Doktora Tezi, Dokuz Eylül Üniversitesi Eğitim Bilimleri Enstitüsü, İzmir.

56. Tabak, A., Polat, M., Coşar, S. ve Türköz, T. (2012). Otantik Liderlik Ölçeği: Güvenirlik ve Geçerlik Çalışması. Is, Guc: *The Journal of Industrial Relations & Human Resources, 14*(4).

57. TDK (Türk Dil Kurumu), http://www.tdk.gov.tr, 17.05.2023'te alındı.

58. Waldman, D., Sully de Luque, M., Washburn, N. et al. (2006).. Cultural and leadership predictors of corporate social responsibility values of top management: a GLOBE study of 15 countries. *J Int Bus Stud 37*, 823–837. https://doi.org/10.1057/palgrave.jibs.8400230

59. Walker, B., & Avolio, B. (2019). Ethical leadership in healthcare: Relationships with organizational culture, climate, and performance. *Journal of Healthcare Management, 64*(1), 52-63.

60. West, M. A., Armit, K., Loewenthal, L., Eckert, R., West, T., & Lee, A. (2015). Leadership and leadership development in healthcare: The evidence base. *Faculty of Medical Leadership and Management*.

61. Wong, C. A., & Cummings, G. G. (2007). Authentic Leadership: A New Theory for Nursing or Back to Basics? *Journal of Health Organization and Management, 21*(6), 522-538.

62. Wong, C. A., Cummings, G. G., & Ducharme, L. (2013). The relationship between nursing leadership and patient outcomes: A systematic review update. *Journal of Nursing Management, 21*(5), 709-724.

63. Yağcı, İ. & Duman, T. (2006). Hizmet Kalitesi- Müşteri Memnuniyeti İlişkisinin Hastane Türlerine Göre Karşılaştırılması: Devlet, Özel ve Üniversite Hastaneleri Uygulaması, *Doğuş Üniversitesi Dergisi, 7*(2): 218–238.

64. Yavuz E., (2009). İş Görenlerin Dönüşümcü Liderlik ve Örgütsel Bağlılık ile İlgili Tutumlarına Yönelik Bir Araştırma, *İşletme Araştırmaları Dergisi, 1*(2): 51–69.

65.Yılmaz H. & Kantek F. (2016). Türkiye'deki yönetici hemşirelerin liderlik tarzları: Literatür inceleme. *Journal of Health and Nursing Management, 2*(3), 110-117.

66. Yukl, G. (2010). *Leadership in Organizations*. Upper Saddle River, NJ: Pearson.

CHAPTER XII

NANOSPONGE-BASED DRUG DELIVERY FOR OVERCOMING SOLUBILITY AND PERMEABILITY CHALLENGES OF POORLY WATER-SOLUBLE DRUGS

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1. Introduction

Solubility plays a crucial role in drug discovery and development, as it is estimated 70% to 90% percentage of drug candidates in the developmental phase and approximately 40% of marketed fall under the category of poorly water-soluble compounds. This limitation results in lower oral bioavailability due to their limited dissolution rate during absorption. Oral drug delivery of these substances may also encounter additional challenges, including rapid metabolism, absence of a consistent blood or plasma concentration of the drug, and inter-individual variability (1). In the past decade, significant research efforts have been dedicated to exploring novel drug delivery systems that can overcome these challenges, and among them, nanosponges have gained significant attention (2).

Nanosponges have been addressed as a promising drug delivery system to tackle and revolutionize the drawbacks related to conventional pharmaceuticals. These ingenious structures possess a porous framework that enables them to capture various drugs and be immersed inside its core. Offering a host of

advantages including biocompatibility, porosity, biomimetic properties, sustained release behavior, and therapeutic efficacy, nanosponges have garnered significant attention. Their ability to enhance the bioavailability, stability, permeability and solubility of therapeutic agents or low soluble drugs is particularly noteworthy, as it facilitates the attainment of desired pharmacokinetic effects. By forming complexes with hydrophilic or lipophilic molecules due to their amphiphilic nature, nanosponges exhibit remarkable versatility in transferring and safeguarding drugs from degradation (3,4). In addition to previously mentioned advantages, Nanosponges are capable of incorporating immiscible liquids and transform fluids into powders, thereby enhancing material processing and formulation efficiency. Moreover, nanosponges exhibit exceptional stability ensuring their integrity and functionality in diverse physiological conditions. With their unique three-dimensional structure, nanosponges enable efficient encapsulation and controlled release of active pharmaceutical ingredients, facilitating precise and sustained drug delivery (5,6).

Nanosponges offer key advantages over microsponges, including smaller diameter less than 1 μm and void sizes from 5 to 300 nm, greater strength and stability (withstanding temperatures up to 300 °C), lipophilic properties for flavor masking and compound transformation, enhanced therapeutic properties with biodegradable polyesters, and a structurally similar size to viruses with a naturally degradable polyester framework. Nanosponges are also derived from natural sources, porous, non-toxic, and insoluble in water and organic solvents, distinguishing them from other nanoparticle (7). Since their introduction in 2006, nanosponges have been extensively studied in pharmaceutics for delivering diverse drugs across multiple indications, including infection, inflammation, diabetes, hyperlipidemia, Parkinson's disease, and cancer therapies (8). In this chapter, we focused on the preparation and characterization of nanosponges and explored their potential applications in overcoming solubility and permeability challenges associated with low-soluble drugs.

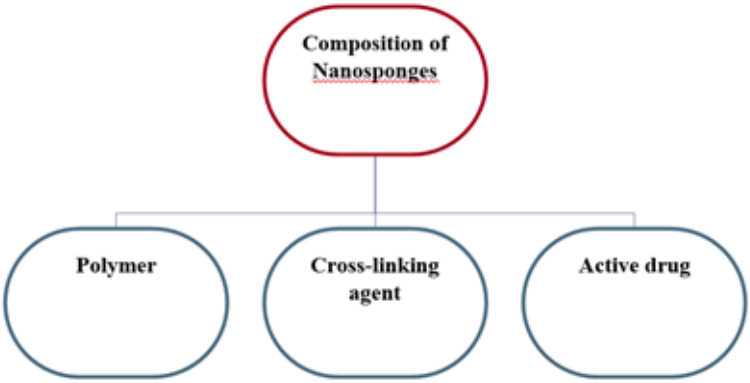

Figure 1. General Composition of Nanopsonges

2. Composition of Nanosponges

Nanosponges are supramolecular, hydrophilic, water-insoluble, nano-sized structures with a porous, 3D, hyper-reticulated nanoporous design made of crosslinked polymers (9). They are mainly composed of polymers mixed with small cross-linking molecules, and the active drug to be loaded as in (Figure 1). The choice of polymer plays a crucial role in determining the efficacy and formation of nanosponges. Examples of polymers used in synthesis of nanosponges include hyper-crosslinked polystyrenes for high surface area, cyclodextrins and their derivatives for molecular encapsulation (e.g., methyl β-cyclodextrin), and copolymers like poly(valerolactone-allylvalerolactone) for tunable properties. In addition to polymers, crosslinking agents like diphenyl carbonate, diisocyanates, and pyromellitic anhydride are used to stabilize the polymer networks. Other agents include carbonyl diimidazoles, epichlorohydrin, glutaraldehyde, carboxylic acid dianhydrides, 2,2-bis(acrylamido)acetic acid, and dichloromethane (10). The ideal drug candidate for nanosponges should have a molecular weight between 100-400 Daltons, no more than five compacted rings, solubility less than 10 mg/ml in water, a melting point below 250 °C (11). Nanosponges are superior to many other nanocarriers because they can be engineered to have specific sizes and can be surface-conjugated with ligands for site-specific drug delivery. As shown in (Figure 2) nanosponges are spherical in shape with multiple cavities for drug storage.

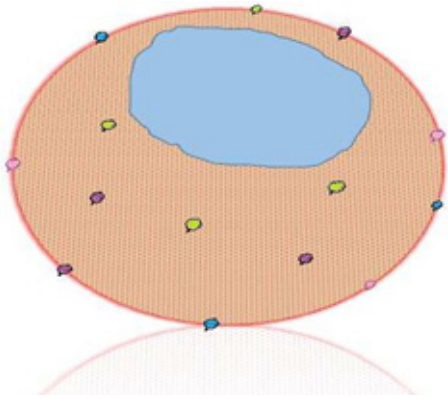

Figure 2. The structure of nanosponges (12)

Nanosponges encompass a diverse range of types including cyclodextrin-based nanosponges, hyper-cross-linked polystyrene, titanium-based nanosponges, and silicon nanosponges. These various types can be customized by incorporating different polymers, adjusting their concentrations, and employing specific preparation methods. When considering the versatility of nanosponges, it becomes evident that beta-CD-based nanosponges hold particular prominence due to their widespread usage and relatively simple formulation process (13).

3. Preparation Methods of Nanosponges

Various methods are employed in the preparation of nanosponges, as depicted in (Figure 3). These methods encompass a diverse range of techniques and play a crucial role in the fabrication of nanosponges for various applications.

3.1. Solvent Method

In solvent method process dimethylformamide and dimethyl sulfoxide act as solvents and facilitate the creation of polymer-based sponges, which constitute the primary structural component. The polymer is introduced into the solvent system and thoroughly mixed. The cross linker-to-polymer ratio is maintained at 8:2 within the mixture, and the resulting blend is allowed to undergo a reaction for a duration of 48 hours at a controlled temperature of 10°C. Subsequently, the reaction is terminated, and the solution is gradually cooled to reach ambient room temperature. To obtain the desired final product, bi-distilled water is employed to extract the cooled solution, which is then subjected to the vacuum filtration process (14).

In a recent study a solvent-based method was developed for the synthesis of cyclodextrin based nanosponges. Initially, anhydrous β-Cyclodextrin is dissolved in 20 mL of Dimethylformamide, followed by the addition of diphenyl carbonate to initiate the crosslinking reaction. After around 4 hours reaction at 100 °C, the resulting transparent nanosponge block was crushed, washed with distilled water, and subjected to Soxhlet extraction using acetone to remove residual reagents. The purified nanosponge was then dried at 60 °C and ground into a fine white powder. In this study, solvent method prepared nanosponges showed consistent size, desirable particle size distribution, and high encapsulation efficiencies. They effectively shielded dithranol from photodegradation, outperforming pure dithranol. Furthermore, a comparison was made between the solvent method and the melt method, and it was found that the nanosponges prepared using the solvent method retained the highest antioxidant activity of dithranol among all the batches produced through solvent evaporation (15).

3.2. Ultrasound-Assisted Method

In this method, nanosponges are synthesized through a cross-linking reaction between polymers and cross-linkers under ultrasonic irradiation. Polymer and cross-linker are carefully combined in a flask at a specific molar ratio. The flask is then placed in an ultrasonic bath and subjected to sonication for around 5 hours at a controlled temperature of 90 °C. Following sonication, the temperature of the reaction mixture is gradually decreased, and subsequent cleavage and washing steps are performed to eliminate unreacted polymer and reagents. The resulting product is then purified using a Soxhlet extraction technique with ethyl alcohol as the extraction solvent. This purification process effectively separates impurities and residual reactants from the nanosponge product. Finally, the nanosponges are dried thoroughly in preparation for subsequent drug loading procedures (16).

Researchers developed 5-Fluorouracil cyclodextrin nanosponges using an ultrasound-assisted technique, aiming to enhance drug accumulation in gastric tumors while minimizing systemic side effects. Nanosponges exhibited nano-sized particles with varied entrapment efficiency ranging from 15.6% to 30% depending on polymer to cross-linker ratio. In vitro dissolution studies revealed of developed formulation achieved a rapid drug release rate, with 56% of the drug dissolved within the first hour. These findings indicate the potential of cyclodextrin nanosponges as a promising anticancer delivery system, offering

improved therapeutic efficacy and reduced systemic side effects for gastric tumor treatment (17). However, Shah et al. compared two methods, ultrasonic-assisted and solvent evaporation synthesis, for developing nimesulide nanosponges. The study aimed to enhance the solubility of the drug and prolong its release using Eudragit L100, PVA, and Dichloromethane. The results showed that the solvent evaporation method yielded higher production yield and drug entrapment. Both methods demonstrated improved dissolution rates in a basic medium and exhibited characteristics similar to previously reported nanosponges in terms of their comb-like structure (18).

3.3. Melt Method

In melt method, the mixture of the cross linker and the polymer are melted through the melting process till homogenization. Then the nanosponges are putted together by washing the product frequently with an appropriate liquid. After cleaning the product, the waste polymer and the reagents that didn't go through the reaction process are removed then the product is divided into the form of nanosponges (19).

Moin et. al, developed polymeric nanosponge tablets for combination therapy by incorporating three drugs into nanosponges prepared using the hot melt method. After optimization, the nanosponges were compressed into tablets and evaluated for drug release. The tablets demonstrated superior drug dissolution properties compared to pure drug counterparts, highlighting the potential of nanosponges for modulating drug release in combination therapy (7).

Rezaei et al. also produced three types of β-cyclodextrin nanosponges using different molar ratios of β-cyclodextrin to diphenyl carbonate as the cross-linker (1:2, 1:4, and 1:8). Anhydrous β-cyclodextrin was reacted with melted diphenyl carbonate at 90ºC for at least 5 hours. The resulting mixture was cooled at room temperature and washed with double distilled water to remove any unreacted β-cyclodextrin. Purification of the product was performed using Sohxlet method with ethanol for 4 hours to eliminate unreacted diphenyl carbonate. The nanosponges were then dried under vacuum and sonicated for 10 minutes (20).

3.4 Emulsion Solvent Diffusion Method

The method consists of two phases: an organic phase and an aqueous phase. In the organic phase, dichloromethane is combined with a solution of ethyl cellulose and the drug. Meanwhile, in the aqueous phase, polyvinyl alcohol is mixed with distilled water. The emulsification process takes place through

dropwise addition, ensuring proper mixing of both phases. Throughout a stirring period of 2 hours at 1000 RPM, the organic solvent evaporates. Following evaporation, the nanosponges are filtered and subsequently dried in an oven at 40ºC for 24 hours. The dried nanosponges are then stored in desiccators until all residual solvents are completely removed (21).

Penjuri et. al., developed Lansoprazole-loaded nanosponges using ethylcellulose, PVA, pluronic F-68, and dichloromethane. The nanosponges exhibited a porous structure with nanochannels, as confirmed by scanning electron microscopy (SEM). The FTIR spectra indicated the stability of lansoprazole within the polymer mixture, with no drug-polymer interactions observed. DSC analysis suggested the involvement of the drug in complexation with the nanosponges. The particle size of the lansoprazole nanoparticles ranged from 83.4 nm to 190.69 nm, and the nanosponges displayed negative zeta potential values, indicating good stability. In-vitro drug release studies revealed extended release for up to 12 hours. The optimized nanosponges were then formulated into an enteric-coated tablet, which exhibited controlled release behavior for 24 hours, following zero-order kinetics (22).

In addition to diffusion method, emulsion solvent evaporation has also been used by Ahmet et al. for incorporating brigatinib into nanosponges. It exhibited optimal characteristics, including a particle size of 261.0 nm, high encapsulation efficiency (85.69%), sustained drug release over 12 hours, and biocompatibility. In vitro studies demonstrated dose-dependent apoptosis activity against A549 cells (23). Furthermore, Ethylcellulose nanosponges were created via an ultrasonic-assisted emulsion solvent evaporation method for the targeted delivery of withaferin-A with anticancer properties, and abemaciclib with sustained-release behavior via an emulsion solvent diffusion technique where the nanosponges showed high stability and sustained release of the drug in 24 h (24).

3.5. Quasi Emulsion Solvent Diffusion Method

Quasi-emulsion method for nanosponge preparation involves the following steps: preparing an inner organic phase by dissolving Eudragit RS 100 in ethyl alcohol, mixing the desired drug with the solution under ultrasonication at 35°C. After that the inner phase is combined with a polyvinyl alcohol (PVA) solution in water as the outer phase and stirring the mixture for 60 minutes at 1000 rpm. Nanosponges are filtered from the suspension, and dried in a hot air oven at 40°C for 12 hours to obtain the final product (25).

By using a quasi-emulsion solvent diffusion method hesperitin nanosponges were developed and optimized with a factorial design approach. The internal phase was prepared by dissolving ethylcellulose and hesperetin at a prearranged ratio into dichloromethane. It was added gradually to the external phase composed of water and polyvinyl alcohol with continuous stirring at 500 rpm for 2 h. By using evaporation method dichloromethane was then removed, the mixture was filtered, and after the hesperitin nanosponges was dried in a hot air oven at 40 °C for 24 h. Hesperetin nanosponges demonstrated delayed drug release, downregulated pro-inflammatory cytokines (IL-1β and IL-6), skin permeation of 18.52% at 8 hours, and reduced inflammation by 33.16% in rats (26).

3.6. Drug Loading into Nanosponges

Nanosponges are initially dissolved in water and then subjected to strong sonication to prevent aggregation. The resulting solution is centrifuged to obtain a colloidal fraction, while the supernatant is separated. The sample is then dried using a freeze dryer or by evaporating the solvent. To incorporate a specific drug, the nanosponge suspension is mixed with the drug and continuously stirred for a specific duration. Centrifugation is employed again to separate noncomplexed drug from the complexed one. Finally, the remaining suspension is dried to obtain solid nanosponge crystals (12). Nanosponges' crystal structure is preferred for drug complexation, as it offers greater loading capacity compared to their paracrystalline counterparts (27).

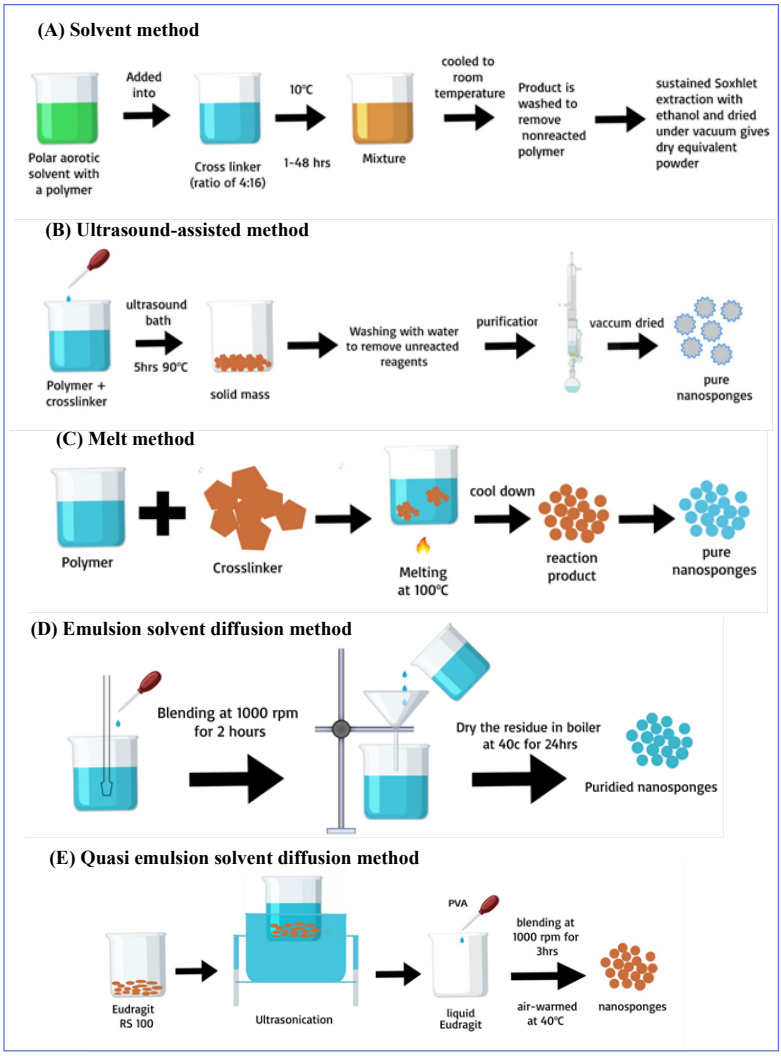

Figure 3. Schematic representation of various preparation methods of nanosponges (13)

4. Characterization of Nanosponges

4.1. Solubility Studies

Phase solubility method, originally proposed by Higuchi and Connors, represents a widely employed approach for investigating inclusion complexation phenomena. Specifically, this method focuses on elucidating influence of nanosponges on drug solubility characteristics. By constructing phase solubility

diagrams, researchers can get insights into the extent and nature of complexation process between drugs and nanosponges. These diagrams provide valuable information regarding the various solubility phases and their respective drug-nanosponge interactions. Through careful analysis and interpretation of phase solubility diagrams degree of complexation between drugs and nanosponges can be detected (28).

Kadian et al, studied solubility of different batches of cyclodextrin nanoparticles and beta-cyclodextrin in comparison to dithranol. Cyclodextrin nanoparticles formulations showed significant improvements in solubilization efficiency compared to free dithranol and beta-cyclodextrin. Molar ratio of beta-cyclodextrin to diphenylcarbazide played a crucial role in enhancing drug solubility, while a low diphenylcarbazide concentration resulted in lower solubility due to the presence of unbound complexes. These findings highlight potential of cyclodextrin nanoparticles as promising carriers for improving the solubility of hydrophobic drugs like dithranol (15).

4.2. Thermo- Analytical Studies

Thermo-analytical methods are used to check if the drug molecule has undergone any change before the nanosponge's thermal degradation. Drug may undergo some changes which indicates complex formation, those changes are evaporation, oxidation, decomposition, melting, or polymeric changes. Thermogram obtained via DTA and DSC can be checked for broadening, shifting and how certain peaks may appear or disappear. (15)

According to Gangadharappa, H.V. et al in an aluminum pan at a 10 °C/min heating rate and in a 25–300 °C temperature range in the process of nitrogen (flow rate of 100 mL/min), 10 mg of the formulation was taken and sealed in. Later on DSC thermogram for samples was done by using DSC- 4000 PerkinElmer Thermal Analyzer, USA (29).

4.3. Stability Studies

For the stability test, it is characterized as the degree to which a product keeps the same properties and characteristics it has at the time of its manufacture, within defined limits, and over its complete storage and usage duration. (30) The formulation was putted in a tightly closed container and held at approved stability chamber at 40± 2°C and 75± 5% Relative humidity for 45 days. Then it was tested at different intervals before and after, for the appearance improvement, pH, drug quality, and in vitro drug release. The results obtained

indicated no significant changes compared for parameters before and after study period, indicating that the developed formulation was stable and passed the stability test (31).

4.4. Microscopic Studies

Transmission Electron Microscopy (TEM) and Scanning Electron Microscopy (SEM) are powerful techniques for examining the microscopic characteristics of drugs, nanosponges, and their complexes. TEM provides high-resolution imaging and allows for detailed analysis of the internal structure and distribution of drug molecules within nanosponges. SEM, on the other hand, offers a surface-level view, enabling observation of particle size, shape, and surface characteristics (7,32). For SEM analysis, a concentrated aqueous suspension is expanded on a slab and dried under vacuum. The sample is enveloped in a cathodic evaporator with a gold layer 20nm thick and diameters were measured to obtain mean particle size (33).

4.5. Powder X-Ray Diffractometry

Powder X-ray diffractometry (XRPD) is utilized to detect inclusion complexation in the solid state. The diffraction pattern of a new substance varies from that of uncomplexed nanosponge because the drug molecule is liquid, and it doesn't have diffraction pattern. There is a comparison that is made between the diffractogram of the complex which is different from every component and the mixture of the drug and polymer molecules in case the drug molecules are in solid state (34). It has been demonstrated that method of preparation may have effect on the crystallinity of the final product of nanosponges. In a study, X-ray powder diffraction was used to evaluate its crystallinity and drug complexation capacity. Nanosponges can be crystalline or paracrystalline, depending on processing conditions. Researchers used ultrasound-assisted and non-ultrasound methods. The ultrasound-assisted method produced a crystalline product, while the non-ultrasound method resulted in a paracrystalline product. XRPD analysis showed increased peak area and decreased intensity-to-width ratio in the paracrystalline product. This indicates differences in structural characteristics between the two forms of Nanosponge formulation (35).

4.6. Particle Size

Particle size and the size distribution can be determined through various techniques such as dynamic light scattering, laser light diffractometry, or zeta

sizer. Through these instruments polydispersity index can also be dtermined (34). Influence of particle size on drug release can be conducted by analyzing the cumulative percentage of drug release from nanosponges over time. This allows for a comprehensive assessment of how different particle sizes impact the release kinetics and efficiency of the drug from the nanosponges (36).

4.7. Fourier Transform Infrared Spectroscopy (FTIR)

Fourier Transform Infrared Spectroscopy (FTIR) is a fundamental analytical technique widely used for verifying the molecular structure of nanosponges. It is also utilized to evaluate the connection between nanosponges and the drug molecules in the solid state. The utilization of the Infra-red spectroscopy is restricted to the drugs with specific bands, like sulfonyl or carbonyl groups. Infrared spectral studies help to know how hydrogen is required in various functional groups. Upon the incorporation of drugs into Nanosuspensions (NS), the FTIR spectra exhibit notable changes such as broadening or shifting of drug peaks. These alterations can be attributed to the molecular interactions occurring between the drug molecules and the NS components. Forinstance, FTIR analysis of cyclodextrin nanosponges reveals important peak characteristics. The absence of the peak at 3450 cm^{-1} indicates presence free primary alcoholic groups in the cross-linking process. Additionally, the peak corresponding to the carbonate group in the cross-linking agent shifts from 1775 cm^{-1} to 1750 cm^{-1}. Notable peaks in the ranges of 1460–1600 cm^{-1} and 1270–1290 cm^{-1} further confirm the successful cross-linking in Cyclodextrin nanpsponges (35,37).

4.8. Determination of Zeta Potential

Zeta potential, also known as electrokinetic potential, plays a crucial role in determining of stability and behavior of nanosponges in solution. Zeta potential refers to the potential difference between the shear plane of nanosponge particles and the surrounding medium when subjected to an electric field. This parameter provides valuable insights into the surface charge and interparticle interactions of nanosponges. A higher absolute zeta potential magnitude indicates increased electrostatic repulsion between particles, leading to enhanced dispersion and reduced aggregation tendency. Generally, a zeta potential threshold of -30 mV to +30 mV is considered as a stability range for nanoparticles but researchers reported surface charges in the range of −20 to −40 mV for nanosponges. (38) By analyzing the zeta potential values, it is possible to assess the stability and

dispersion characteristics of nanosponges, allowing for the optimization of their formulation and performance in various applications (15,39).

4.9. Porosity

Porosity of nanosponge can be evaluated by determining its tapped and untapped (bulk) densities using a marked cuvette with a known volume. The bulk density is determined by inserting a measured amount of nanosponge powder into the cuvette, while the tapped density is obtained by vertically tapping the cuvette against a padded benchtop for 50 cycles. By comparing initial and final mass quantities, the mass data is analyzed. The true density is obtained by dividing the sample weight by its volume. Significantly, the nanosponge exhibits higher porosity compared to parent polymer and co-polymers used for its preparation, owing to its porous nature.

Percent porosity of nanosponges is calculated as follows (40):

$$\text{Porosity} = \frac{\text{bulk density}}{\text{tap density}} \times 100$$

4.10. Loading Efficiency

Assessment of loading efficiency in nanosponges involves the deduction of the unentrapped drug amount from the total drug content. To determine the quantity of untrapped drug, several effective analytical approaches can be utilized. Dialysis, gel filtration, and ultra-centrifugation are among the commonly employed techniques used to isolate and separate the unentrapped drug from the nanosponges. Loading efficiency can be calculated from the following equation (27):

$$\text{Loadingefficiency} = \frac{\text{Actual drug content}}{\text{Theoritical drug content}} \times 100$$

5. Enhancing Performance of Poorly Soluble Drugs with Nanosponges

Nanosponges are able to take in both lipophilic and hydrophilic drug substances, essentially (BCS-class II) which are the drugs that drugs molecules that belong to the biopharmaceutical classification system also the poorly water-soluble drug. For drug delivery, due to their tiny cavity structure nanosponges

can accept drugs that are water insoluble. They can be used to enhance the dissolution rate, solubility and permeability of drugs. The β-cyclodextrin based nanosponges are reported to be three or five times more effective to deliver the drug to the targeted site than injection (41–43). Ansari et al. synthesized nanosponges (NS) by reacting cyclodextrin with a cross-linker to enhance the solubility, stability, and permeation of resveratrol. The resveratrol-loaded NS showed excellent encapsulation, solubility, stability, and permeation properties. The NS complex exhibited a significantly improved release rate compared to the pure drug and demonstrated superior photostability. Cytotoxicity studies revealed increased cytotoxic activity of the resveratrol NS complex compared to plain resveratrol. Moreover, the permeation and accumulation results indicated potential applications of the resveratrol NS complex in buccal and topical delivery (44).

Cyclodextrin nanosponges have been introduced as safe carriers of drugs/therapeutic agents for the treatment of various diseases, especially cancers/tumors. In addition to that, Cyclodextrins can be directly co-polymerized with other monomers or grafted onto organic/inorganic compounds due to the presence of hydroxyl groups with the capability of a substitution/elimination process. These nanosystems are resistant to organic solvents and can show good thermal stability (45).

Pawar and Shende synthesized Hyper-crosslinked cyclodextrin nanosponges via a solvent evaporation technique and loaded with artemether and lumefantrine (antimalarial agents) to improve their solubility and to acquire a controlled-release profile. These nanosponges showed good stability at 40 C for 3 months and were used for the targeted delivery of doxorubicin (46).

Moreover, Dynamic DNA nanosponges enable efficient gene regulation and tumor-targeted drug delivery. Their stability, biocompatibility, and tumor accumulation capabilities make them promising for multimodal imaging and targeted drug delivery system (47)**.** Jin et al. developed sponge-like nanoplatforms for DNAzyme-mediated gene regulation and programmable tumor-targeted delivery with high efficiency. These nanosponges were employed for the photothermal therapy of cancers to overcome thermal resistance (48).

Cyclodextrin-based nanosponges serve as advantageous carriers for enzymes, proteins, and gases in the biomedical field. These nanosponges enable the adsorption and encapsulation of proteins and macromolecules, extending their activity, enhancing efficiency, and facilitating sustained release. Moreover, nanosponges containing oxygen can effectively deliver this gas to hypoxic tissues,

addressing oxygenation deficiencies (49). Nanosponges are versatile entities that can be utilized in various dosage forms, including oral, parenteral, topical, and inhalation formulations. In the context of tablet and capsule preparations, nanosponge complexes can be effectively dispersed within a well-suited matrix comprising lubricants, diluents, and anti-cracking agents. This formulation strategy ensures the proper integration of nanosponges into solid dosage forms, optimizing their stability, uniformity, and overall performance. By employing suitable excipients and carefully designing the formulation, the nanosponge complexes can be seamlessly incorporated into tablets and capsules, facilitating their efficient administration and enhancing therapeutic outcomes (50).

Considering some studies, it shows that the Nanosponges are classified as a group of nanoparticles that can contain small drug molecules which may lead to a dose drop because of an early dissolution of crosslinker since the degree of crosslinking may have an effect on the drug loading capacity. The crosslinking determines the void space available in the Nanosponges which can be used in drug loading which will necessitate the evolution of crosslinkers as well as the establishment of new production methods (51). These studies provide valuable insights into the use of nanosponges for permeability and solubility enhancement and shows how the field of nanosponges is continuously evolving.

6. Conclusion

Nanosponges have cavities that can contain both hydrophilic and lipophilic drug molecules and enhance the solubility of poorly water-soluble substances. They are able to deliver the drug in a controlled system at a target site and for adequate time which helps to reduce reduces side effects, improve stability, and improve formulation flexibility and better patient compliance. Nanosponges comprise into topical preparation such as lotions, cream, ointments, oral, parenteral and inhalation. Further research and development in this area will undoubtedly uncover the full potential of nanosponges in drug delivery and other diverse applications

References

1. Khan KU, Minhas MU, Badshah SF, Suhail M, Ahmad A, Ijaz S. Overview of nanoparticulate strategies for solubility enhancement of poorly soluble drugs. *Life Sciences*. 2022;291:120301. doi:https://doi.org/10.1016/j.lfs.2022.120301

2. Bhavin SE, Anuradha G. Nanosponge Approach -A Plethora of Opportunities as a Promising Nanocarrier for Novel Drug Delivery. *Recent Pat Nanotechnol*. 2022;16(4):271-282. doi:10.2174/1872210515666210720141736

3. Mostafavi E, Iravani S, Varma RS. Nanosponges: An overlooked promising strategy to combat SARS-CoV-2. *Drug Discovery Today*. 2022;27(10):103330. doi:10.1016/j.drudis.2022.07.015

4. Kaur S, Kumar S. The nanosponges: an innovative drug delivery system. *Asian Journal of Pharmaceutical and Clinical Research*. Published online 30 2019:60-67. doi:10.22159/ajpcr.2019.v12i7.33879

5. S S, S A, Krishnamoorthy K, Rajappan M. Nanosponges: a novel class of drug delivery system--review. *J Pharm Pharm Sci*. 2012;15(1):103-111. doi:10.18433/j3k308

6. Ahire PS, Bhambere DS, Patil MP, Kshirsagar SJ. Recent advances in nanosponges as a drug delivery system. *Indian J Drugs*. 2020;8:8-17.

7. Moin A, Roohi NKF, Rizvi SMD, et al. Design and formulation of polymeric nanosponge tablets with enhanced solubility for combination therapy. *RSC Adv*. 10(57):34869-34884. doi:10.1039/d0ra06611g

8. Allahyari S, Zahednezhad F, Khatami M, Hashemzadeh N, Zakeri-Milani P, Trotta F. Cyclodextrin nanosponges as potential anticancer drug delivery systems to be introduced into the market, compared with liposomes. *Journal of Drug Delivery Science and Technology*. 2022;67:102931. doi:10.1016/j.jddst.2021.102931

9. Iravani S, Varma RS. Nanosponges for Drug Delivery and Cancer Therapy: Recent Advances. *Nanomaterials*. 2022;12(14). doi:10.3390/nano12142440

10. Gedam SS, Basarkar GD. Nanosponges: An Attractive Strategy for Enhanced Therapeutic Profile. In: ; 2019.

11. Jenny AV, Merima P, Alberto F, Trotta F. Role of beta cyclodextrin nanosponges in polypropylene photooxidation. In: ; 2011.

12. Bhowmik H, Venkatesh D, Kuila A, Kumar K. Nanosponges: A review. *International Journal of Applied Pharmaceutics*. 2018;10:1. doi:10.22159/ijap.2018v10i4.25026

13. Tiwari K, Bhattacharya S. The ascension of nanosponges as a drug delivery carrier: preparation, characterization, and applications. *Journal of materials science Materials in medicine*. 2022;33(3):28. doi:10.1007/s10856-022-06652-9

14. Rubin Pedrazzo A, Caldera F, Zanetti M, Appleton SL, Dhakar NK, Trotta F. Mechanochemical green synthesis of hyper-crosslinked cyclodextrin polymers. *Beilstein journal of organic chemistry*. 2020;16:1554-1563. doi:10.3762/bjoc.16.127

15. Kadian V, Dalal P, Kumar S, Kapoor A, Rao R. Comparative evaluation of dithranol-loaded nanosponges fabricated by solvent evaporation technique and melt method. *Future Journal of Pharmaceutical Sciences*. 2023;9(1):13. doi:10.1186/s43094-023-00461-9

16. Asfaram A, Ghaedi M, Dashtian K. Ultrasound assisted combined molecularly imprinted polymer for selective extraction of nicotinamide in human urine and milk samples: Spectrophotometric determination and optimization study. *Ultrasonics sonochemistry*. 2017;34:640-650. doi:10.1016/j.ultsonch.2016.06.018

17. Jasim IK, Alhammid SNA, Abdulrasool AA. Synthesis and evaluation of B-cyclodextrin Based Nanosponges of 5- Fluorouracil by Using Ultrasound Assisted Method. *Iraqi Journal of Pharmaceutical Sciences (P-ISSN 1683 - 3597 E-ISSN 2521 - 3512)*. 2020;29(2):88-98. doi:10.31351/vol29iss2pp88-98

18. Shah PA, Syed HK, Sohail AR, et al. Comparison of solvent evaporation and ultrasonicassisted production methods in the development of nimesulide nanosponges and their characterization. *Tropical Journal of Pharmaceutical Research*. 2022;21(6):1139-1145. doi:10.4314/tjpr.v21i6.1

19. Rao MR, Bhingole RC. Nanosponge-based pediatric-controlled release dry suspension of Gabapentin for reconstitution. *Drug development and industrial pharmacy*. 2015;41(12):2029-2036. doi:10.3109/03639045.2015.1044903

20. Rezaei A, Varshosaz J, Fesharaki M, Farhang A, Jafari SM. Improving the solubility and in vitro cytotoxicity (anticancer activity) of ferulic acid by loading it into cyclodextrin nanosponges. *Int J Nanomedicine*. 2019;14:4589-4599.

21. Li D, Ma M. Nanosponges : From inclusion chemistry to water purifying technology. In: ; 1999.

22. Penjuri S, Ravouru N, Damineni S, Bns S, Poreddy S. Formulation and Evaluation of Lansoprazole Loaded Nanosponges. *Turkish Journal of Pharmaceutical Sciences*. 2016;13:304-310. doi:10.4274/tjps.2016.04

23. Ahmed MM, Fatima F, Anwer MK, Ansari MJ, Das SS, Alshahrani SM. Development and characterization of ethyl cellulose nanosponges for sustained

release of brigatinib for the treatment of non-small cell lung cancer. *Journal of Polymer Engineering*. 2020;40(10):823-832. doi:10.1515/polyeng-2019-0365

24. Anwer MK, Fatima F, Ahmed MM, et al. Abemaciclib-loaded ethylcellulose based nanosponges for sustained cytotoxicity against MCF-7 and MDA-MB-231 human breast cancer cells lines. *Saudi Pharmaceutical Journal*. 2022;30(6):726-734. doi:https://doi.org/10.1016/j.jsps.2022.03.019

25. Sharma R. Evaluation of the Kinetics and Mechanism of Drug Release from Econazole nitrate Nanosponge Loaded Carbapol Hydrogel. *Indian Journal of Pharmaceutical Research and Education*. 2011;45(1):25-31. doi:Nill

26. Rodrigues K, Nadaf S, Rarokar N, et al. QBD approach for the development of hesperetin loaded colloidal nanosponges for sustained delivery: In-vitro, ex-vivo, and in-vivo assessment. *OpenNano*. 2022;7:100045. doi:https://doi.org/10.1016/j.onano.2022.100045

27. Yadav GV, Panchory HP. "Nanosponges – A boon to the targeted drug delivery system. *Journal of Drug Delivery and Therapeutics*. 2013;3(4):151-155. doi:10.22270/jddt.v3i4.564

28. Trotta F, Zanetti M, Cavalli R. Cyclodextrin-based nanosponges as drug carriers. *Beilstein J Org Chem*. 2012;8:2091-2099. doi:10.3762/bjoc.8.235

29. Rita L, Amit T, Chandrashekhar G, Kundnani PKM. Current trends in β-cyclodextrin based drug delivery systems. In: ; 2011.

30. Gangadharappa HV, Chandra Prasad SM, Singh RP. Formulation, in vitro and in vivo evaluation of celecoxib nanosponge hydrogels for topical application. *Journal of Drug Delivery Science and Technology*. 2017;41:488-501. doi:10.1016/j.jddst.2017.09.004

31. Vyas A, Saraf S, Saraf S. Cyclodextrin based novel drug delivery systems. *Journal of Inclusion Phenomena and Macrocyclic Chemistry*. 2008;62(1):23-42. doi:10.1007/s10847-008-9456-y

32. Cavalli R, Trotta F, Tumiatti W. Cyclodextrin-based Nanosponges for Drug Delivery. *J Incl Phenom Macrocycl Chem*. 2006;56(1):209-213. doi:10.1007/s10847-006-9085-2

33. Swaminathan S, Pastero L, Serpe L, et al. Cyclodextrin-based nanosponges encapsulating camptothecin: Physicochemical characterization, stability and cytotoxicity. *European Journal of Pharmaceutics and Biopharmaceutics*. 2010;74(2):193-201. doi:https://doi.org/10.1016/j.ejpb.2009.11.003

34. Silpa G, Manohar D, Dharan SS. Formulation and Evaluation of Nanosponges Loaded Hydrogel Using Different Polymers Containing Selected Antifungal Drug. In: ; 2021.

35. Ahmed RZ, Patil G, Zaheer Z. Nanosponges – a completely new nano-horizon: pharmaceutical applications and recent advances. *Drug Development and Industrial Pharmacy*. 2013;39(9):1263-1272. doi:10.3109/03639045.2012.694610

36. Seema G, Kumar SA, Manoj B. Development and evaluation of curcumin loaded nanosponges for colon drug delivery. In: ; 2015. Accessed June 30, 2023. https://www.semanticscholar.org/paper/DEVELOPMENT-AND-EVALUATION-OF-CURCUMIN-LOADED-FOR-Seema-Kumar/263becac994167f9cc3c673f6a4e24811e71aad3

37. Swaminathan S, Pastero L, Serpe L, et al. Cyclodextrin-based nanosponges encapsulating camptothecin: Physicochemical characterization, stability and cytotoxicity. *European journal of pharmaceutics and biopharmaceutics : official journal of Arbeitsgemeinschaft für Pharmazeutische Verfahrenstechnik eV*. 2009;74:193-201. doi:10.1016/j.ejpb.2009.11.003

38. Osmani RA, Kulkarni P, Manjunatha S, et al. Cyclodextrin Nanosponges in Drug Delivery and Nanotherapeutics. In: Dasgupta N, Ranjan S, Lichtfouse E, eds. *Environmental Nanotechnology: Volume 1*. Environmental Chemistry for a Sustainable World. Springer International Publishing; 2018:279-342. doi:10.1007/978-3-319-76090-2_9

39. Bhattacharjee S. DLS and zeta potential – What they are and what they are not? *Journal of Controlled Release*. 2016;235:337-351. doi:10.1016/j.jconrel.2016.06.017

40. Moin A, Roohi NKF, Rizvi SMD, et al. Design and formulation of polymeric nanosponge tablets with enhanced solubility for combination therapy. *RSC Adv*. 2020;10(57):34869-34884. doi:10.1039/d0ra06611g

41. Trotta F, Dianzani C, Caldera F, Mognetti B, Cavalli R. The application of nanosponges to cancer drug delivery. *Expert opinion on drug delivery*. 2014;11(6):931-941. doi:10.1517/17425247.2014.911729

42. Pandey P, Purohit D, Dureja H. Nanosponges -A Promising Novel Drug Delivery System. *Recent Pat Nanotechnol*. 2018;12(3):180-191.

43. Gidwani B, Vyas A. A Comprehensive Review on Cyclodextrin-Based Carriers for Delivery of Chemotherapeutic Cytotoxic Anticancer Drugs. *BioMed research international*. 2015;2015:198268. doi:10.1155/2015/198268

44. Ansari KA, Vavia PR, Trotta F, Cavalli R. Cyclodextrin-based nanosponges for delivery of resveratrol: in vitro characterisation, stability, cytotoxicity and permeation study. *AAPS PharmSciTech*. 2011;12(1):279-286. doi:10.1208/s12249-011-9584-3

45. Asela I, Donoso-González O, Yutronic N, Sierpe R %J P. β-Cyclodextrin-Based Nanosponges Functionalized with Drugs and Gold Nanoparticles. 2021;13.

46. Pawar S, Shende P. Dual drug delivery of cyclodextrin cross-linked artemether and lumefantrine nanosponges for synergistic action using 23 full factorial designs. *Colloids and Surfaces A: Physicochemical and Engineering Aspects*. 2020;602:125049. doi:https://doi.org/10.1016/j.colsurfa.2020.125049

47. Luo D, Lin X, Zhao Y, et al. A dynamic DNA nanosponge for triggered amplification of gene-photodynamic modulation. *Chemical Science*. 2022;13(18):5155-5163. doi:10.1039/D2SC00459C

48. Jin Y, Liang L, Sun X, et al. Deoxyribozyme-nanosponges for improved photothermal therapy by overcoming thermoresistance. *NPG Asia Materials*. 2018;10(5):373-384. doi:10.1038/s41427-018-0040-7

49. Jitendra G, Prabakaran L, Reena G, Mohan G. Nanoparticles formulation using counterion induced gelification technique: invitro chloramphenicol release. In: ; 2011.

50. Cavalli R, Akhter AK, Bisazza A, Giustetto P, Trotta F, Vavia P. Nanosponge formulations as oxygen delivery systems. *International Journal of Pharmaceutics*. 2010;402(1):254-257. doi:10.1016/j.ijpharm.2010.09.025

51. Osmani R, Thirumaleshwar S, Bhosale R, Kulkarni P. Nanosponges: The spanking accession in drug delivery-An updated comprehensive review. *Der Pharmacia Sinica*. 2014;5:7-21.