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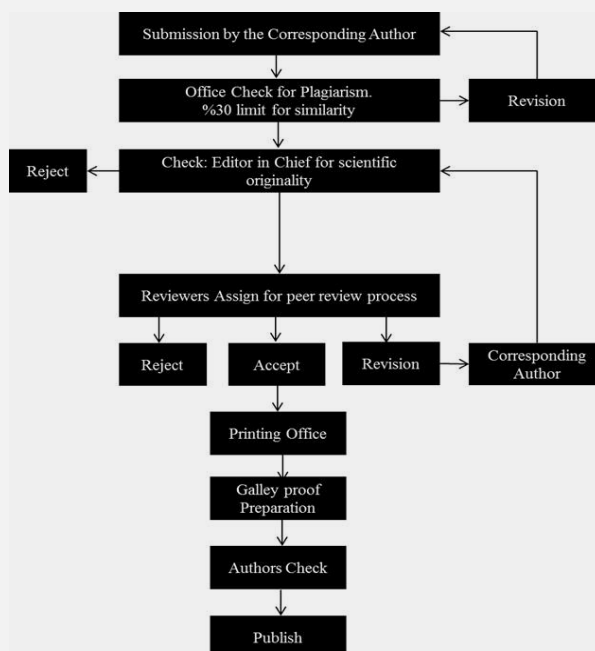
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## Assessment of communication needs and planning communication actions during health crises

Athanasios Zafeirakis<sup>1\*</sup>, Panagiotis Efstathiou<sup>2</sup>

### Abstract

Health crisis communication (HCC) is a challenging and urgent task of the emergency preparedness planning of any welfare state. In this paper some particular reasons for that will be more specifically analyzed. The action flow of HCC includes the phases of preparedness, warning, response, recovery and evaluation. For a successful HCC detailed guidelines are also needed, along with profound knowledge of how the crisis stakeholders should deal with the psychological needs of the citizens and the mass media, as well as with some specific technical items. The ultimate implication of HCC is that the public is aware of its right to make informed choices after having been actively involved in the procedure of risk decisions making.

**Keywords:** Health crisis, crisis communication, preparedness, public emergencies

### Introduction

#### Crisis communication meaning

Crisis communication refers to the production, assessment and interactive exchange of information and messages with the public and institutions, concerning specific risks and hazards, before, during and after a hazardous incident (1). It aims to reduce damage, initiate recovery, manage responsibilities, promote support and justify all required actions (2); it also aims to inform the public concerning an emergency, identify and avoid risks and adverse outcomes, stir actions and thus to protect both individuals and the whole community from the consequences of a crisis incident (3). Furthermore, crisis communication refers to the coordinated efforts on behalf of crisis stakeholders to afford urgent, often incomplete or unverified, information to the public, under extreme time pressure and with unpredicted outcome, ultimately helping the public to accept the imperfect nature of alternatives during a critical situation (4).

#### Guidelines for communication during health crises

HCC is a challenging task due to the needs for a large scale response of various stakeholders, the limited and compressed time span of required actions, the diffusely uncertain or unstable situation -at least in the beginning of the crisis- and the emotionally unforeseen reactions of the public under stress (3). Fast and understandable information leads to high quality crisis decisions, whilst misinformation or communication collapse can make a health crisis even worse and increase damage (5). Practical guidelines for HCC are the following:

1. Communication network establishment, aiming at various stakeholders and public groups (6);
2. Recognition of the inherent uncertainty of emerging risks (7);
3. Paying attention to public perceptions and fears of the danger (8);
4. Honesty, compassion and credibility (9);
5. Provision of self-efficacy messages to the public, for physical and psychological protection against the consequences of crisis (7, 9);
6. Maintaining accessibility for the media, to act appropriately (10);
7. Continuous evaluation and revision of crisis mitigation planning, along with testing of validity of theories and concepts, to prevent similar future challenges (11).

#### Psychology of HCC

Health crises and emergencies tend to take a grave toll on human lives due to their complexity, to the radical changes of routine, and the high degree of uncertainty they introduce in normal life (2). Even more, when crises are manmade, they tend to raise even stronger psychological and emotional reactions (12). In such crises the affected population tends to receive and process data and act according to crisis information in a different or even exaggerated way in comparison to what they are used to do



in normal life (13). Thus, in order to achieve an effective crisis communication and to reduce the psychological impact of a hazardous incident, three psychological levels must be deeper apprehended by the health crisis managers: The physical level, that focuses on the public's basic requirements for food and shelter, the psycho-social level, that focuses on social and psychological requests for family and society support and the interpretative level, that focuses on the means by which various social, cultural, economical, etc, groups apprehend messages and process crisis information (14). From the psychological point of view the health crisis messages should be:

1. Simple, otherwise under the intense stress and possible information overload the key message may be lost or misinterpreted (15);
2. Consistent and convincing; it should come from credible sources and be repeated to maintain public composure;
3. Adhered to current beliefs; counterintuitive instructions, such as evacuating a seemingly, or for nonce, safe area need delicate communication approaches (16);
4. The first to be received; people under stress tend to believe the first message, either rumor or reality (17);
5. Sent on multiple communication channels; people want always to verify the validity of risk messages (18);
6. Focusing on practical steps and on specific protective actions (19).

### **Crisis communication actions flow**

For a successful HCC, credibility, trust and accuracy of information are required; these result from fast transmission of clear and realistic instructions in ordinary language, in a reassuring, non-dogmatic and prioritizing way. Moreover, dignity, protection of privacy and confidentiality in the handling of sensitive personal data are also necessary, along with special care in avoiding the "infodemic" status, i.e. confusing facts with fiction, partly due to the dissemination of an enormous amount of inaccurate or misleading information that is aided mainly because of the global distribution of the social media. Nevertheless, the chain of generally non-confidential crisis information should include easily apprehensible clinical and epidemiological details -when available-, along with explicit communication channels, yet discouraging undue panic reactions from the public (20). Underneath follow the five main phases of HCC management:

#### **1. Preparedness**

The communication in the preparedness phase gathers information concerning the public's comprehension and behavior towards a health crisis situation along with its trust to authorities. Scientific experts are usually accepted by the public as more effective and credible spokespersons than ordinary politicians. Concerning in particular the public, better compliance with preventive measures and overall better preparedness outcomes are correlated with older age and higher socioeconomic status, as well as broad

family, social and interpersonal networks (21), female gender (22), urban place of residence versus rural areas (19) and perhaps most of all, the trust in the messages from public authorities (23). To accomplish an integrated pre-crisis communication strategy, the following targets should be identified (24, 25):

1. Clear objectives are set and needs are assessed;
2. National and international laws and guidelines regarding crisis communication are reviewed;
3. An integrated crisis communication plan involving detailed reporting protocols in multiple channels is formulated and periodically reviewed;
4. Crisis communication monitoring mechanisms along with relative tests and simulations are carried out to assess the effectiveness of the communication system and to verify and fix potential operational flaws before emerging a real crisis situation;
5. Detailed and updated databases of relevant stakeholders and health care individuals are prepared and a broad communication network is established;
6. Functions and responsibilities of involved operational branches and communication teams are defined;
7. Official communication delegates are designated and trained to effectively manipulate the mass media, to maintain their accessibility to facts and to motivate the public to positive actions;
8. Communication channels with all community structures and cultures are planned to distribute valuable information, also incorporating the necessary feedback;
9. Special emphasis is given to identify vulnerable groups or specific target populations;
10. Suitable prominent personalities and celebrities are enlisted, who may share information and support communication efforts through campaigns, public statements, etc.

During the preparedness phase three sets of communication are required: Broad and detailed information, i.e. what exactly has happened, what is going to be done next; practical directives for the population, i.e. what to do and what not to do; and key messages, i.e. how people can find help and maintain their self-control. And all these message sets must be repeatedly issued on multiple communication channels to allow efficient distribution and to ensure sufficient absorption of information of all social, economical and educational classes (25).

#### **2. Warning**

The pre-crisis warning period is a rather challenging and mostly brief, latent period which lasts between the identification of an oncoming threat and the time point when the threat presents its full potential. In sudden crises this period may often not appear at all; such sorts of crises emerge, or better erupt without previous warning signs. Major challenges of the warning period are first, the motivation of the community and state to appropriate

measurements and second, the preparation of steps and procedures necessary for the following stages of the evolving crisis. The media, which regularly cover the crisis on a 24h/7d basis, may well be used as a valuable assistant tool to the communication management (24).

### 3. Response

The emergency response period is the core phase of the crisis outbreak. Major communication challenges of the response phase are the mobilization of personnel and resources and the skilful motivation of specific actions and procedures to aid in damage minimization, operating under constantly changing conditions. More specifically in that phase (24):

1. Consistent and clarified crisis information is obtained and the severity of the situation is determined;
2. The crisis leader is thoroughly informed;
3. Trust and realistic expectations are promoted;
4. Messages, preferably pre-prepared and optimistic, are distributed to the public;
5. The media-contact delegates are activated to monitor the information flow from the media, to ensure correctness and promptness of official crisis information and also to establish a bidirectional communication between the media and the public;
6. Appropriate press-rooms and spokespersons are activated; the latter start continuous information for the public about appropriate actions they should undertake and they furthermore fight potential noxious rumor dissemination; moreover the spokespersons organize meetings with opinion leaders from public and private stakeholders;
7. Call centers are activated along with a variety of information channels, e.g. dedicated phone hotlines, live radio/television broadcasts and continuously updated web pages.

### 4. Recovery

In that phase mental and physical recovery of the affected population, along with reconstruction of infrastructures is the only way to get back to the stage of routine. Relevant communication challenges are (24):

1. Positively informing the public that the danger has faded away -or is on the way to it- and that normal health services are being resumed;
2. Encouragement of the public to continuously support the response stakeholders and the resiliency efforts of the society, also avoiding in that time to assign responsibilities and blames for the crisis incident.

### 5. Evaluation

The principal challenges in this period are to assess the effectiveness of the entire crisis communication system, the lessons learned in each of the previous phases, as well as the mistakes in actions of preceding phases, so that the best practices in future responses to be documented. These have to be done in a systematic way so that preparedness for

similar future incidents to be accomplished based on collected data from previous phases, by implementing the necessary structural changes and reformations and by decisive reallocation of funds and budgets (24).

### Citizens

According to CDC, the emergency crisis information to the citizens is defined as “the ability to develop, coordinate, and disseminate information, alerts, warning, and notifications to the public and incident management responders.” (26). During health crises, emergency communication to the public is critically important; thus to be efficient, health crisis communication needs a deep apprehension of the target population, which is often a notably diverse cultural mosaic, as regards not only language and socio-economical parameters, but also the psycho-social perception of the very danger (23). Communication inequalities are translated into differences among individuals and social groups in terms of apprehending and utilizing crisis information (27). By knowing those cohorts translates into comprehension of prejudices and false perceptions concerning the proper behavior of each population group towards crisis. That apprehension also includes knowledge of who are the disabled, chronically ill and medicated people, the immigrants and all other sorts of vulnerable population cohorts. The knowledge of the public relies on various information sources, such as existing demographic data, case studies, or statistical research resulting from social sciences, insurance agencies, governmental departments and academic institutes (14).

### Media

Mass media have a central role in risk communication (28, 29); the reaction of the public is heavily determined by media coverage (13), since those have the power to convert a minor event into a genuine crisis incident. Although well-documented research proved that the source and the form of information do actually influence the behavior of the public towards health crises (30, 31), trust to the media is especially important in situations of extreme hazards where the public feels they have no personal control over critical risks (32). During health crises vital preventive and supportive relevant information may be disseminated through many communication channels to different receivers (33). Prompt communicational response is herein necessary because the media habitually lurk for rapid information sources (34) and it is thus improbable that there will be any time delay between a major incident and media coverage. Furthermore, according to empirical studies, the social media platforms may well engage the public in rapid response reactions in such cases (35, 36) and yet individuals seem then to prefer sharing crisis information via interpersonal social channels than via private personal communication (37). Important issues to be considered in addressing health crisis messages to the public are (14):

1. High levels of message uniformity, with a minimum number of spokespersons; the principle is valid mostly in relatively homogenous populations;



2. A multi-voice, multi-language approach, addressing to rather heterogeneous societies;
3. Clear, scientifically solid and unambiguous messages, with special care to avoid inconsistencies and disagreement among health crisis experts; the key-message should be simple and easily conceivable even from non-experts and should give practical guidelines for stepwise actions of the public to confront the threat;
4. Above all, the main message should be honest and credible; truth should always be told at any cost, in any phase of the actions and in all situations; vital information should never be withheld on grounds of concerns that it might provoke panic.
3. A medical analysis system, for automated information verification to avoid heterogeneous, ambiguous or unverified data collection or duplication of verified data and to control massive data overflow.
4. A sophisticated system for continuous surveillance of sensitive or strategic civil areas (e.g. cities or forests), with broad camera-surveillance networks based on advanced, interactive, interconnected and interworking mapping applications, which after electronic data-flow processing, trigger actions according to certain hazard thresholds.

### Technical items

Operational branches required in health crisis communication are at minimum the following:

1. A Crisis Telecommunication team, which utilizes emergency crisis communication services and infrastructure to share urgent crisis information among delegates from national and international operational stakeholders from both public and private health agencies.
2. A Data Management team, which gathers data and controls the operational information sharing for proper mobilization of resources and for coordination among relevant experts and stakeholders from both country-level and international stakeholders, such as the E-CDC or the WHO.

The necessary high technology communication infrastructure and equipment should be available before the health crisis. That equipment should be potent, efficient, all-weather resistant and self-sustained; it should be also able to provide timely and continuous web-based information flow and accurate data management on a 24h/7d basis, according to high level security standards. Such equipment should include dedicated computers, printers, fax machines, specific software programs, internet servers, landline phones and CB radios -in case of a cell phone network overload/collapse-, along with a portable generator-based and UPS-supported power supply. Importantly, integrated and professional audio-visual equipment, such as TVs, portable microphones, sound systems, projectors, screens and recording devices, is required for press-conference purposes. Among the various technical challenges in the communication chain the following might be listed:

1. A fast Early Warning and Alert System (EWAS), for the coordinated recall of health staff during health crises emergency.
2. An information sharing platform to allow for data exchange to multiple receivers in unified format; such is the Global Disaster Alert and Coordination System (GDACS), aiming to facilitate world-wide disaster information exchange beyond cross-national organizational and bureaucratic boundaries (38).

### Conclusions

The communication in health crises is a critical component of the emergency preparedness planning, regarding unexpected and devastating events, requiring urgent actions in a narrow time span and is thus a challenging task for any welfare state. Ultimate implication of the crisis communication planning is that the public has the right to make informed choices after actively involved in risk decisions implementation.

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### References

1. Fearn-Banks K. Crisis communications. 2nd ed., Mahwah, NT: Lawrence Erlbaum Associates, Inc; 2002. ISBN-13: 978-0805836042.
2. Seeger MW, Sellnow TL and Ulmer RR. Communication and organizational crisis. Westport (CT): Praeger 2003.
3. Seeger M, Pechta L, Price S, Lubell K, Rose D, Sapru S, et al. A Conceptual Model for Evaluating Emergency Risk Communication in Public Health. *Health Security*. 2018; 16(3):193-203.doi: 10.1089/hs.2018.0020.
4. Reynolds B. Crisis and Emergency Risk Communication; September 2002. Centers for Disease Control and Prevention.
5. Veil SR, Reynolds B, Sellnow TL and Seeger MW. CERC as a theoretical framework for research and practice. *Health Promotion Practice* 2008; 9(4):26-34. Available from: <https://doi.org/10.1177/1524839908322113>.
6. Spence P, Lachlan K and Griffin D. Crisis communication, race, and natural disasters. *Journal of Black Studies* 2007; 37: 539-54. Available from: <https://doi.org/10.1177/0021934706296192>
7. Ulmer R, Sellnow T and Seeger M. *Effective Crisis Communication: Moving from Crisis to Opportunity*, Thousand Oaks, CA: Sage, 2007. ISBN 9781412914192.
8. Sandman PM. *Responding to Community Outrage: Strategies for Effective Risk Communication*; American Industrial Hygiene Association 1993, Fairfax, VA. ISBN 0-932627-51-X. Available from: <http://psandman.com/media/RespondingtoCommunityOutrage.pdf>.

9. Sellnow T and Vidoloff K. Getting crisis communication right. *Food Technology* 2009; 63(9):40-5.
10. Veil SR and Ojeda F. Establishing media partnerships in crisis response. *Communication Studies* 2010; 60(4):412-29. Available from: <https://doi.org/10.1080/10510974.2010.491336>.
11. Shari R, Veil R and Husted A. Best practices as an assessment for crisis communication. *Journal of Communication Management* 2012; 16(2):131-145. Available from: <https://doi.org/10.1108/13632541211217560>.
12. Novac A. Traumatic stress and human behavior. *Psychiatric Times* 2012; 18(4). Available from: <https://www.psychiatrictimes.com/view/traumatic-stress-and-human-behavior>.
13. Glik DC. Risk communication for public health emergencies. *Ann Rev Public Health* 2007; 28: 33-35. Available from: <https://doi.org/10.1146/annurev.pubhealth.28.021406.144123>.
14. Lee Y-I and Jin Y. Crisis Information Seeking and Sharing (CISS): Scale Development for Measuring Publics' Communicative Behavior in Social-Mediated Public Health Crises. *Journal of International Crisis and Risk Communication Research* 2019; 2(1):13-38. Available from: <https://doi.org/10.30658/jicrcr.2.1.2>.
15. Hill D. Why they buy. *Across the Board* 2003; 40(6):27-33.
16. Brehm SS, Kassir S and Fein S. *Social Psychology* 2005. 6th ed. Boston: Houghton Mifflin Co. ISBN-13: 978-0547126425.
17. Solso RL. *Cognitive Psychology* 2001. 6th ed. Boston: Allyn and Bacon. ISBN-13: 978-0205309375.
18. Sellnow TL, Ulmer RR, Seeger MW and Littlefield RS. *Effective risk communication: A message-centered approach*. New York 2009: Springer. ISBN 978-0-387-79727-4.
19. Wray R, Becker S, Henderson N, Glik D, Jupka K, Middleton S, et al. Communicating with the public about emerging health threats: lessons from the Pre-Event Message Development Project. *Am J Public Health* 2008; 98(12):2214-2222. Available from: <https://doi.org/10.2105/AJPH.2006.107102>.
20. Memish Z, Steffen R, White P, Dar O, Azhar EI, Sharma A, et al. Mass gatherings medicine: public health issues arising from mass gathering religious and sporting events (Review) *Lancet* 2019; 393: 2073-84. Available from: [https://doi.org/10.1016/S0140-6736\(19\)30501-X](https://doi.org/10.1016/S0140-6736(19)30501-X).
21. Murphy MW, Iqbal S, Sanchez CA and Quinlisk MP. Post-disaster health communication and information sources: the Iowa flood scenario. *Disaster Med Public Health Prep* 2010; 4(2):129-134. Available from: <https://doi.org/10.1001/dmp.2010-v4n2-hre10009>.
22. Hilyard KM, Freimuth VS, Musa D, Kumar S and Quinn SC. The vagaries of public support for government actions in case of a pandemic. *Health Aff (Millwood)* 2010; 29(12):2294-2301. Available from: <https://doi.org/10.1377/hlthaff.2010.0474>.
23. Savoia E, Lin L and Viswanath K. Communications in Public Health Emergency Preparedness: A Systematic Review of the Literature Biosecurity and Bioterrorism: Biodefense Strategy, Practice, and Science 2013; 11(3). Available from: <https://doi.org/10.1089/bsp.2013.0038>.
24. [Author unknown]. Checklist for planning a national risk communication strategy. Pan American Health Organization 2014. Available from: <https://www.paho.org/hq/dmdocuments/2014/2014-cha-checklist-risk-comm-strategy.pdf>.
25. WHO. Communicating risk in public health emergencies: A WHO guideline for emergency risk communication (ERC) policy and practice 2017. World Health Organization. ISBN 978-92-4-155020-8. Available from: <https://www.who.int/risk-communication/guidance/download/en/>
26. [Author unknown]. Vaccination coverage among Adults in the United States, National Health Interview Survey, 2017. National Center for Immunization and Respiratory Diseases Available from: <https://www.cdc.gov/vaccines/imz-managers/coverage/adultvaxview/pubs-resources/NHIS-2017.html>
27. Taylor-Clark K, Viswanath K, Blendon R. Communication inequalities during public health disasters: Katrina's wake. *Health Commun* 2010; 25(3):221-229. Available from: <https://doi.org/10.1080/10410231003698895>.
28. Ma R. Media, crisis and SARS: An introduction. *Asian Journal of Communication* 2005; 15:241-6. Available from: <https://doi.org/10.1080/01292980500260656>.
29. Lariscy RW, Avery EJ, Sweetser KD and Howes P. An examination of the role of online social media in journalists' source mix. *Public Relations Review* 2009; 35:314-316. Available from: <https://doi.org/10.1016/j.pubrev.2009.05.008>.
30. Austin L, Liu B and Jin Y. How audiences seek out crisis information: Exploring the social-mediated crisis communication model. *Journal of Applied Communication Research* 2012; 40: 188-207. Available from: <https://doi.org/10.1080/00909882.2012.654498>.
31. Liu BF, Fraustino JD and Jin Y. How disaster information form, source, type, and prior disaster exposure affect public outcomes: Jumping on the social media bandwagon? *Journal of Applied Communication Research* 2015; 43:44-65. Available from: <https://doi.org/10.1080/00909882.2014.982685>.
32. Frewer L. The public and effective risk communication. *Toxicology Letters* 2004; 149: 391-7. Available from: <https://doi.org/10.1016/j.toxlet.2003.12.049>.
33. Vijaykumar S, Jin Y and Nowak G. Social media and the virality of risk: The risk amplification through media spread (RAMS) model. *Journal of Homeland Security and Emergency Management* 2015; 12: 653-677. Available from: <https://doi.org/10.1515/jhsem-2014-0072>.
34. Coombs T. *Ongoing crisis communication: Planning, managing and responding*. Thousand Oakes 1999; CA: Sage. ISBN13: 9781452261362.
35. Freberg K, Saling K, Vidoloff KG and Eosco G. Using value modeling to evaluate social media messages: The case of hurricane Irene. *Public Relations Review* 2013; 39:185-192. Available from: <http://dx.doi.org/10.1016/j.pubrev.2013.02.010>.
36. Smith BG and Gallicano TD. Terms of engagement: Analyzing public engagement with organizations through social media. *Computer in Human Behavior* 2015; 53: 82-90. Available from: <http://dx.doi.org/10.1016/j.chb.2015.05.060>.
37. Liu BF, Fraustino JD and Jin Y. Social media use during disasters: How information form and source influence intended behavioral responses. *Communication Research* 2016; 43:626-646. Available from: <http://doi.org/10.1177/0093650214565917>.
38. WHO. Summary report of systematic reviews for public health emergency operations centres. Plans and procedures; communication technology and infrastructure: minimum datasets and standards; training and exercises. World Health Organization 2015; ISBN 978 92 4 150978 7.



## Experimental model for the evaluation of metabolic status after gastric bypass in obese rats

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### Abstract

**Objective:** Obesity is an exceedingly current pathology with many clinical, molecular, and psychological implications. The number of obese people has doubled in the past ten years, and we can observe an early onset of obesity. Bariatric surgery is an effective treatment for severe obesity and type 2 diabetes mellitus (T2DM); Roux-en-Y gastric bypass (RYGB) and sleeve gastrectomy (SG) are the two most prevalent types of this procedure. Nevertheless, no single mechanism has emerged that thoroughly explains the metabolic benefit and subsequent long-term effects after surgery. Webster's new collegiate dictionary defines the noun model as: 'a descriptive or analogy used to help visualize something that cannot be directly observed.' Sustained by this definition, the animal models in nutritional research are fundamental to improve human conditions. Due to the recent boost in experimental surgery, our aim in this study is to set the main technical characteristics of the gastric bypass operation and specific animal care in the metabolic surgery field we have been undertaking in our center.

**Materials and methods:** We chose Wistar rats fed with a high caloric diet (HCD) 82g / 100 g fat, 53/100 g saturated fat, 64/100 g carbohydrates. After 7-15 weeks of this diet, depending on the protocol understudy, a consistent three-fold greater weight gain is achieved than the usual range of the free eating chow. After approximately three months on an HCD, the obese rats manifest biochemical features of the metabolic syndrome.

**Results:** The weight loss for group B (By-pass) was  $125 \pm 16.16$  g, and for group BS (By-pass + Sulodexid), it was  $133.10 \pm 14.38$  g. Although the weight loss was higher in group BS, it is not statistically significantly higher than in group B ( $p = 0.345$ ), despite administering a pharmacologically active substance in group BS. Although small (approximately 40-50 g), the difference between the control group and groups B and BS is statistically significant with  $p = 0.016$  and  $0.026$  and Pearson index of 0.674 and 0.628, respectively. Statistical significance also kept the difference between group C and group S ( $p = 0.028$ , Pearson Coefficient = 0.621).

**Conclusions:** First of all, Metabolic surgery is the most effective weight-loss method and improvement or even remission of some diseases associated with obesity. Like today's high-calorie diet, the diet administered, mostly of adolescents, generates both obesity and its associated diseases: diabetes, hypertriglyceridemia, hypercholesterolemia, thus increasing mortality and overall morbidity. Second, metabolic surgery radically improves the parameters targeting obesity (weight, % EBWL) and its associated conditions: diabetes mellitus, hypertriglyceridemia, hypercholesterolemia, strongly associated with decreasing life expectancy of the general population. Parameters targeted by gastric bypass: glycemia, TGL, CHO, hepatic steatosis, testicular atrophy registering significant improvements.

**Keywords:** Gastric bypass, Obesity, Hypercaloric nutrition

### Introduction

The worldwide pandemics of obesity and diabetes are devastating in severity, extent, and rate of growth. Over two billion adults worldwide, approximately 30% of the population are either overweight (body mass index (BMI) > 25) or obese (BMI > 30)(1).

Obese patients have substantially increased morbidity and mortality from obesity-related complications, including type 2 diabetes mellitus (T2DM), cardiovascular disease, and several types of cancer (2).

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In addition to obesity in the adult population, the obesity rate amongst children and adolescents in the USA has doubled during the past two decades. Currently, 17% of children aged 2–19 years are obese (3). If this trend continues, obesity and its associated co-morbidities threaten the health of future generations. Given the severity of the obesity pandemic, there is an urgent need for a detailed understanding of the mechanisms underlying the pathophysiology of obesity to enable effective therapy.

There is a limited understanding of the precise mechanisms by which RYGBP induces and maintains substantial weight loss. In continuous numerical growth, evidence for this hypothesis suggests that profound changes in body weight and metabolism cannot be explained by simple mechanical restrictions or by malabsorption techniques of gastric bypass. (4, 6). Several groups have shown that gastric bypass affects hunger and increases or accelerates the experience of satiety after meals. Gastric bypass has been shown to alter the release of various gastrointestinal hormones, including ghrelin, GLP-1, YY peptide, and glucose-dependent insulintropic peptide, generating an endocrine response that results in a drastic reduction in appetite and early postprandial satiety. (5).

In the absence of effective non-invasive treatments, bariatric surgery remains the only practical option that can lead to long-term sustained weight loss (6–8). In RYGB (Fig. 1a), the stomach is divided into two sections, creating a small gastric pouch (about 1–2% of total gastric volume) (9,10). The small intestine is also divided and rearranged to create Y-shaped anatomy, causing the food to bypass the more extensive section of the stomach and upper intestine, restricting stomach capacity and proper intestine length. Although the development of laparoscopic methods has substantially reduced the mortality and complications associated with RYGB, it is still a complex operation that can lead to postoperative complications (about 0.5% mortality rates), after which patients require intensive postoperative care and life-long nutritional management(11-13).

Another view on adipose tissue and obesity was due to research that led to the discovery of leptin and the identification of adiponectin, and later, after two years and other protein structures specific to adipose tissue both visceral and subcutaneous, having hormonal activity, both autocrine and paracrine. It is well known today that adipose tissue has an extraordinarily complex and insufficiently documented role(14).

What is the future of adipose tissue as an endocrine organ? This is a question that not only endocrinologists but also metabolic surgeons ask themselves. Several hormones derived from adipose tissue have been identified, but even those factors that are relatively well known, such as leptin, require further evaluation to precisely define their physiological roles. In addition to known genes, up to 40% of genes expressed in adipose tissue are newly discovered genes, and 20-30% of them can secrete proteins. Ongoing research into these new genes aims to reveal a more in-depth perspective on adipose tissue's endocrine function and the relationship between energy homeostasis and other physiological systems (14-16).

Further studies are also needed to clarify the contribution of adipose tissue's individual cellular components to determine how these components function as a unit. Finally, understanding the endocrine function of adipose tissue is likely to allow for more rational approaches to treating the metabolic consequences of excess and deficiency of fatty tissue. (17)

## Material and Methods

Experimental design included a number of 40 Wistar rats divided into four lots (exemplified below). They were fed for 12-16 weeks with a high-calorie diet (with a very high energy value) consisting of 82g / 100 g fat, 53/100 g saturated fat, 64/100 g carbohydrates; they were practically fed 82% butter and carbohydrate-enriched chocolate to reach the stage of obesity, weighing over the 75th percentile of age. These were added discretionary feeding with rat granules and water ad libitum with 35 g sugar / 100 g water. Thus, an attempt was made to simulate a hyperlipidemic hyperglycemic diet found today in the daily diet of the population that consumes fast food and sweet carbonated drinks. The exception to this rule is the control group throughout the experiment, the regular diet specific to laboratory animals with grains and water ad libitum. Thus, we tried to simulate a hyperlipidemic hyperglycemic diet that is found today in the daily diet of the population that consumes fast food and sweet carbonated drinks. Subsequently, they underwent gastric bypass surgery. They were kept permanently in individual cages at a temperature of  $21 \pm 3$  degrees Celsius and 45-60% humidity, with specific water and granules for ad libitum feeding and 3-5 hypercaloric meals (described above) per day (except for the control group). (Figure 1). Reaching the weight of over 75th percentile (declared obese), those in groups 2 and 4 were operated on. At the time of surgery, blood samples were collected to analyze the above parameters. After 28 days, they were sacrificed, at which time biological samples were collected again, intraperitoneal characters were studied, and organs were collected for further study.



**Figure 1.** Rats in individual cages

Anesthesia and pain control was performed, following a pre-weighing, by weight, with an anesthetic mixture of

Acepromazine (0.2 ml/kg) and Ketamine (100mg / kg) administered intramuscularly. If necessary, during the surgery, 0.1 ml of Ketamine was administered for better anesthetic control. The sulodexide solution provided by Alfa Sigma Company was administered by intramuscular injection.

Before the day of surgery, 18-24 hours, the rats were deprived of oral nutrition, being only hydrated with water ad libitum. The equipment consists of: a scalpel, anatomical and surgical forceps, 2 Pean forceps, 2 Kocher forceps, a dissection scissors, and a needle holder. The sutures used were polyglactin 4-0 (Vicryl) for anastomoses and polypropylene or polyglactin 3-0 or 4-0 for layer closure.



**Figure 2:** Peritoneal cavity inspection with identification of anatomical landmarks: 1- stomach, 2- jejunum, 3- ileum, 4- cecum, 5- transverse colon.

The operating device included The rat being fixed to the heated operating table, with the abdomen prepared for the incision. Sterile operating field Autoclave or bead sterilize instruments before use. Prepare an aseptic operative field by covering the operation table with a sterile drape. Place the sterile surgical instruments and a heating pad on the table. Turn on the heat pad and set the temperature to 37 °C. Cover the heating pad with a sterile drape. Set up the rodent anesthesia machine.

The sterile operating field is followed by laparotomy on a length of about 3 cm (Laparotomy: skin incision - sectioning of the right abdominal muscles - penetration into the peritoneal cavity without damaging the internal organs). After entering the peritoneal cavity, it is inspected, and the targeted anatomical elements are identified (Figure 2).

A gastric reservoir of approximately 1/4 of the total surface of the stomach is made by pinching and sectioning it with scissors, After a minimal dissection at the small gastric curvature level and the Hiss angle to enter the retro gastric space. (Figure 3)



**Figure 3:** Closing with a 3-0 or 4-0 thread in the surget of the remaining stomach

Close the remaining stomach with a thread in the thigh as in the image below and then proceed to prepare the jejunal loop to be anastomosed to the gastric trench of the gastric reservoir. The small intestine is sectioned approximately 16 cm from Treitz's angle, this being the biliopancreatic loop. Hoffmaster-Finsterer T-L gastro-jejunal anastomosis is performed with two 4-0 threads, one for the posterior layer and one for the anterior layer. (figure 3) The jejune-jejunal anastomosis is made at about 25 cm from the gastrojejunal anastomosis, thus remaining for the food loop 25 cm and the common one around 20-25 cm. Hemostasis is checked and supplemented, where necessary (Figure 4,figure 5 and figure 6 A and B).

Parietoraphy is done on anatomical layers with 3-0 threads in the surget. (Figure 7) Wash the wound and leave the rat to wake up spontaneously from anesthesia. The temperature and pulse are monitored continuously throughout the operation.





**Figure 4:** Preparation and sectioning of the small intestine after measuring 16 cm of biliary-pancreatic loop.



**Figure 5:** Beginning of gastrojejunal anastomosis: 1- biliary pancreatic loop, 2- food loop, 3- gastric reservoir.



**Figure 6A:** The final appearance of gastrojejunal anastomosis



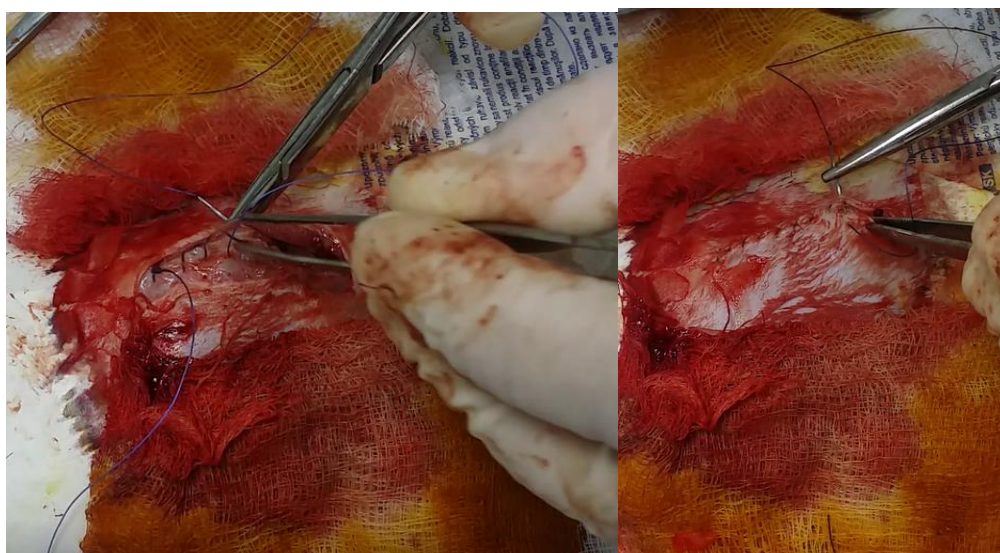
**Figure 6B:** Final appearance of jejunio jejunal anastomosis: 1- biliopancreatic loop, 2- food loop, 3- common loop

The average duration of such an intervention was around 55-70 min. The first 48 hours postoperatively, rehydration fluids are administered without resuming a solid oral diet.

For all rats, when the intestinal transit for feces is resumed, the appearance of possible postoperative complications is followed (the most frequently cited in the literature are the digestive fistula and the thromboembolic complications).

The subjects are 28 days after the operation period corresponding to 10-14 months in human subjects. After the anesthetic induction, which is done according to the same protocol mentioned above, the median laparotomy is performed with penetration into the abdominal cavity where the cavity is inspected, and the volume is evaluated, the quality of parietal-visceral and visceral-visceral adhesions and neovascularization is assessed on a scale specific to each rat (using the van der Ham scale - see table below: Table 1).

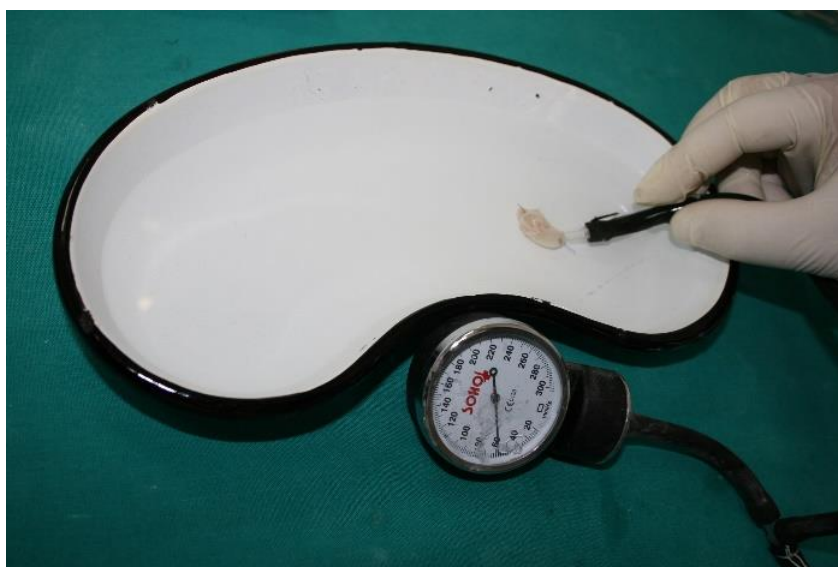
After adhesiolysis is performed, such a rating is also given for the appearance and quality of the anastomosis, according to the table below (Table 1.).



**Figure 7:** Anatomical parietorrhaphy with two resorbable threads in the surjet

**Table 1:** Table of quantitative and qualitative evaluation of intraperitoneal adhesions and the appearance of anastomoses

Scala van der Ham														
Post operative day	GJ anastomosis				Adesiolysis			Neovascularisation			JJ anastomosis			
	-	+	++	+++	Less (<3)	Moderate (3-5)	Many (>5)	+	++	+++	-	+	++	+++
28														



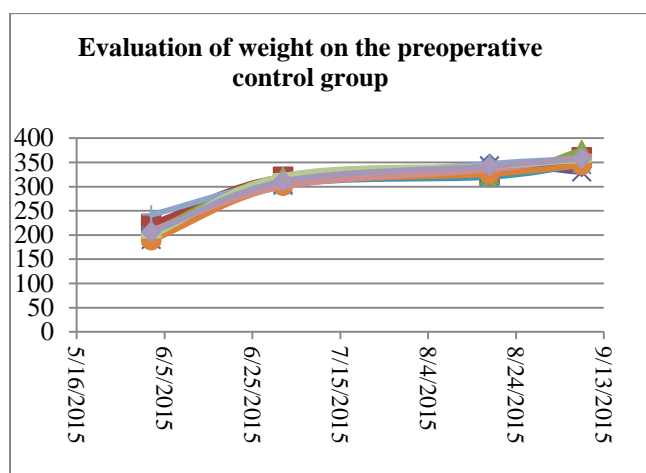
**Figure 8:** Immersion testing of anastomosis resistance to pressure.

After completing the previous stages, the resection of the digestive tract segment containing the gastro-jejunal anastomosis with a safety margin of approximately 5-10 mm is performed, considering that at this level is the highest risk of anastomotic fistula. Determine the pressure that the anastomosis withstands with a device like the one in the adjacent figure.

The determination of this pressure is performed by immersion (see figure 8) with the help of a blowing pair and a manometer that records the pressure at which intestinal dehiscence occurs (rupture of the anastomosis). After removing excess tissue, intestinal anastomosis is taken in containers with PFA / formalin to make histology slides in trichrome hemalaun-eosin-methylene blue to assess the degree of fibrosis and granulation tissue.

## Results

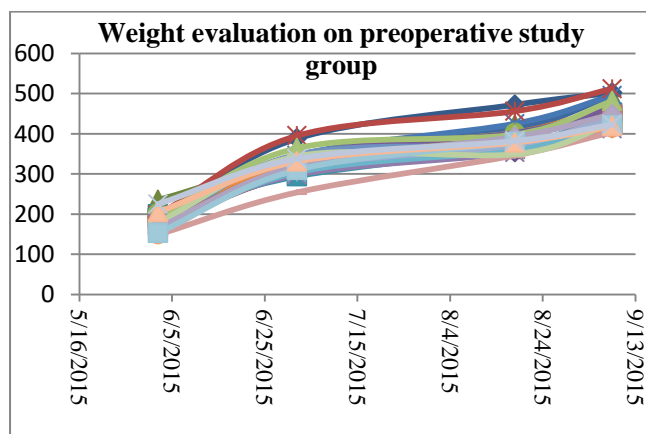
The evolution of the weights preoperatively shows an essential increase in the first interval followed by a certain plateau. In the last interval of time, adding another addition is not so significant as in the first time interval (figure 9).



**Figure 9:** Evolution in time of weight (gr) in preoperative control group.

In the case of the study groups, the rats fed with a hypercaloric diet are observed a constant ascending weight gain, the most significant evolution being also in the first time interval. The difference between the average weights in the control group compared to the study groups is statistically significant, making the diet presented in the methodology chapter an experimental model of obese Wistar rats on which various aspects can be studied. The table below shows a difference of up to 162 grams between the control group and the study groups with a hypercaloric diet, the average being  $98\text{g} \pm 7.2$  (Mean  $\pm$  StDev) between them. The difference is statistically significant ( $p < 0.05$ ).

As shown in the graph and table below, there is a statistically significant difference between the mean weight in the control group and B, BS, and S (values expressed as Mean  $\pm$  StDev). I am having the importance of p and Pearson's coefficient described in the table below. The most significant difference being between the control group and the S group (figure 10).

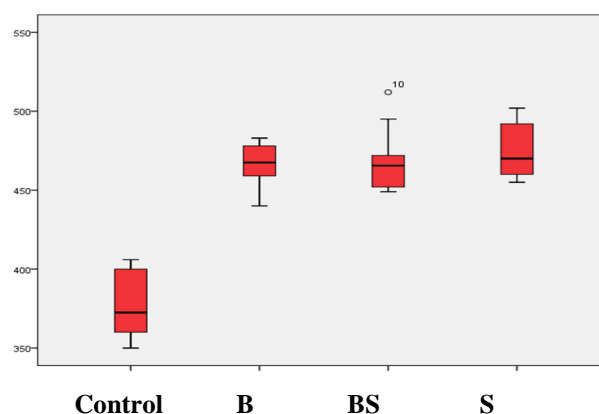


**Figure 10:** Evolution in time of preoperative study groups in weight parameter (gr)

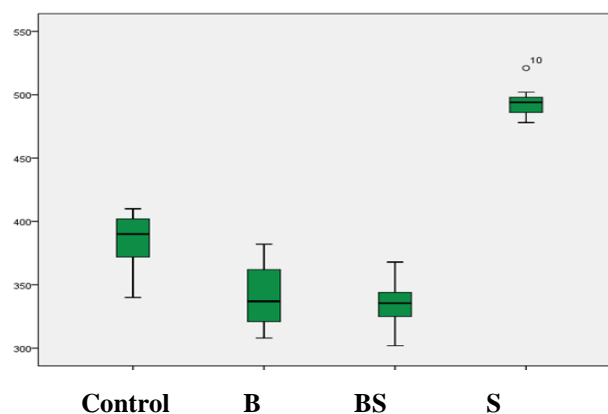
Postoperatively, groups B and BS's average weight decreases significantly; their values are below the control group (see table below).(figure 12, figure 13) In contrast, the importance of group S increased by about 19g on average. No statistical significance is reported between groups B and BS in postoperative weight, although group BS has a lower average than in group B. However, there is a statistically significant difference (almost 160g) between B, BS, and group S postoperative.(table 2)

Although small (approximately 40-50 g), the difference between the control group and groups B and BS is statistically significant with  $p = 0.016$  and  $0.026$  and Pearson index of  $0.674$  and  $0.628$ , respectively. Statistical significance also kept the difference between group C and group S ( $p = 0.028$ , Pearson Coefficient =  $0.621$ ). (table 2, table 3)

We notice that the weight loss for group B (By-pass) was  $125 \pm 16.16$  g, and for group BS (By-pass + Sulodexid), it was  $133.10 \pm 14.38$  g. Although the weight loss was higher in group BS, it is not statistically significantly higher than in group B ( $p = 0.345$ ), despite administering a pharmacologically active substance in group BS. (figure 11)



**Figure 11:** Preoperative weight on each batch (Mean  $\pm$  StDev). **C:** Control  $376.30 \pm 19.71$  gr, **B:**  $465.50 \pm 12.88$  gr **BS:**  $468.80 \pm 20.32$  gr **S:**  $474.70 \pm 16.93$  gr



**Figure 12:** Chart with postoperative weights at 28 days (Mean  $\pm$  StDev). **C:** Control:  $383.40 \pm 24.30$  gr, **B:**  $342.16 \pm 24.96$  gr **BS:**  $335.70 \pm 18.60$  gr **S:**  $493.90 \pm 12.29$  gr

**Table 2:** Evolution and correlations of pre- and postoperative weight differences.

Group (n)	Preoperative Weight (gr)	Postoperative Weight (gr)	P value	Pearson	Average pre / postop difference	The significance of the difference
<b>C (10)</b>	376.30±19.71	383.40±24.30	<b>0.05*</b>	0.421	-7.1±17.05	0.652
<b>B (12)</b>	465.50±12.88	342.16±24.96	<b>0.012*</b>	0.691	125.4±16.16	<0.001
<b>BS (10)</b>	468.80±20.32	335.70±18.60	<b>0.008*</b>	0.730	133.1±14.38	<0.001
<b>S (10)</b>	474.70±16.93	493.90±12.29	<b>0.001*</b>	0.907	-19.2±7.75	<0.05

\* It was considered statistically significant p &lt;0.05

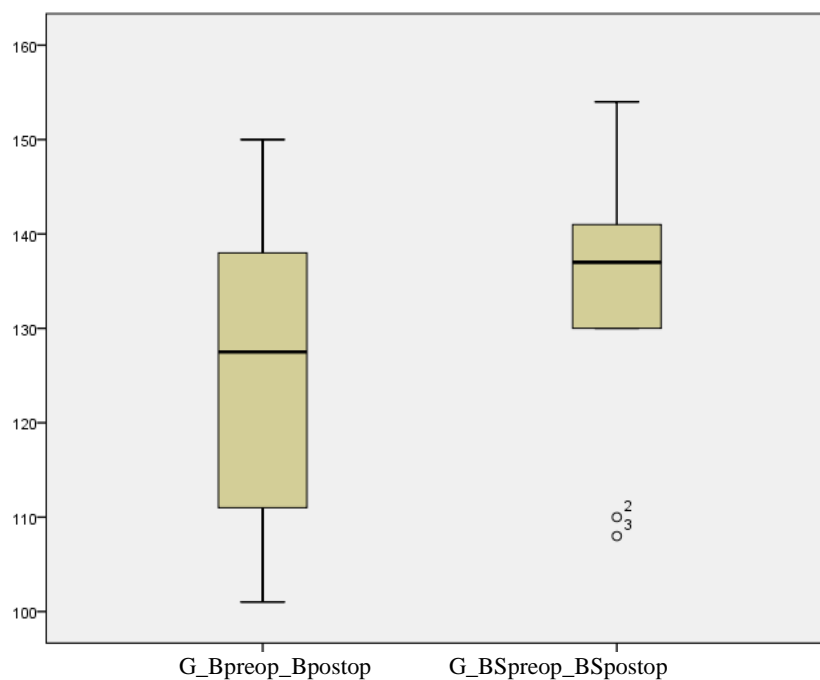
**Table 3:** Correlations between batches - for the Weight parameter

	C_post-op.	B_post-op.	BS_post-op.	S_post op.
<b>C_pre-op.</b>	p=0.01* Coef Pearson = 0.719	p=0.013* Coef Pearson = 0.696	p=0.132 Coef Pearson = 0.390	p=0.013* Coef Pearson = 0.695
<b>B_pre-op.</b>	p=0.014* Coef Pearson = 0.691	p=0.016* Coef Pearson = 0.620	p=0.046* Coef Pearson = 0.561	p=0.154 Coef Pearson = 0.360
<b>BS_Pre-op.</b>	p=0.074 Coef Pearson = 0.492	p=0.035* Coef Pearson = 0.595	p=0.008* Coef Pearson = 0.730	p=0.033* Coef Pearson = 0.602
<b>S_pre-op.</b>	p=0.027* Coef Pearson = 0.624	p=0.102 Coef Pearson = 0.439	p=0.043* Coef Pearson = 0.569	p=0.001* Coef Pearson = 0.907

\* It was considered statistically significant p &lt;0.05

**Table 4:** Correlations of pre- and postoperative weight differences for groups B and BS

		G_Bpreop_Bpostop	G_BSpreop_BSpostop
G_Bpreop_Bpostop	Pearson Correlation	1	-.144
	Sig. (1-tailed)		.345
	Sum of Squares and Cross-products	2352.400	-302.400
	Covariance	261.378	-33.600
	N	10	10
G_BSpreop_BSpostop	Pearson Correlation	-.144	1
	Sig. (1-tailed)	.345	
	Sum of Squares and Cross-products	-302.400	1862.900
	Covariance	-33.600	206.989
	N	10	10

**Figure 13:** Pre- and postoperative weight difference for groups B and BS (Mean ± StDev)



## Discussion

Regarding the diet administered to rats to induce intake obesity, Vincent E. de Meijer et al. showed that a diet consisting of 60% fat can cause an increased level of the fat index and can generate obesity; it can also induce thus insulin resistance and hepatic steatosis administered for a long time, diseases directly related to obesity. (15-18) The diet administered in our research was achieved by combining a hypercaloric, hyperlipidemic diet, cited in the literature, with a diet present today in most of the population in medium and well-developed countries, a diet rich in high-calorie foods, sweets, and sweet carbonated drinks. It contained 82g / 100g fat, 53 / 100g saturated fatty acids, 64 / 100g carbohydrates, were practically fed with 82% butter and carbohydrate enriched chocolate to reach the stage of obesity. It is considered at weight over 75 percentile according to age. These were added a discretionary diet with granules for rats and water ad libitum enriched with 35g of sugar / 100g of water (similar to the carbohydrate addition from carbonated drinks). In this way, we tried to simulate as well as possible the conditions in a society where the consumption of fast food and sweet carbonated drinks is continuously increasing (19).

The administered diet determined the appearance of obesity in the study rats, the obesity being considered the weight over the 75th percentile of age. The statistically significant difference in food intake in the study groups compared to the control group that received regular feeding was up to 162 grams, more than half the rats' body weight in the control group. Data from the literature abound in various diets with similar results, important being the final weight at which the recordings are made. (20) It is known that a patient, or in our case, a laboratory subject if he is obese for a significant period, will also develop diabetes in direct relation to obesity induced by food intake. (21)

Postoperatively, the effects of surgery are observed for the weight and blood glucose values and the TGL and CHO values, all registering statistically significant values lower than the preoperative values on the respective groups. The difference is both between pre- and postoperative moments and between groups B, BS, and group Control (C). In addition to the surgery that is the determining factor generating these differences, the rats in the study groups returned to regular feeding. After surgery, without a hypercaloric/hyperlipidemic diet and carbohydrate supplementation in the food consumed (22-23).

Given that laboratory animals do not have the same parameters for assessing postoperative weight status, After a thorough study of the literature, we chose to replace the variable% EBWL, which we can not calculate according to the formula given to rats, not having their height as a well-defined variable, with pre- and postoperative weights difference at 28 days, respectively (22).

Weight loss was the most important in the case of the BS group. In group B, the weight loss, although very significant, did not reach the value of group BS. We can thus suspect that the difference between groups B and BS in terms of postoperative weight loss, the difference in favor of group BS, is due to sulodexide administration, a

fact not yet confirmed in the literature. Since sulodexide does not have a role in weight loss, we can consider that its implication in tissue remodeling by changing the status of MMPs is what determines that increase in a weight loss of the BS group. The involvement of sulodexide in MMP metabolism is also demonstrated by Mannello et al. in his study published in 2013. (23)

A meta-analysis with an extremely strong scientific impact published by Ribaric, Buchwald and McGlennon in 2014 in a reference journal for the medical scientific community conducted on a number of 6131 patients with a mean follow-up of  $17.3 \pm 5.7$  months revealed 9.8 to 15.8 times the efficiency of bariatric/metabolic surgery compared to conventional therapy in terms of weight loss, reduction of TGL, CHO, HbA1C (glycosylated hemoglobin) and fasting blood glucose. These data implicitly lead to the conclusion of the effectiveness of the surgical treatment of obesity for weight loss and remission of type II diabetes (18).

## Conclusions

First of all, metabolic surgery is the most effective weight-loss method and improvement or even remission of some diseases associated with obesity.

Like today's high-calorie diet, the diet administered, mostly of adolescents, generates both obesity and its associated diseases: diabetes, hypertriglyceridemia, hypercholesterolemia, thus increasing mortality and overall morbidity.

Second, metabolic surgery radically improves the parameters targeting obesity (weight, % EBWL) and its associated conditions: diabetes mellitus, hypertriglyceridemia, hypercholesterolemia, strongly associated with decreasing life expectancy of the general population. Parameters targeted by gastric bypass: glycemia, TGL, CHO, hepatic steatosis, testicular atrophy registering significant improvements.

**Author Contributions:** Radu Mirica, Alexandra Mirica, Octav Ginghina, Razvan Iosifescu, Andrei Vacarasu, Claudiu Ungureanu, Nicolae Iordache: Project design, Review of the literature, Data collection **Radu Mirica:** Writing and Revisions

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**Ethical issues:** All authors declare originality of research.

## References

1. Boden G, Song W, Pashko L, K. K. (2008) 'In vivo effects of insulin and free fatty acids on matrix metalloproteinases in rat aorta', *Diabetes*, 57(476).
2. Bouloumié, A. et al. (2001) 'Adipocyte Produces MMP2 and 9 Involvement in Adipose Differentiation', *Diab*, 50(9), pp. 2080–2086.
3. Booth, H. et al. (2014) 'Incidence of type 2 diabetes after bariatric surgery: Population-based matched cohort study', *The Lancet Diabetes and Endocrinology*. Booth et al. Open Access article distributed under the terms of CC BY, 2(12), pp. 963–968. Broekhuizen, L. N. et al. (2010) 'Effect of sulodexide on glycocalyx and vascular permeability in patients with type 2 diabetes', *Diabet*, 53(12), pp. 2646–2655.

4. Mirica, R. M. et al. (2017) 'Quality of Life Assessment After Metabolic Surgery — a Single-Center Experience'. *Indian Journal of Surgery*.
5. Mirica, R. M. et al. (no date) 'The Important Roles of Matrix MMP in the Pathophysiology of Obesity', pp. 1481–1484.
6. Mingrone, G. et al. (2015) 'Metabolic surgery versus conventional medical treatment in obese patients with type 2 diabetes: 5 Year follow-up of an open-label, single-centre, randomised controlled trial', *The Lancet*. Elsevier Ltd, 386(9997), pp. 964–973.
7. Nickel, F., Schmidt, L., Bruckner, T., Billeter, A. T., et al. (2017) 'Gastrointestinal Quality of Life Improves Significantly After LSG and RYGB—a Prospective Cross-Sectional Study Within a 2-Year Follow-up', *Obesity Surgery*. *Obesity Surgery*, 27(5), pp. 1292–1297.
8. Nickel, F., Schmidt, L., Bruckner, T., Büchler, M. W., et al. (2017) 'Influence of bariatric surgery on qol, body image, and general self-efficacy within 6 and 24 months—a prospective study', *Surgery for Obesity and Related Diseases*. Elsevier, 13(2), pp. 313–319.
9. Nicolae Angelescu, F. P. (2010) *Caiet de tehnici chirurgicale*.
10. Noel P, Nedelcu M, Nocca D, Schneck AS, Gugenheim J, Iannelli A, G. M. (2014) 'Revised sleeve gastrectomy: an option for weight loss failure after LSG', *Surg Endosc*, 28(4), pp. 1096–1102.
11. POPESCU, L.A., VIRGOLICI, B., STEFAN, D.C.A., LIXANDRU, D., TIMNEA, O., VIRGOLICI, H., ORASEANU, D., SINESCU, C., MOHORA, M. (2017) 'Vascular Reactivity and Proinflammatory Cytokines in the Obese Children', *Rev. Chim.(Bucharest)*, 68(4), pp. 758–768.
12. Popov Y, S. D. (2009) 'Targeting liver fibrosis: strategies for development and validation of antifibrotic therapies', *Hepatology*, 50: 1294–3.
13. Qin DD, Yuan W, Z. W. (2007) 'Do reproductive hormones explain the association between body mass index and semen quality?', *Asian J Androl*, 9, pp. 827–34.
14. Regan JP, Inabnet WB, Gagner M, P. A. (2003) 'Early experience with two-stage laparoscopic RYGB as an alternative in the super obese patient.', *Obes Surg.*, 13(6), pp. 861–864.
15. Röss, C. et al. (2010a) 'Influence of significant weight loss on serum MMP-7 levels', *European Cytokine Network*, 21(1), pp. 65–70.
16. Röss, C. et al. (2010b) 'Influence of significant weight loss on serum MMP-7 levels', *European Cytokine Network*, 21(1), pp. 65–70.
17. Reynolds, C. L., Byrne, S. M. and Hamdorf, J. M. (2017) 'Treatment Success: Investigating Clinically Significant Change in QoL Following Bariatric Surgery', *Obesity Surgery*. *Obesity Surgery*, 27(7), pp. 1842–1848.
18. Ribaric, G., Buchwald, J. N. and McGlennon, T. W. (2014) 'Diabetes and weight in comparative studies of bariatric surgery vs conventional therapy: A review and meta-analysis', *Obesity Surgery*, 24(3), pp. 437–455.
19. Sacks, J. et al. (2018) 'Effect of RYGB on liver mitochondrial dynamics in a rat model of obesity', *Physiological Reports*, 6(4), p. e13600.
20. Salminen, P. et al. (2018) 'Effect of LSG vs LRYGB on Weight Loss at 5 Years Among Patients With Morbid Obesity', *Jama*, 319(3), p. 241.
21. Santoro, S. et al. (2014) 'Sleeve gastrectomy with anti-reflux procedures', *Einstein (São Paulo)*, 12(3), pp. 287–294.
22. Serra, R. et al. (2014) 'The effects of sulodexide on both clinical and molecular parameters in patients with mixed arterial and venous ulcers of lower limbs', pp. 519–527.
23. Sierżantowicz R1, Lewko J2, Hady HR3, Kirpsza B1, Trochimowicz L3, D. J. (2017) 'Effect of BMI on quality of life and depression levels after bariatric surgery.', *Adv Clin Exp Med*, 26(3), pp. 491–496.

## Correlation between severity of diabetes mellitus, polyneuropathy and nasal mucociliary clearance

Mirac Aysen Unsal<sup>1\*</sup>, Suphi Bulgurcu<sup>2</sup>

### Abstract

**Objective:** To examine the relationship between the severity of polyneuropathy and nasal mucociliary clearance times in patients with polyneuropathy and investigate how the presence and severity of electrophysiological polyneuropathy might affect mucociliary clearance in patients with diabetes mellitus.

**Material and Methods:** This prospective cross-sectional study was carried out in the Neurology and Ear, Nose and Throat (ENT) clinics in a tertiary hospital. The study included three groups of patients with 20 participants in each group (Group 1, patients with diabetic poly-neuropathy; Group 2, patients with non-diabetic polyneuropathy; and Group 3, diabetes mellitus patients with a normal nerve conduction study) Nasal saccharin test was performed on all patients.

**Results:** There was a statistically significant difference in the duration of nasal mucociliary clearance among the groups ( $p = 0.001$ ). There was a positive, statistically significant ( $p = 0.007$ ) correlation between the nasal mucociliary clearance duration and the severity of poly-neuropathy. The nasal mucociliary clearance duration increased with the severity of polyneuropathy.

**Conclusion:** Patients with diabetes mellitus are a special group, and preventable problems should be taken into consideration when examining nasal pathologies. It should be kept in mind that nasal mucociliary clearance dysfunction can be both a cause and a result in management of diabetes mellitus patients, and thus it should be evaluated carefully.

**Keywords:** nasal mucociliary clearance, saccharin test, diabetes mellitus, diabetic polyneuropathy

### Introduction

Effective ciliary activity and regularly regenerated airway surface liquids are required for the respiratory mucosal surfaces to remove accumulated foreign particles. Being essential for normal physiology, these two features are commonly referred to as mucociliary activity. The combined effect of the mucus and ciliary system is evaluated by mucociliary clearance measurement. And, mucociliary clearance (MC) can be determined by measuring the elimination time of inhaled aerosols (1)

Various environmental, metabolic and pharmacological factors have an effect on the mucociliary activity, including mucosal dryness, smoking, room temperature, hypoxia, hypercarbia, dehydration, pH changes, cystic fibrosis, primary ciliary dyskinesia, pharmacological agents such as phenylephrine, epinephrine, lidocaine, atropine, antihistamines, bacterial and viral infections, and allergic rhinitis. The abnormal ciliary function causes chronic upper and lower respiratory tract infections (2-4).

Assessment of mucociliary function started in the 1830s, and migration of various substances placed on the nasal mucosa was examined by means of direct observation. Saccharin test is one of the methods used for this measurement (5-7). The circulation of the nasal airway is mediated by the autonomic nervous system. There are sympathetic and parasympathetic fibers of the autonomic system in the nose (8). Previous studies reported that nasal mucociliary clearance (NMC) in people with diabetes mellitus (DM) differed from the normal population due to dryness in the nasal membrane, increased nasal mucous viscosity, and abnormalities in small vessels (9, 10). We think that autonomic involvement of diabetic and non-diabetic polyneuropathies may also be a prominent factor in the disruption of the MC. The present study aimed to evaluate how NMC is affected by the presence of electrodiagnostic polyneuropathy in patients with DM, and compare the severity of diabetic and non-diabetic polyneuropathy to that of nasal mucociliary dysfunction.

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## Materials and Methods

### Patients

This prospective cross-sectional study was carried out in the Neurology and Ear, Nose, and Throat (ENT) clinics in a tertiary hospital. Before the study, an approval was obtained from the local Ethics Committee as well as written informed consent from each participant.

The study included three groups of patients, homogenous in terms of age and gender, with 20 participants in each group (Group 1, patients with Diabetic Polyneuropathy (DPN), DM patients with abnormal nerve conduction study; Group 2, patients with non-diabetic polyneuropathy (N-DPN); and Group 3, DM patients with a normal nerve conduction study)

A detailed history was taken from all patients. Data on duration of DM and the presence of peripheral neuropathy were collected. A detailed general physical examination and neurologic examination were performed and documented by the same physicians. Nasal endoscopy was performed with a complete ENT examination and was evaluated in detail for nasal pathology. Patients who had upper respiratory tract infection, nasal surgery, septum deviation, concha hypertrophy, chronic otitis media, nasal discharge and nasal polyps and patients who were smoking and using pharmacological agents that would affect ciliary activity were excluded. Patients with systemic diseases that might have affected ciliary activity were also excluded.

Saccharin test was done at least 3 hours after fasting and between 10-12 a.m. so that other factors such as coffee drinking, nutrition and circadian rhythm did not affect the test result.

### Saccharin test

The saccharin test has been described by Andersen and modified by Rutland and Cole (6,7). MC measurement was performed for all patients and the control group by the saccharin test. The test was carried out at a room temperature with the patient seated and his/her head in the upright position. First of all, the patient was asked to clear the secretions in his/her nose. A 1x1x1 millimeter (1/4 saccharin tablet) size saccharin tablet was placed 1-centimeter posterior from the anterior border of the inferior turbinate medial surface in one of the nasal cavities with the help of port cotton. The patient was asked not to sneeze, sniff, and bend his/her head forward. The patient was instructed to tell the moment when (s)he felt the taste, which was recorded as the NMC time.

### Evaluation of the severity of polyneuropathy with electromyography (EMG)

All patients underwent nerve conduction study (NCS) using Medelec Synergy machine, Oxford, UK for measurement of sensory and motor latency, amplitude, and velocity. Motor conduction studies consisted of stimulating the median nerve, ulnar nerve, common peroneal nerve and posterior tibial nerve while sensory conduction studies consisted of stimulating the median nerve, ulnar nerve and sural nerve.

In motor conductions, distal latency, conduction velocity, amplitude and F wave were evaluated. In sensory conduction, distal latency, conduction velocity and amplitude were evaluated. Patients with DPN and N-DPN in the first and second groups were divided into 3 groups according to the severity of their neuropathy. All NCS-positive patients were classified into following subgroups according to their outcomes: Mild-prolonged sensory latency and/or decreased velocity in sensory and motor nerves, with normal amplitude; moderate - above with mildly decrease sensory and/or motor amplitude; and severe - absent sensory potentials or severely reduced sensory and/or motor amplitudes.

### Statistical analysis

A power analysis was performed before the study was designed. Twenty patients were enrolled each in to three groups, with a total number of 60 participants. For statistical analysis, data were entered in Microsoft Excel format. Descriptive and analytic statistics were performed using the Statistical Package for the Social Sciences (SPSS) software for Windows, version 21.0 (SPSS Inc., Chicago, IL, USA). Independent t-test and One way-ANOVA test were applied to compare nominal data between the groups. The relationship between NMC duration and severity of polyneuropathy (both in DPN and N-DPN groups) was examined by Pearson's correlation analysis. A P-value < 0.05 was regarded as statistically significant.

## Results

Among the participants, the number of females and males was 12 (60%) and 8 (40%) in the first group, 11 (55%) and 9 (45%) in the second group, and 11 (55%) and 9 (45%) in the third group, respectively. The average age was  $56.15 \pm 8$ ,  $58.75 \pm 11.4$  and  $57 \pm 13.3$  years in the first, second and third groups, respectively. The groups were statistically homogeneous in terms of gender and age ( $p > 0.05$ )

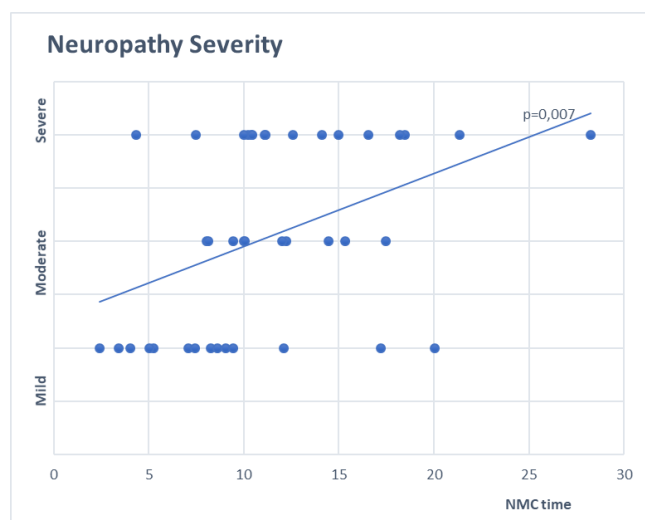
The average duration of NMC in the DPN, N-DPN and DM without polyneuropathy patient groups was  $12.69 \pm 6.3$ ,  $10.11 \pm 4$ , and  $6.76 \pm 1.4$  minutes, respectively. There was a statistically significant difference in the duration of NMC among the groups ( $p = 0.001$ ) (Table 1).

**Table1:** Duration of NMC in each group of patients

Patient groups	Duration of NMC (minutes)	p-value
Patients with diabetic polyneuropathy	$12.69 \pm 6.3$	0,001
Patients with non-diabetic polyneuropathy	$10.11 \pm 4$	
DM patients without polyneuropathy	$6.76 \pm 1.4$	



There was a positive, statistically significant ( $p = 0.007$ ,  $r = 0.42$ ) correlation between the NMC duration and the severity of polyneuropathy. The NMC duration increased with the severity of polyneuropathy (Figure 1)



**Figure 1:** The NMC duration increased with the severity of polyneuropathy.

## Discussion

Mucociliary clearance is one of the most important defense mechanisms of nasal respiratory epithelium. Harmful substances are retained in this mucous membrane and then removed from the nasal cavity by movements of cilia. Mucociliary interaction varies by environmental temperature, humidity, partial oxygen pressure, inhaled agents such as  $PO_2$ , pH, trauma and sulfur dioxide, formaldehyde, ozone, chlorine, and smoking. It has been also reported that MC differs by age, hypertension, viral infections, chronic sinusitis, chronic and allergic rhinitis, anatomical obstacles such as adenoid hypertrophy, cystic fibrosis, chronic bronchitis, septum deviation, secondary to surgeries, bronchial asthma, multiple sclerosis, and DM (10-12).

There are many studies and case reports on susceptibility to DM and ENT diseases in the literature. The most common presentations in the head and neck are otitis externa, hypoacusis, vertigo, dry mouth, dysphagia, fungal, and recurrent bacterial infections. However, changes in the nasal mucosa in DM are not well known. The main complaints of patients about the nose are xeromycteria, hyposmia, and reduced degrees of patency of the nose. Symptoms of chronic atrophic rhinitis, septal perforation, nasal mucosa ulcer, alar necrosis, staphylococcus or fungal infection may be seen in the otolaryngology examination (13-15). Yue compared mucociliary function in 50 insulin-dependent DM patients and 50 non-diabetic patients. He found that the mean value of nasal mucociliary clearance was significantly reduced in diabetic patients due to dry nose and increased pH values. He argued that the nasal mucociliary clearance is impaired as a result of increased viscosity due to electrolyte and water loss from the whole body and osmotic diuresis due to small vessel abnormalities (9).

There are consecutive studies in the literature confirming impaired nasal mucociliary clearance in diabetic patients (16-17).

In our study, the presence of polyneuropathy in DM patients was correlated with prolonged NMC duration. Polyneuropathy and autonomic nervous system involvement are one of the most common and most challenging complications of DM (18). DPN is the most common form of neuropathy worldwide and recent cross-sectional studies from the US and Europe have reported a prevalence of diabetic peripheral neuropathy ranging from 6% to 51% depending on the population studied (19,20). Although the presence of electrodiagnostic poly-neuropathy depends on multiple variables, chronic hyperglycemia significantly increases the risk and quantitatively reflect the severity of polyneuropathy in diabetic patients (21).

The most exciting finding of our study was that the severity of polyneuropathy and duration of NMC were significantly correlated. Dyck et al's classification of polyneuropathy is the most reliable classification used to measure the severity of DSPN in a given patient, to predict symptoms, symptoms, neurophysiological test results, and overall severity of DSPN, to conduct medical practice and epidemiological studies and randomized controlled clinical trials (21). Although it can be attributed to multiple factors, autonomic control of the nasal mucosa should also be kept in mind. The autonomic nervous system richly innervates the nasal cavity and paranasal sinuses. Nonetheless, its contribution to sinonasal symptoms is poorly re-searched and understood (8).

The biggest limitation of our study was that no autonomic nervous system test, including sudomotor axon reflex test was performed in the routine of our electromyography laboratory. Assessment of sudomotor dysfunction would contribute to detection of autonomic dysfunction in DPN. Although we could not specifically show autonomic dysfunction in our study, we speculate that it affects the nasal dryness that was emphasized in previous studies with DM patients. In our study, it was clearly seen that the severity of diabetic or non-diabetic polyneuropathy increased with the duration of NMC. In both of our neuropathy groups, there was a prolonged duration of NMC compared to the group of patients without neuropathy. In the DPN group, even longer duration of NMC was not statistically significant compared to the N-DPN group.

Our two most significant results were the presence of electrodiagnostic polyneuropathy, causing more pronounced NMC dysfunction compared to DM patients without polyneuropathy, and increase in the severity of diabetic or non-diabetic polyneuropathy with the NMC dysfunction.

The fact that nasal mucociliary activity is negatively affected in diabetes mellitus is not a new phenomenon. However, according to our knowledge; this is the first and only study to show that the presence and degree of polyneuropathy in DM, in other words, the presence of prolonged hyperglycemia, affects NMC more prominently.

## Conclusion

In conclusion, patients with diabetes mellitus are a special group, and preventable problems should be taken into consideration when examining nasal pathologies. It should be kept in mind that NMC dysfunction can be both a cause and a result in the management of DM patients, and thus it should be evaluated carefully.

**Author Contributions:** MAU: Project design, review of the literature, writing and revisions; SB: data collection and analyzes

**Ethical issues:** All authors declare originality and ethical approval of research. Responsibilities of research, responsibilities against local ethics commission are under the authors responsibilities. The study was conducted under defined rules by the local ethics commission guide-lines and audits.

**Conflict of Interest:** The authors declare that they have no competing interest.

## References

- Bustamante-Marin XM, Ostrowski LE. Cilia and Mucociliary Clearance. *Cold Spring Harb Perspect Biol.* 2017;9(4): a028241.
- Houtmeyers SE, Gosselink R, Gayan-Ramirez G, Decramer M. Regulation of mucociliary clearance in health and disease. *Eur respir J* 1999; 13:1177-1188.
- Xavier RF, Ramos D, Ito JT, Rodrigues FMM, Bertolini GN, Macchione M, et al. Effects of Cigarette Smoking Intensity on the Mucociliary Clearance of Active Smokers. *Respiration* 2013; 86:479-485.
- Jiao J, Zhang L. Influence of Intranasal Drugs on Human Nasal Mucociliary Clearance and Ciliary Beat Frequency. *Allergy Asthma Immunol Res.* 2019;11(3):306-319.
- Corbo GM, Foresi A, Bonfitto P, Mugnano A, Agabiti N, Cole PJ. Measurement of nasal mucociliary clearance. *Arch Dis Child.* 1989;64(4):546-550.
- Grossan M. Mucociliary Clearance: Measures and Therapies. *Otolaryngol (Sunnyvale)* 2017;7: 336.
- Rutland J, Cole PJ. Nasal mucociliary clearance and ciliary beat frequency in cystic fibrosis compared with sinusitis and bronchiectasis. *Thorax.* 1981;36(9):654-658.
- Yao A, Wilson JA, Ball SL. Autonomic nervous system dysfunction and sinonasal symptoms. *Allergy Rhinol (Providence).* 2018; 9:2152656718764233.
- Yue WL. Nasal mucociliary clearance in patients with diabetes mellitus. *The Journal of Laryngology and Otology* 1989; 103: 853-55.
- Proença de Oliveira-Maul J, Barbosa de Carvalho H, Goto DM, Maia RM, Fló C, Barnabé V, et al. Aging, diabetes, and hypertension are associated with decreased nasal mucociliary clearance. *Chest.* 2013 Apr;143(4) 1091-1097.
- Deniz M, Uslu C, Ogredik EA, Akduman D, Gursan SO. Nasal mucociliary clearance in total laryngectomized patients. *Eur Arch Otorhinolaryngol* 2006; 263: 1099-1104.
- Sahin E, Hamamcı M, Kantekin Y. Measurement of mucociliary clearance in the patients with multiple sclerosis. *Eur Arch Otorhinolaryngol.* 2020; 277(2) 469-473
- Müller M, Betlejewski S. Nasal mucosa in patients with diabetes mellitus. *Otolaryngol Pol.* 2003; 57:361-364.
- Zhang Z, Adappa ND, Lautenbach E, Chiu AG, Doghramji L, Howland TJ, et al. The effect of diabetes mellitus on chronic rhinosinusitis and sinus surgery outcome. *Int Forum Allergy Rhinol.* 2014 Apr;4(4):315-20.
- Dinesh R, Avatar S, Haron A, Suhana MD, Azwarizan MD. Nasal septal abscess with uncontrolled diabetes mellitus: case reports. *Med J Malaysia.* 2011 ;66(3):253-4.
- Sachdeva A, Sachdeva OP, Gulati SP, Kakkar V. Nasal mucociliary clearance & mucus pH in patients with diabetes mellitus. *The Indian Journal of Medical Research.* 1993; 98:265-268.
- Selimoglu MA, Selimoglu E, Kurt A. Nasal mucociliary clearance and nasal and oral pH in patients with insulin-dependent diabetes. *Ear, Nose, & Throat Journal.* 1999; 78:585-588.
- Dyck PJ, Albers JW, Andersen H, Arezzo JC, Biessels GJ, Bril V, et al; Toronto Expert Panel on Diabetic Neuropathy. Toronto Expert Panel on Diabetic Neuropathy. Diabetic polyneuropathies: update on research definition, diagnostic criteria and estimation of severity. *Diabetes Metab Res Rev.* 2011; 27:620-628.
- Iqbal Z, Azmi S, Yadav R, Ferdousi M, Kumar M, Cuthbertson DJ, et al. Diabetic Peripheral Neuropathy: Epidemiology, Diagnosis, and Pharmacotherapy. *Clin Ther.* 2018; 40:828-849.
- Hicks CW, Selvin E. Epidemiology of Peripheral Neuropathy and Lower Extremity Disease in Diabetes. *Curr Diab Rep.* 2019; 19: 86.
- Lee WJ, Jang S, Lee SH, Lee HS. Correlation Between the Severity of Diabetic Peripheral Polyneuropathy and Glycosylated Hemoglobin Levels: A Quantitative Study. *Ann Rehabil Med.* 2016;40(2):263-270.

## The treatment of chronic wounds with boric acid polyurethane sponges combined with negative pressure wound treatment: a multi-center, prospective, randomized study

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### Abstract

**Objective:** This study aimed to compare the outcomes of the use of polyurethane boric acid sponges and sponges containing silver nitrate over the negative pressure wound treatment.

**Materials and methods:** The patients were separated as Group 1 (n:30) treated with boric acid group, and Group 2 (n:34) treated with silver nitrate group (Group 2). The wound healing of each patient was evaluated quantitatively on a cellular basis from a table of macroscopic and histopathological scoring.

**Results:** No statistically significant difference was determined between the groups applied with boric acid and silver nitrate in respect of the culture results from the first visit (p:0.705). In the final evaluation of wound dimensions on day 21, a significant reduction was seen in wound width (p=0.001), wound length (p=0.003), and wound depth (p<0.001) in the boric acid group, and no significant results were obtained in the silver nitrate group. In the quantitative cellular evaluations, a statistically significant difference was determined in favor of the boric acid group in respect of inflammatory cell count, angiogenesis, granulation, and re-epithelialization (p<0.001 for all).

**Conclusion:** Sponges with boric acid can have a positive effect on chronic wound recovery by improving cellular proliferation, cellular differentiation and cellular migration in addition to antimicrobial properties when used in combination with the negative pressure wound treatment system. Therefore, boric acid sponges seem to be a good alternative to silver nitrate sponges.

**Keywords:** chronic wounds, boric acid, negative pressure treatment

### Introduction

The healing of chronic wounds tends to not advance beyond the inflammatory stage (1). For the wound to emerge from that stage and enter the healing period, the infection must be brought under control, angiogenesis must be provided and the epithelialization phase should be accelerated. Many methods and technological products are used to reach these goals in the treatment of chronic wounds. One of the most important of these is the vacuum-assisted wound closure (VAC) system, also known as negative pressure wound treatment (NPWT). However, the debate continues about the optimum physicochemical structure of the interface contact material in this system.

Silver nitrate polyurethane sponges are currently used as wound interface material. The most important property of these sponges is the broad spectrum anti-bacterial effect. However, local and systemic complications of this substance, which are not dose-dependent, have raised concerns in clinicians (2-4). Boric acid has long been used in topical wound treatments, such as eye and ear drops, and in recent years, promising results have been obtained from in vitro and in vivo studies of infected wounds (5-7). In addition to the valuable anti-microbial effect of boric acid against yeast and fungi, it also has the effect of increasing cellular proliferation, macrophage migration, and levels of growth factor and dermal cell gene expression.

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It also plays a significant role in the wound healing process by increasing the extra-cellular matrix synthesis through proteoglycan, collagen and protein production (6, 8, 9).

The properties demanded of wound interface material are both a broad-spectrum anti-bacterial effect and a cellular proliferative effect. Previous studies have reported that boric acid has both these properties (5, 7). The aim of this prospective, randomized, multi-center, clinical study was to macroscopically and histopathological compare the anti-microbial, angiogenetic, macrophage stimulating and epithelialization effects of boric acid sponges with silver nitrate sponges.

## Material and Methods

The study protocol was approved by the Clinical Research Ethics Committee of Health Sciences University Medical Faculty Adana Training and Research Hospital (decision no:660- MON 665.144.1, dated:08.01.2020) and by the Turkish Ministry of Health Drugs and Medical Supplies Institute (decision no: 68869993-511.06-E.27180, dated: 03.12.2019). The study was conducted in the Orthopedics and Traumatology Departments and the Plastic Reconstructive and Aesthetic Surgery Departments in 4 different university hospitals, determined by the Ministry of Health.

The study included 64 patients with chronic wounds of different etiological causes (diabetic, secondary to trauma, decubitus ulcer etc). The patients were separated as Group 1 (n:30) treated with boric acid and Group 2 (n:34) treated with silver nitrate. The wounds were evaluated macroscopically and microscopically on first presentation, then on days 7, 14, and 21, by a researcher blinded to the treatment groups. Patients were excluded from the study if they were aged <18 years or >90 years, were pregnant, had venous ulcers, septicemia/septic shock, or were receiving immunosuppressive therapy or chemotherapy.

Following patient evaluation and physical examination, randomization to the treatment groups was applied using software that included the electronic Case Report Form. In the group applied with boric acid sponge treatment, the boric acid levels were measured in blood and urine samples on days 7, 14, and 21. The general healing of the wound was evaluated with a Visual Analog Scale (VAS), also on days 7, 14, and 21.

## Technique

Boric acid sponges are produced from polyurethane, and have a broad absorption surface with pores of 400-600µm. On these sponges, boric acid is distributed evenly on all the surfaces at the rate of 3-5% at dimensions of 300 x 70 µm. Silver nitrate sponges are polyurethane structures with hydrophobic properties containing silver nitrate conforming to ISO-10993-10 and ISO10993-11.

After macroscopic examination of the chronic wounds, radical debridement was performed. Deep wound cultures and biopsies were taken.

Following the application of the sponges appropriate to the postoperative wound, the same make of VAC device was applied to all the patients in both groups, using the same pressure and the same duration. The interface materials, i.e. the sponges, were changed every 7 days in all patients. No antibiotics were administered to any patient. The histopathological evaluations applied to the patients are shown in Table 1 (9).

## Statistical Analysis

Data included in the study were analyzed statistically using SPSS 21 software. Conformity of continuous variables to normal distribution was assessed with the Shapiro Wilk test. As the variables did not show normal distribution, logarithmic transformation was applied to provide normal distribution, and analysis was made with parametric methods.

The Student's t-test was applied in the comparisons of the mean values of the boric acid and silver nitrate groups in respect of the measurements taken by the clinician, the VAS scores reported by the patient, and the width, depth, and length measurements of the wound.

As the VAS scores, and the width, depth, and length measurements of the wound were taken at 4 different times, the change over time was determined using Repeated Measurements Variance Analysis, which was also used to observe time x group interaction. The re-epithelialization, granulation, inflammatory and angiogenesis measurements were evaluated categorically.

In the comparison of these measurements between the groups, the Chi-square and Fisher's Exact tests were used. In the separate comparisons of the groups of the measurements taken at 4 different times, the Marginal Homogeneity test was used.

Descriptive statistics were stated as mean  $\pm$  standard deviation, minimum and maximum values for continuous variables, and as number (n) and percentage (%) for categorical variables. A value of  $p < 0.05$  was accepted as statistically significant.

## Adverse Events:

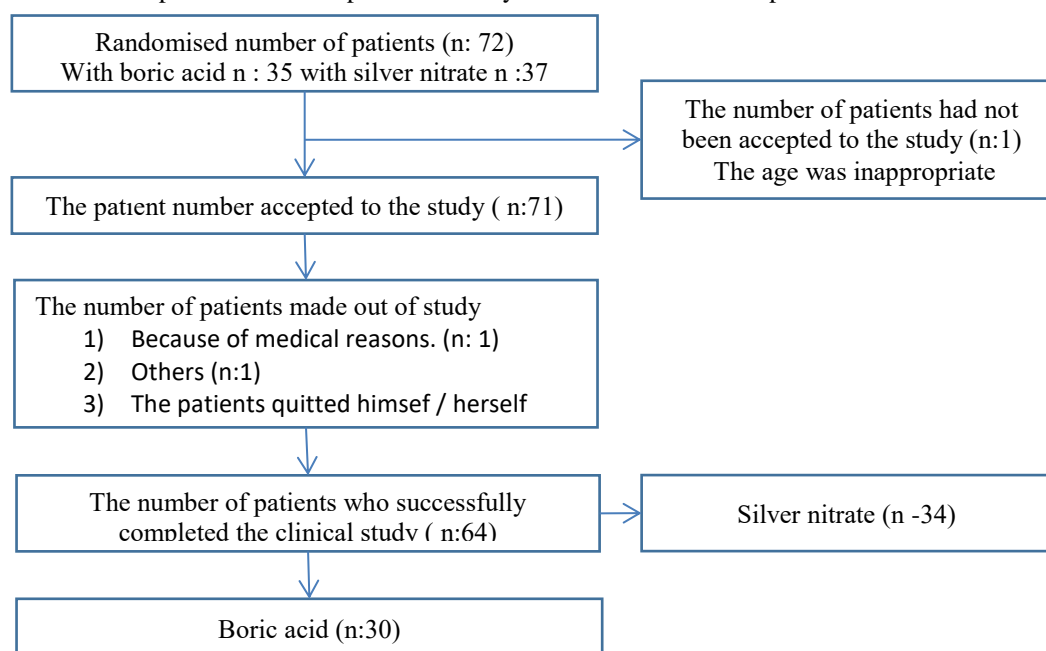
All the adverse events were recorded on the electronic case report form, were coded according to the MedDRA coding system, and presented in detailed explanatory tables.

## Results

Statistical analysis of the demographic data of the patients: No statistically significant difference was determined between the groups in respect of age and height ( $p > 0.05$ ). A statistically significant difference was determined between the groups in respect of mean weight ( $p = 0.037$ ). The general demographic characteristics of the patients, such as gender, general habits, comorbidities and wound localization were seen to be homogenous in the two groups ( $p = 0.092$ ) (Table 2 and 3).

**Table 1:** The histopathological scroing of the wounds

Score	Reepitelization	Granulation	Inflammatory Cells	Angiogenesis
0	No epithelial proliferation in more than 70% of tissue	Immature and inflamed tissue in more than 70% of tissue	13-15 inflammatory cells per histopathological site	Angiogenesis deficiency, presence of bleeding, edema and congestion
1	Poor epithelization in more than 60% of tissue	Fine immature and inflamed tissue in more than 60% of tissue	10-13 inflammatory cells per histopathological area	Presence of 1-2 vessels per area, presence of bleeding, edema and congestion
2	Incomplete epithelization of more than 40% of tissues	Medium regeneration on more than 50% of texture	7-10 inflammatory cells per histopathological site	There are 3-4 vessels per area. Moderate edema congestion
3	Moderate epithelialisation in 60% of tissues.	Thick granulation tissue and well-formed collagen tissue in more than 60% of tissue	4-7 inflammatory cells per histopatological site	Presence of 5-6 vessels per area, small amount of congestion and edema
4	Completed epithelization	Finished granulation tissue	1-4 inflammatory cells per histopathological site	7 veins per field

**Table 2:** Number of patients who completed the study in accordance with the protocol**Table 3:** Comparison of the demographic findings of both groups in terms of age, height and weight

	Boric acid		Silver Nitrate		p1
	Mean + SD	Min-Max	Mean + SD	Min-Max	
Age	54,43±13,2	21,00-77,0	59,68±12,4	25,00-83,00	0,108
Height	175,37±9,1	159,00-194,00	172,09±8,4	161,00-187,00	0,139
Length	80,10±15,0	55,00-110,00	87,97±14,5	62,00-113,00	<b>0,037</b>
	n	%	n	%	p2
Gender					
Male	25	83,3	22	64,7	0,092
Female	5	16,7	12	35,3	

**Group 1** included 30 patients, comprising 25 males and 5 females with a mean age of  $53.43 \pm 13.28$  years, mean height of  $175.37 \pm 9.10$  cm, and mean weight of  $80.10 \pm 15.03$  kg. The blood and urine values of boric acid of all the patients were at a level that could not be measured. Chronic disease in at least two systems was present in 24 patients. The wounds were localized in the foot in 18 (60%) patients, in the sacrum in 6 (20%), on the thigh in 2 (6.6%), in the gluteal region in 2 (6.6%), on the anterior wall of the chest in 1 (3.3%), and in the perineal region in 1 (3.3%). On first presentation, the wound width was mean  $57.43 \pm 62.85$  mm, length was mean  $32.73 \pm 20.66$  mm, and mean depth was  $39.37 \pm 45.95$  mm.

**Group 2** included 34 patients, comprising 22 males and 12 females with a mean age of  $59.68 \pm 12.42$  years, mean height of  $172.09 \pm 8.40$  cm, and mean weight of  $87.97 \pm 14.52$  kg. Chronic disease in at least two systems was present in 31 patients. The wounds were localized in the foot in 24 (70.5%) patients, in the sacrum in 6 (17.64%), on the thigh in 2 (5.88%), on the forearm in 1 (2.94%), and in the hand region in 1 (2.94%). On first presentation, the wound width was mean  $44.41 \pm 25.50$  mm, length was mean  $32.88 \pm 21.44$  mm, and mean depth was  $35.38 \pm 40.21$  mm.

## 2. Distribution of VAS scores at different measurement times

**2.1. Clinician evaluations:** No statistically significant difference was determined between the groups in the scoring made by the clinician at the first visit. In the boric acid group, the mean clinician score was 4.66 at baseline and decreased at subsequent measurement times to 1.50 at the final visit. The difference between the scores at the 4 measurement times was determined to be statistically significant ( $p < 0.001$ ). In the silver nitrate group, the initial clinician score was mean 4.86, and this decreased to 3.65 at the final measurement, with no statistically significant difference determined in the change over time ( $p = 0.087$ ).

**2.2. Patient Evaluation:** No statistically significant difference was determined between the groups in the scoring made by the patient at the first visit ( $p = 0.088$ ). No significant difference was determined between the groups in respect of the scoring at subsequent measurement times. In the boric acid group, the baseline mean score of 3.66 decreased statistically significantly to 1.13 at the final measurement ( $p < 0.001$ ). In the silver nitrate group, no statistically significant difference was determined between the baseline score of mean 2.89 and the final mean score of 2.21 ( $p = 0.201$ ).

**3. Blood and urine levels of boric acid:** In the blood and urine samples of the patients applied with sponges containing boric acid in the negative pressure system, boric acid was reported to be at levels that could not be measured in both analyses.

**4. Changes in culture results over time:** In Group 1, culture positivity was determined in 22/30 patients (73.3%), and of these, mixed infection was present in 12 (54.54%). The most common pathogen micro-organisms were *Staphylococcus Aureus* and *Pseudomonas Aeruginosa*. In

Group 2, culture positivity was determined in 16/34 patients, and of these, mixed infection was present in 8. The most common pathogen micro-organisms were *Staphylococcus Aureus* and *Pseudomonas Aeruginosa*. No statistically significant difference was determined between the groups in respect of the culture results ( $p = 0.705$ ). At the second visit, the number of patients with positive culture results had decreased in the boric acid group. In the comparisons made at the second, third and fourth visits, a statistically significant difference was determined between the groups in respect of culture positivity ( $p = 0.021$ ,  $p = 0.008$ ,  $p < 0.001$ , respectively).

## 5. Statistical analysis of wound dimensions:

In the boric acid group, a decrease was observed in wound width ( $p = 0.001$ ), and all the changes were found to be statistically significant compared to the previous measurements ( $p < 0.05$ ). In the silver nitrate group, no statistically significant difference was determined in the wound width over time ( $p = 0.200$ ). The difference between the groups in respect of the change in wound width over time was observed to be statistically significant ( $p = 0.010$ ).

In the boric acid group, a significant decrease in wound length was determined ( $p = 0.003$ ). The third measurement was statistically significantly different to the second measurement, and the fourth measurement to the first measurement ( $p < 0.05$ ). In the silver nitrate group, no statistically significant difference was observed in wound length over time ( $p = 0.276$ ). The difference between the groups in respect of the change in wound length over time was observed to be statistically significant ( $p = 0.019$ ).

In the boric acid group, a decrease was observed in wound depth ( $p < 0.001$ ), and all the changes were found to be statistically significant compared to the previous measurements ( $p < 0.05$ ). In the silver nitrate group, a statistically significant difference was determined in the wound depth over time ( $p = 0.002$ ) and all the changes were found to be statistically significant compared to the previous measurements ( $p < 0.05$ ). The difference between the groups in respect of the change in wound depth over time was observed to be statistically significant ( $p = 0.002$ ) (Table 4).

**6. Analysis of adverse events:** In 3 (4.22%) patients, an adverse event was reported, one of which was recorded as "possibly" related to the drug used, and 2 as "probably". All the adverse events that developed were identified as silver allergy.

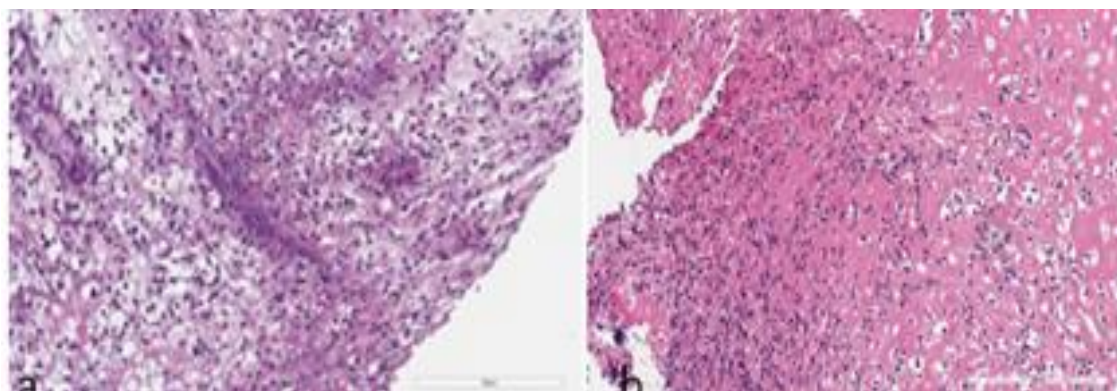
## 7. Macroscopic and microscopic features of the wounds and the statistical analysis of the changes over time

On day 7, there were small foci of necrosis and the wound surfaces were inflamed in both groups. A shrinking of the wound size was not determined at this stage, and similar microscopic features were present in both groups. Generally, in addition to lymphocyte and macrophage predominance, there was observed to be endothelial damage and capillary vasoconstriction. On day 14, there was a noticeable vascular proliferation in both groups, and this was more evident in Group 1 (Figure 1 and 2).

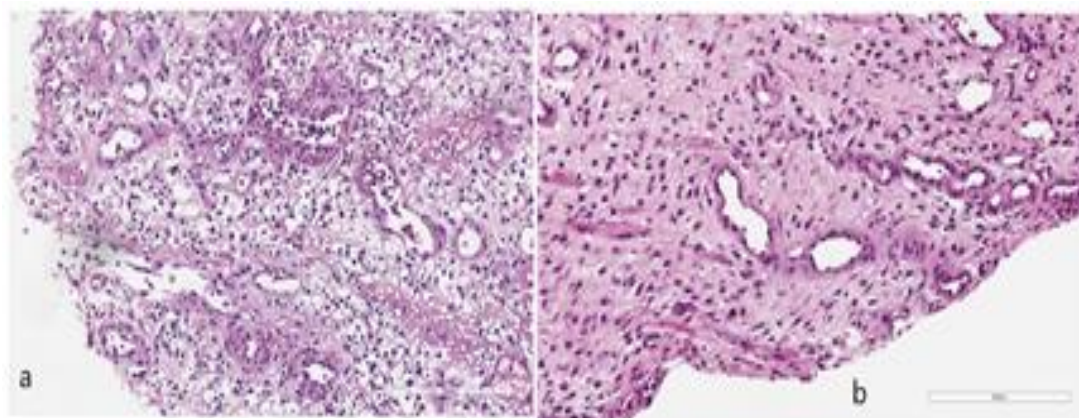


**Table 4:** Comparison of the wounds before and after treatment in 3D

		Boric acid		Silver Nitrate		p <sub>group</sub>	p <sub>general</sub>
Wound size		Mean+SD	Min-Max	Mean +SD	Min-MaX		
Width	Pre-treatment	57,43±62,8	10-350	44,41±25,5	15-110	0,570	0,006
	Post-treatment	38,83±43,32	0-230	43,18±26,1	0-110	0,132	
	Ptime	<0,001		0,276			
Length	Pre-treatment	32,73±20,6	8-75	32,88±21,4	5-75	0,885	0,021
	Post-treatment	26,00±23,2	0-100	40,68±33,7	0-170	0,068	
	Ptime	0,008		0,756			
Depth	Pre-treatment	39,37±45,9	5-200	35,38±40,2	6-170	0,862	0,006
	Post-treatment	18,33±24,1	0-90	25,44±31,4	0-115	0,096	
	P time	<0,001		0,008			



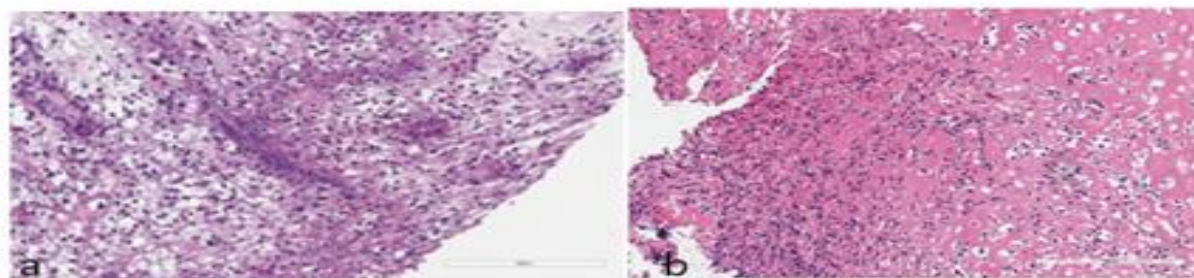
**Figure 1:** Histopathological images of first application biopsies. a: The patient who is applied a silver sponge. Ulceration of the squamous epithelium and fibrinoleukocytic exudate. b: The patient was treated with Acid Boric. Biopsy: Surface ulceration of squamous epithelium, fibrinoleukocytic exudate, Hematoxylin Eosin staining x200



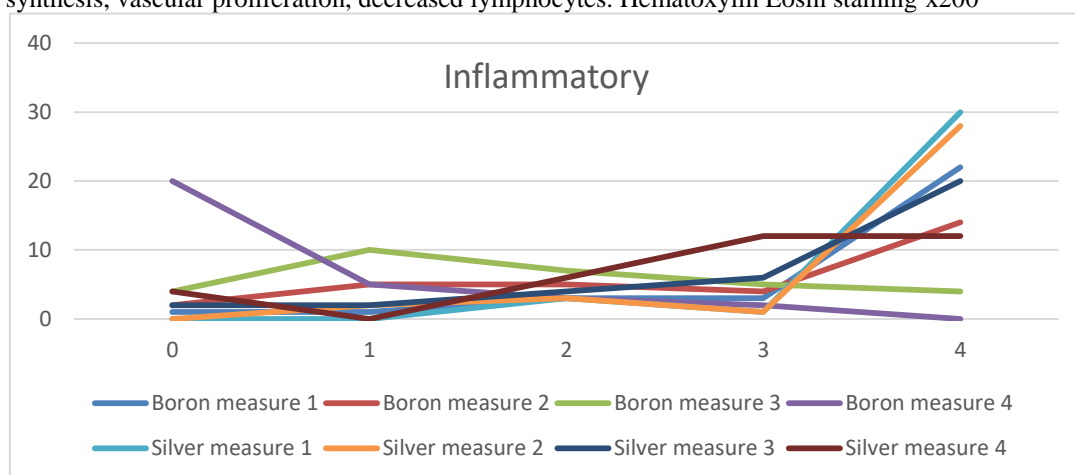
**Figure 2:** Histopathological images of specimens taken on the 7th day. (a): The patient who was treated with a silver sponge. Vascular proliferation by dense neutrophilic polymorphs and lymphocytes. (b) The patient to whom a boron sponge is applied. Vascular proliferation, collagen synthesis, lymphoplasmacytic cell infiltration. Hematoxylin Eosin staining x200

On day 21, macroscopically vascularization of the wound bed was seen to be better in Group 1, and the wound diameter had shrunk noticeably from the periphery. These features were less evident in Group 2. Microscopically, vascular proliferation, fibroblastic activation, and epithelialization were more intense in Group 1. These features were present in Group 2, but not so intensely (Figure 3).

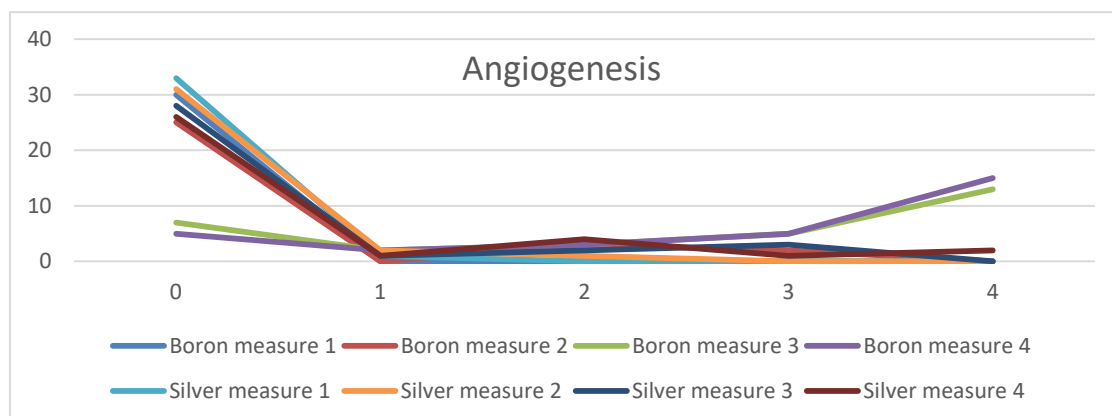
**7.1a. Inflammation phase and cell characterization:** In the boric acid group and the silver nitrate group, the ratio of inflammatory cells showed a statistically significant change at all measurement points compared to the previous measurement ( $p < 0.05$ ,  $p < 0.05$ ). A statistically significant difference was determined between the boric acid group and the silver nitrate group in respect of the ratio of inflammatory cells at the final measurement ( $p < 0.001$ ) (Graphic 1).



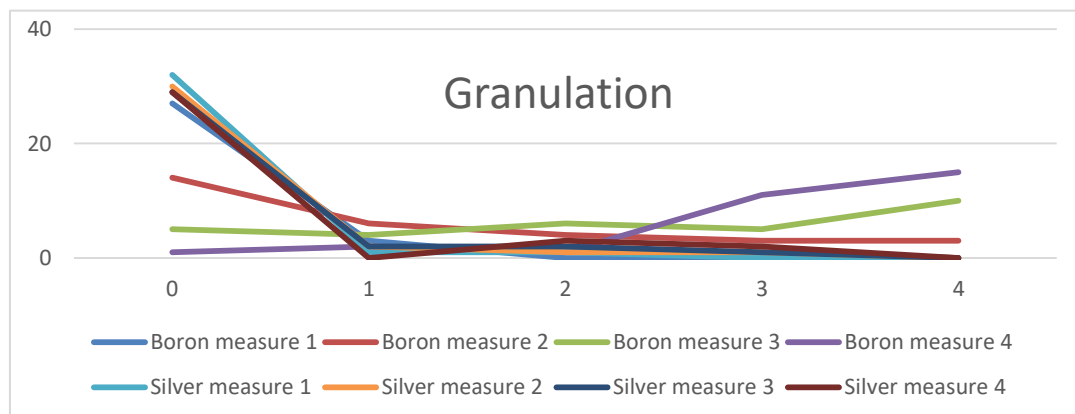
**Figure 3:** Histopathological images of specimens taken on the 14th day. (a) Patient with silver sponge applied Vascular proliferation and lymphoplasmacytic cell infiltration. (b) The patient to whom a boron sponge is applied. Pronounced collagen synthesis, vascular proliferation, decreased lymphocytes. Hematoxylin Eosin staining x200



**Graphic 1:** Comparison of inflammation level among two different groups at different visits



**Graphic 2:** Comparison of angiogenesis between two different groups at different visits



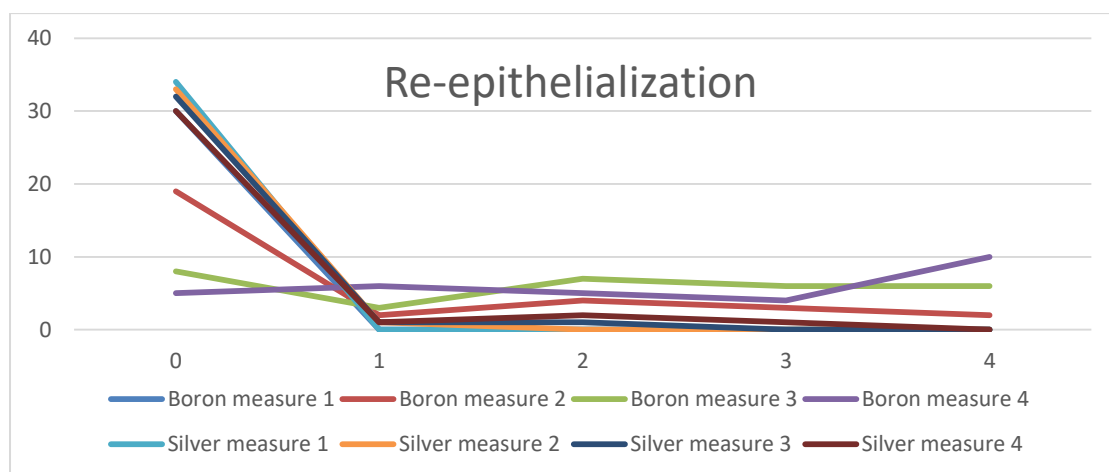
**Graphic 3:** Comparison of granulation between two different groups at different visits

**7.1b. Angiogenesis:** In the boric acid group the rate of angiogenesis showed a statistically significant change at all measurement points compared to the previous measurement ( $p < 0.05$ ). In the silver nitrate group, no statistically significant change was seen at all measurement points compared to the previous measurement ( $p > 0.05$ ). A statistically significant difference was determined between the boric acid group and the silver nitrate group in respect of the angiogenesis rate at the final measurement ( $p < 0.001$ ) (Graphic 2).

**7.1c. Granulation:** In the boric acid group the rate of granulation showed a statistically significant change at all measurement points compared to the previous measurement ( $p < 0.05$ ). In the silver nitrate group, a statistically significant change was seen in the granulation rate of the second measurement compared to the first measurement ( $p < 0.05$ ). No significant change was seen between the third and second measurements or between the fourth and third measurements in the silver nitrate group ( $p > 0.05$ ).

No statistically significant difference was determined between the boric acid group and the silver nitrate group in respect of the first and second measurements of the granulation rate ( $p < 0.05$ ), and the difference between the groups at the final measurement was statistically significant ( $p < 0.001$ ) (Graphic 3).

**7.1d. Re-epithelialization:** In the boric acid group the rate of re-epithelialization showed a statistically significant change at all measurement points compared to the previous measurement ( $p < 0.05$ ). In the silver nitrate group, no statistically significant change was seen at all measurement points compared to the previous measurement ( $p > 0.05$ ). A statistically significant difference was determined between the boric acid group and the silver nitrate group in respect of the re-epithelialization rate at the final measurement ( $p < 0.001$ ) (Graphic 4).



**Graphic 4:** Comparison of re-epithelialization between two different groups at different visits

## Discussion

It is estimated that by 2040 there will be 640 million patients diagnosed with diabetes, and it is thought that 10% of these will have diabetic ulcers (10). Despite these data, chronic wounds continue to be one of the largest and growing problems around the world. In particular, the rapid increase in the geriatric population has caused an increase in the prevalence of diabetes mellitus and other chronic diseases, and an increase in the incidence of chronic wounds. In addition to having a negative effect on quality of life, chronic wounds are comparable to cancer in respect of mortality rates (11-13).

The optimal physiochemical properties of sponges applied as the intermediate unit in NPWT systems, are still not fully known (14, 15). Silver nitrate is currently used as the filling material in sponges. Silver is an effective bactericide against both gram-positive and gram-negative bacteria. However, at high concentrations, it is toxic to mammalian cells. Complications of silver such as leukopenia, kidney and liver dysfunction, and neuropathy are still unresolved severe problems (16-20).

Another agent which is accepted as broad-spectrum is boric acid, which has been shown in several studies to be effective against bacteria such as *S. aureus*, *K. pneumoniae*, *C. Albicans*, *A. Niger*, *E. coli*, and *P. Aeruginosa* (21, 22). In a recent study was compared boric acid sponges and silver nitrate sponges, and they reported that anti-infective properties were much more evident in boric acid sponges compared to silver nitrate sponges (23). In addition to the anti-infective property of boric acid, the authors claimed that this effect was formed by the proliferation of local and systemic defense cells. In the current study, culture positivity was determined in 73.3% of the patients in the boric acid group, and 54.54% of those patients had mixed-type infection. No statistically significant difference was determined between the two groups in respect of culture positivity on first presentation, whereas a significant difference was observed in the final culture results ( $p < 0.001$ ).



Various studies have shown that boric acid shows notable antimicrobial effects against yeast and fungi and significantly increases cellular proliferation, macrophage migration, growth factor and dermal cell gene expression (24-26). In another study, it was reported that boric acid played an important role in the wound healing process by increasing extra-cellular matrix synthesis through proteoglycan, collagen and protein production (6). In the current study, the wound healing process was evaluated microscopically and macroscopically. Macroscopic evaluation of the wound was made according to metric measurements of the wound dimensions and VAS scores. In the metric evaluation of the wound dimensions in particular, there was a significant decrease in wound width in the boric acid group ( $p=0.001$ ), and there was no statistically significant difference in the wound width compared to the baseline measurement or over time in the silver nitrate group ( $p=0.200$ ). While a significant decrease was seen in wound length in the boric acid group ( $p=0.003$ ), no significant difference was determined over time in the silver nitrate group ( $p=0.276$ ). The wound depth showed a significant difference over time in the boric acid group ( $p<0.001$ ) and no difference was determined in the silver nitrate group ( $p>0.05$ ).

In the scoring applied by the clinicians, no significant difference was determined between the groups at the first visit, and in the subsequent visits, significant differences were determined between the groups at each measurement time ( $p<0.001$ ). In the silver nitrate group, no statistically significant difference was found in the subsequent clinician evaluations compared to the baseline measurement ( $p=0.087$ ). In the subjective patient evaluation (VAS), no significant difference was determined between the groups in the initial evaluation ( $p=0.088$ ). In the comparisons of the groups at subsequent visits, significant differences were observed in favor of the boric acid group ( $p<0.001$ ).

When the cases were examined microscopically, although there was no significant difference in the histopathological features of the biopsies taken at 7 days compared to the first presentation, there was observed to be a decrease in chronic inflammatory cells such as lymphocytes and macrophages in both groups. However, there was no statistically significant difference between the two groups in respect of the number of inflammatory cells. In the boric acid group, chronic inflammatory cells were seen to be extremely sparse at this stage and the most important formation was a noticeable vascular germination.

At this stage, there was no significant difference between the groups in respect of inflammatory cells, angiogenesis, granulation, or epithelialization. As a result of the histopathological examination of the final biopsies taken on day 21, significant differences were determined at every stage in the boric acid group. Although vascular proliferation, fibroblastic activation, and granulation tissue were clearly noticeable at this stage, they were less evident in the silver nitrate group.

As a result of the macroscopic evaluation, the boric acid sponges were determined to have shrunk the wounds in all three dimensions. Furthermore, in the microscopic

analyses, cell proliferation was determined to be responsible for wound healing. The determination of statistically significant improvements, especially in vascular proliferation, fibroblastic activation and granulation tissue, seemed to confirm the theory that boric acid had proliferative effects on a cellular basis.

Although this was a multi-center, prospective, randomized study, there were also limitations of the low number of cases included and that the results were not examined at the molecular level.

## Conclusion

The results of this study showed that while the inflammatory cell count and ratios were similar in both groups on first presentation, there was a significant decrease in inflammatory cells in the boric acid group as the treatment progressed over time. In the silver nitrate group, there was no noticeable decrease in inflammatory cells even at the late stages. In the samples taken at the second and third measurement points, the formation of collagen was seen to have started earlier and in broader areas in the boric acid group. Collagen formation started later in the silver nitrate group and was observed in a few focal areas. Vascular proliferation was seen at an early stage in the boric acid group and angiogenesis increased more as the treatment progressed. However, in the silver nitrate group, vascular proliferation was less evident and was observed to be focal. In the samples taken at the final examination, epithelialization had started in both groups, and was more evident in the boric acid group.

In addition to anti-microbial effects, boric acid sponges can be considered to form a synergic effect with the NWPT system in cell proliferation, differentiation and migration, which are necessary for wound healing. The use of these sponges can be considered a good alternative to silver nitrate sponges, which also have a broad-spectrum anti-microbial effect, but for which the same effect could not be determined in cell proliferation.

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## References

1. Falanga V. Classifications for wound bed preparation and stimulation of chronic wounds. Wound repair and regeneration: official publication of the Wound Healing Society [and] the European Tissue Repair Society. 2000;8(5):347.
2. Gaisford S, Beezer A, Bishop A, Walker M, Parsons D. An in vitro method for the quantitative determination of the antimicrobial efficacy of silver-containing wound dressings. International journal of pharmaceutics. 2009;366(1-2):111.
3. McDonnell G, Russell AD. Antiseptics and Disinfectants: Activity, Action, and Resistance. Clinical Microbiology Reviews. 1999;12(1):147.



4. Siegel HJ, Herrera DF, Gay J. Silver Negative Pressure Dressing With Vacuum-assisted Closure of Massive Pelvic and Extremity Wounds. *Clinical Orthopaedics and Related Research*. 2014;472(3):830.
5. Borrelly J, Blech M, Grosdidier G, Martin-Thomas C, Hartemann P, editors. Contribution of a 3% solution of boric acid in the treatment of deep wounds with loss of substance. *Annales de chirurgie plastique et esthetique*; 1991.
6. Nzietchueng R, Dousset B, Franck P, Benderdour M, Nabet P, Hess K. Mechanisms implicated in the effects of boron on wound healing. *Journal of trace elements in medicine and biology: organ of the Society for Minerals and Trace Elements (GMS)*. 2002;16(4):239.
7. Tepedelen B, Soya E, Korkmaz M. Boric Acid Reduces the Formation of DNA Double Strand Breaks and Accelerates Wound Healing Process. *Biological trace element research*. 2016;174(2):309.
8. Samman S, Foster M, Hunter D. The Role of Boron in Human Nutrition and Metabolism. *Boron Science: New Technologies and Applications*. 2016:73.
9. Gupta A, Kumar P. Assessment of the histological state of the healing wound. 2015.
10. Armstrong D, Wrobel J, Robbins J. Guest Editorial: are diabetes-related wounds and amputations worse than cancer? *International wound journal*. 2007;4(4):286.
11. Hopman W, Harrison M, Coe H, Friedberg E, Buchanan M, VanDenKerkhof E. Associations between chronic disease, age and physical and mental health status. *Chronic diseases in Canada*. 2009;29(3):108.
12. Bickers DR, Lim HW, Margolis D, Weinstock MA, Goodman C, Faulkner E, et al. The burden of skin diseases: 2004: A joint project of the American Academy of Dermatology Association and the Society for Investigative Dermatology. *Journal of the American Academy of Dermatology*. 2006;55(3):490-500.
13. Kapukaya R, Ciloglu O. Treatment of tibial intercalary defects secondary to tumour with modified fibular flap technique. *European Journal of Plastic Surgery*. 1-8.
14. Orgill D, Manders E, Sumpio B, Lee R, Attinger C, Gurtner G, et al. The mechanisms of action of vacuum assisted closure: more to learn. *Surgery*. 2009;146(1):40.
15. Wu M, Sun M, Dai H, Xu J, Guo R, Wang Y, et al. Negative-pressure wound therapy: An effective adjunctive treatment to assist flap survival and wound closure. *Journal of plastic, reconstructive & aesthetic surgery: JPRAS*. 2018;71(11):1664.
16. Sudmann E, Vik H, Rait M, Todnem K, Andersen K, Julsham K, et al. Systemic and local silver accumulation after total hip replacement using silver-impregnated bone cement. *Medical progress through technology*. 1994;20(3-4):179.
17. Trop M, Novak M, Rodl S, Hellbom B, Kroell W, Goessler W. Silver-coated dressing acticoat caused raised liver enzymes and argyria-like symptoms in burn patient. *The Journal of trauma*. 2006;60(3):648.
18. Chung I, Lee M, Shin D, Jung H. Three systemic argyria cases after ingestion of colloidal silver solution. *International journal of dermatology*. 2010;49(10):1175-7.
19. Wan A, Conyers R, Coombs C, Masterton J. Determination of silver in blood, urine, and tissues of volunteers and burn patients. *Clinical chemistry*. 1991;37(10 Pt 1):1683.
20. Drake P, Hazelwood K. Exposure-related health effects of silver and silver compounds: a review. *The Annals of occupational hygiene*. 2005;49(7):575.
21. Özcan KM, Sitesi Y, Blok A. The efficacy of boric acid in otomycosis: an in vitro study. *Mediterr J Otol*. 2005;2:00-.
22. Yilmaz MT. Minimum inhibitory and minimum bactericidal concentrations of boron compounds against several bacterial strains. *Turkish Journal of Medical Sciences*. 2012;42(Sup. 2):1423-9.
23. Kapukaya R, Ciloglu O. Treatment of chronic wounds with polyurethane sponges impregnated with boric acid particles: A randomised controlled trial. *International Wound Journal*. 2020;17(5):1159-65.
24. Doğan A, Demirci S, Çağlayan A, Kılıç E, Günel M, Uslu U, et al. Sodium pentaborate pentahydrate and pluronic containing hydrogel increases cutaneous wound healing in vitro and in vivo. *Biological trace element research*. 2014;162(1-3):72.
25. Demirci S, Doğan A, Karakuş E, Halıcı Z, Topçu A, Demirci E, et al. Boron and Poloxamer (F68 and F127) Containing Hydrogel Formulation for Burn Wound Healing. *Biological trace element research*. 2015;168(1):169.
26. Demirci S, Doğan A, Aydın S, Dülger E, Şahin F. Boron promotes streptozotocin-induced diabetic wound healing: roles in cell proliferation and migration, growth factor expression, and inflammation. *Molecular and cellular biochemistry*. 2016;417(1-2):119.

## Changes in liver function in patients with COVID-19 and the demographics of the disease in a mid-sized city of Turkey: A retrospective analysis

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### Abstract

**Objective:** To define the clinical characteristics of patients hospitalized with COVID-19 in ICU and ward and to evaluate the significance of liver function test results. A new disease caused by SARS-CoV-2 has been devastating the world affecting millions of patients worldwide and leading the significant mortality and morbidity.

**Material and Methods:** The demographic features of a cohort of 125 hospitalized patients between March 2020 and May 2020 were recorded. The clinical characteristics, laboratory findings, and mortality rates were analyzed.

**Results:** A total of 125 patients hospitalized for COVID-19 diagnosis where 113 (90.4%) were followed-up in the ward and 12 (9.6%) were in ICU were included in the study. The mean age of the patients was  $44.05 \pm 16.95$  and 88 (70.4%) of the patients were male. The most common symptoms were in the following order: cough in 80 patients (64%), dyspnea in 40 (32%) and fever in 33 (26.4%). The leukocyte and neutrophil counts were significantly higher in ICU patients with COVID-19 compared to patients in the ward ( $p=0.002$ ,  $p<0.05$ ; respectively). The CRP and D-Dimer levels were found elevated in ICU patients with COVID-19 than in the ward ( $p<0.05$ ;  $p<0.05$ , respectively). The AST level of patients with COVID-19 in ICU was significantly heightened than patients in the ward.

**Conclusion:** The present study revealed that patients with elevated AST level were at great risk of progressing to severe disease those require close monitoring.

**Keywords:** coronavirus, epidemiology, pandemics, Severe Acute Respiratory Syndrome, AST, ALT, Liver enzymes

### Introduction

Since February 2019 a new disease caused by severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) has been devastating the world. As of today, 12 million people have been diagnosed with coronavirus disease-19 (COVID-19) and more than 550,000 patients died. The US with 3 million infected patients and 132,000 deaths is the leading country followed by Brazil (1,2). In relation, approximately 200,000 confirmed cases have been detected in Turkey with 5,000 deaths which is low compared to several European Countries. Several reports have been coming about the clinical manifestations and blood biochemical results those might play an essential role for the estimation of the progress of the patients and the management of therapy. However, there is no specific treatment defined.

Thus, the distribution of the clinical and demographic characteristics of COVID-19 in different regions seems to be important to prevent the spread of the disease. A recent study showed that SARS-CoV-2 have the ability to bind to angiotensin-converting enzyme 2 (ACE2) on cholangiocytes causing disruption in the cell functions and triggering a systemic inflammatory response led to liver injury (1). In addition, various studies revealed elevated levels of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) enzymes ranging from 14% to 53% (3,4). Furthermore, pathological findings have also indicated moderate microvesicular steatosis and mild lobular and portal activity (5). The present study aims to report the clinical and biochemical course of the patients hospitalized with COVID-19, and the importance of liver function test results on the course of the disease.



## Materials and Methods

After approval of the Ethics Committee and Ministry of Health, this study was initiated with a retrospective design enrolled patient hospitalized with COVID-19 in the study hospital between March 2020 and May 2020. The study was completed in compliance with the World Medical Association Declaration of Helsinki. All patients had a positive PCR (polymerase chain reaction) test result of SARS-CoV-2. Patients aged below 18, had a history of liver malignancy, liver failure, cirrhosis, hepatitis, receiving medical treatment led to impairment in liver function tests were excluded. The demographic features, clinical and laboratory findings were obtained from hospital database. All data were recorded into a standardized data sheet.

### Statistical Analysis

The distribution of the variables was analyzed using Kolmogorov-Smirnov test. Quantitative data were presented as mean and standard deviation where qualitative data as median (interquartile range [IQR] 25%-75%) values, and also with numbers and percentage. The analysis of the demographic characteristics was conducted using Fisher's exact test. The comparison of the laboratory findings was completed by Mann-Whitney U test. All data was analyzed using Statistical Package for Social Sciences (SPSS) version 20 program. The significance level was set at  $p < 0.05$ .

### Results

A total of 125 patients were hospitalized for COVID-19 diagnosis where 113 (90.4%) were followed-up in the ward and 12 (9.6%) were in ICU. The mean age of the patients was  $44.05 \pm 16.95$  and 88 (70.4%) were male.

A total of nine patients (7.2%) had asthma where only two (1.6%) had chronic obstructive pulmonary disease (COPD). For cardiovascular comorbidities; 26 patients (20.8%) had hypertension where seven patients (7.2%) had coronary artery disease.

A total of 12 patients (9.6%) had type 1 or type 2 diabetes. A total of 41 patients (32.8%) had one or more comorbidities, of those 32 (28.3%) were received treatment in the ward and 9 (75%) in ICU ( $p = 0.002$ ). The most common symptoms were in the following order: cough in 80 patients (64%), dyspnea in 40 (32%) and fever in 33 (26.4%). Other symptoms including sore throat (6.4%), fatigue (4%) and myalgia (3.2%) are the less common symptoms (Table 1). The leukocyte and neutrophil count, and CRP and D-Dimer levels were significantly higher in ICU patients with COVID-19 compared to patients in ward ( $p = 0.002$ ;  $p < 0.05$ ;  $p < 0.05$ ;  $p < 0.05$ , respectively). The AST levels of patients with COVID-19 in ICU were significantly heightened than patients in ward. A total of 11 patients have died of COVID-19, and all of 11 have died while receiving treatment in ICU (Table 3). The mortality rate was 8.8% with a mean age of 66 years ranged from 50 to 81 years. Eight (72.7%) of the deaths had one or more comorbidities where three of them had none ( $p = 0.006$ ). One of the eight deaths with comorbidities had COPD and seven of them had mainly cardiovascular diseases. Patients with higher AST levels are prone to be died from COVID-19 compared to patients with normal AST levels ( $p = 0.01$ ; Table 4). It is also found that patients who died from COVID-19 had significantly higher AST levels than patients with COVID-19 discharged from hospital.

**Table 1.** Demographic characteristics of patients hospitalized with COVID-19

Variable	All patients	Ward	ICU	p
Age, years, (median, range)	44 (17 – 84)	41 (17 – 84)	64 (48 – 81)	$< 0.05^{*a}$
<b>Gender</b>				
Female	37 (30%)	35 (31%)	2 (17%)	0.507 <sup>b</sup>
Male	88 (70%)	78 (69%)	10 (83%)	
<b>Comorbidities</b>				
Hypertension	26 (21%)	20 (18%)	6 (50%)	<b>0.018<sup>*b</sup></b>
Diabetes	12 (9.6%)	8 (7.1%)	4 (33.3%)	<b>0.016<sup>*b</sup></b>
Coronary artery disease	7 (5.6%)	5 (4.4%)	2 (16.7%)	0.135 <sup>b</sup>
COPD	2 (1.6%)	1 (0.9%)	1 (8.3%)	0.183 <sup>b</sup>
Asthma	9 (7.2%)	8 (7.1%)	1 (8.3%)	$> 0.05^b$
<b>Symptoms</b>				
Fever	33 (26.4%)	31 (27.4%)	2 (16.7%)	0.515 <sup>b</sup>
Cough	78 (62.4%)	71 (62.8%)	7 (58.3%)	0.763 <sup>b</sup>
Dyspnea	40 (32%)	30 (26.5%)	10 (83.3%)	<b><math>&lt; 0.05^{*b}</math></b>
Fatigue	5 (4%)	4 (3.5%)	1 (8.3%)	0.402 <sup>b</sup>
Myalgia	4 (3.2%)	3 (2.7%)	1 (8.3%)	0.336 <sup>b</sup>
Sore throat	8 (6.4%)	8 (7.1%)	—	$> 0.05^b$
Headache	8 (6.4%)	8 (7.1%)	—	$> 0.05^b$
Abdominal pain	1 (0.8%)	—	1 (8.3%)	0.096 <sup>b</sup>

\* $p < 0.05$ . <sup>a</sup>Mann-Whitney U test. <sup>b</sup>Fisher's exact test. ICU, intensive care unit; COPD, chronic obstructive pulmonary disease.

**Table 2:** Laboratory results of the patients hospitalized with COVID-19

	Normal range <sup>†</sup>	Total	Ward	ICU	p
Leukocyte (10 <sup>3</sup> /μL)	4.8 – 10.8	6.23 (4.36 – 15.37)	6.08±.45	9.3±3.43	<b>0.002*</b>
Lymphocyte (10 <sup>3</sup> /μL)	1 – 4.8	1.46 (0.62 – 3.69)	1.65±0.71	1.72±0.95	0.887
Neutrophil (10 <sup>3</sup> /μL)	1.8 – 7.7	4.26 (2.72 – 13.6)	3.79±1.97	6.9±3.19	<b>&lt;0.05*</b>
NLR	–	2.43 (0.21 – 13.08)	2.7±1.74	5.44±4.2	<b>0.014*</b>
AST (U/L)	0 – 35	38.38 (18 – 71.2)	27.66±22.72	43.23±27.19	<b>0.022*</b>
ALT (U/L)	0 – 45	27.2 (19 – 63.3)	26.88±31.25	32.69±25.27	0.139
TBL (mg/dL)	0.3 – 1.2	0.59 (0.31 – 1.34)	0.46±0.25	0.59±0.3	0.118
ALB (g/dL)	3.5 – 5.2	4 (3.46 – 5.01)	4.4±0.38	3.43±0.59	<b>&lt;0.05*</b>
GGT (U/L)	0 – 55	33 (16 – 373)	26.76±23.57	78.25±100.6	<b>0.003*</b>
Creatinine (mg/dL)	0.88 – 1.06	0.88 (0.79 – 1.32)	0.9±0.19	0.97±0.41	0.557
CRP (mg/L)	0.3 – 1.2	36.89 (2.07 – 378)	11.35±16.21	142.94±126.52	<b>&lt;0.05*</b>
D-Dimer (ng/mL)	< 500	180 (10 – 718)	308.4±811.06	865.5±969.8	<b>&lt;0.05*</b>
ESR	0 – 30 mm/h	31 (10 – 70)	28.72±20.89	48.25±20.75	0.117

\*p<0.05. †Obtained from the medical biochemistry laboratory of the hospital. Mann-Whitney U test. ICU, intensive care unit; NLR, neutrophil to lymphocyte ratio.

**Table 3:** The outcomes of the patients hospitalized with COVID-19

		Total n (%)	Ward n (%)	ICU n (%)	p
Status	Discharge	114	113	1 (8.3)	<b>&lt;0.05*†</b>
	Died	11	–	11 (91.7)	
		Mean±SD	Mean±SD	Mean±SD	
Hospital stay (days)		7 (4 – 25)	7.88±2.93	11±5.73	<b>0.022*‡</b>

\*p<0.05. †Chi-square test. ‡Mann-Whitney U test. ICU, intensive care unit.

**Table 4:** The distribution of liver function test results

		Status		p
		Discharge n (%)	Died n (%)	
AST	Normal	94 (94.9)	5 (5.1)	<b>0.01*†</b>
	High	20 (76.9)	6 (23.1)	
ALT	Normal	96 (92.3)	8 (7.7)	0.393 <sup>‡</sup>
	High	18 (85.7)	3 (14.3)	
		Median (Range)	Median (Range)	p
AST		21.65 (8.3 – 160)	36.9 (15.7 – 96)	<b>0.026*‡</b>
ALT		18 (3.6 – 222)	21.9 (6.4 – 93)	0.187 <sup>‡</sup>

\*p<0.05. †Fisher's exact test. ‡Mann-Whitney U test. AST, aspartate aminotransferase; ALT, alanine aminotransferase.

Normal range for AST: Female (0 – 31 U/L); Male (0 – 35 U/L). Normal range for ALT: Female (0 – 34 U/L); Male (0 – 45 U/L). Ranges obtained from the medical biochemistry laboratory of the hospital.

## Discussion

To the best of our knowledge, this study was the first one including hospitalized patients infected with SARS-CoV-2 from the study site. An 9.6% of the patients required ICU admission. Hypertension with the rate of 50% and diabetes with 33.3% alone were the evident comorbidities for admission to the ICU. Although the most frequently detected symptom was cough (62.4%), the prevalence of dyspnea (83.3%) was higher in patients admitted to ICU. The overall mortality rate was 8.8%; and 81.8% of the deaths were above 60 years with a mean age of 66 years. Among the 11 deaths, three had no comorbidities where the rest of the deaths had one or two of the following: diabetes, hypertension, and coronary artery disease.

The leukocyte and neutrophil count, and AST, GGT, CRP, and D-Dimer levels were found elevated in patients admitted to ICU, where albumin level was lower in the same patients. Increased AST levels were shown in patients who died from COVID-19. Chronic diseases including hypertension and diabetes may trigger the conditions to prone the patient to be susceptible for being infected with SARS-CoV-2. It is known that chronic diseases share common pathways with infectious diseases including proinflammatory changes and the beginning of the immune response. As of today, the pathophysiology of the disease related to the comorbidities remains unexplained, only various hypotheses have been suggested.



Studies have demonstrated that hypertension is one of the most frequent comorbidities in patients with COVID-19 which had a reported prevalence of 15% and 23.4% in patients with severe COVID-19 (2,6). A meta-analysis showed that hypertension is rare in discharged patients compared to patients died from COVID-19 and a positive correlation was found between hypertension and disease severity as well as ICU admission(7). Li et al. revealed that the prevalence of hypertension in patients with COVID-19 is 17.1%, and it is two-folds higher in ICU patients compared to the ward (8). The present study showed a frequency of 21% among patients with COVID-19, with a 3.6 folds incidence in patients admitted to ICU than in the ward. It is very close to the Asian population reported in the previous studies. This finding could be associated with the usage of Angiotensin-Converting Enzyme (ACE) inhibitors and Angiotensin II Receptor Blockers (ARBs) among hypertensive patients which mostly result in an upregulation of ACE-II which is believed to be the location of SARS-CoV-2 surface antigen binding site.

Furthermore, diabetes is the other most prevalent comorbidity among patients with COVID-19. While the frequency of diabetes in patients with COVID-19 varies from one study to another, a recent meta-analysis demonstrated that the prevalence of diabetes is found higher in ICU patients than in the ward (9,10). Chinese data showed a prevalence between 15% and 25% and it is reported as two to four folds higher in ICU patients (6,11,12). Despite Chinese reports, a prevalence over the 50% has been shown in the United States in patients admitted to ICU with severe COVID-19 (13). One study from China revealed significant data that milder symptoms would be seen at the beginning of the infection such as fever was less prevalent leading a delay in the diagnosis of the disease (14). In addition, diabetes causes elevated inflammatory markers, tissue enzymes, and clotting abnormalities which may be associated with severe multi-organ damage and tendency to thromboembolic events as well as cytokine storm which is an aggravating factor for COVID-19 (14,15). A retrospective analysis showed that diabetes or cardiovascular disorders are detected at 85.5% of severe patients with COVID-19 than mild counterparts (16). Several studies indicate variable frequencies in which the present study showed a prevalence of 9.6% in the overall study group and 33.3% in ICU patients with COVID-19 compared to 7.1% in the ward (2,4,17,18). The difference between ICU and ward suggested that diabetes seems to be a prognostic factor for the poor prognosis of COVID-19. However, it is inevitable that this conclusion requires a study with larger sample size.

Moreover, COVID-19 appears to have variable clinical presentations where the most prevalent symptoms in the severe disease group were cough, fever, and fatigue. However, cough, fever, and dyspnea were the most frequent symptoms admitted to the ICU. The prevalence of dyspnea was reported as 67.2% in ICU admitted patients with COVID-19 compared to 10.2% in the ward. Dyspnea could be a sign of pulmonary involvement of the disease, where some reports indicated a phenomenon called "silent hypoxia" characterized by progressive respiratory failure

without the presence of dyspnea (19). Chinese data showed that the frequency of dyspnea was 37.2% in severe cases admitted to ICU which was 14.7% in cases of the ward (20,21). In contrast to the information obtained from Chinese studies, the present study revealed a prevalence of 83.3% in ICU admission for dyspnea, where 26.5% in the ward with an overall frequency of 32%. The higher percentage of dyspnea in ICU deserves an importance for predicting ICU admission, therefore future research should focus on the evaluation of early hospital admission and treatment management in patients with dyspnea.

Nevertheless, leukocyte count has been reported as decreased or normal in patients with COVID-19, however critical cases have leukocytosis frequently. A comprehensive study by Guan et al.(6) revealed that leukocytosis was observed in 25% of the severe cases. In general, and asymptomatic patients seem to be normal or decreased in patients with COVID-19 during admission and tend to elevate with disease progression (22,23). In contrast to the leukocyte count that was higher in ICU patients compared to the ward, the number of leukocytes is in the normal range in the present study. It can be declared that leukocytosis in ICU patients with COVID-19 may be associated with several factors including co-infections, medications, and the variable response of the immune system. Similar findings have been found for neutrophil numbers as it was higher in ICU patients than in the ward, however, it stayed in normal ranges. Most of the studies reported that neutrophil count was normal in non-severe cases but was elevated in patients with severe disease (24,25). In relation, neutrophil to lymphocyte ratio (NLR) is accepted as a prognostic factor for COVID-19. Studies indicated that it consistently elevated in severe patients with COVID-19 (25,26). Feng et al. reported that higher NLR detected during admission might be an independent predictor of severe pneumonia in patients with COVID-19 (27). Another study conducted by Zhang et al. noted that %94 of the patients who died from COVID-19 had an NLR over 5 (28). The current study has shown a higher NLR in severe patients admitted to the ICU than in the ward, hence it suggested that increased NLR may be a useful tool for predicting high-risk patients associated with the consistency and proven importance of it.

Another biochemical finding of the present study is slight hypoalbuminemia in ICU patients with COVID-19 which was normal in the ward. A cohort study from Spain revealed that hypoalbuminemia was common in critical patients infected with SARS-CoV-2. The authors also concluded that albumin levels might be associated with poor outcomes as increased mortality and hospital stay (29). It can be speculated that the slight decrease in albumin level can be related to the inflammatory response of the host to the SARS-CoV-2 infection.

Inflammatory markers including CRP and D-Dimer have been reported to be elevated in patients with COVID-19 (3,29,30). Studies revealed an increased CRP and D-Dimer ranged from 60.7% to 86.3% and 36.4% to 46.4% of the patients infected with SARS-CoV-2, respectively (2,4,6,31). Significantly higher levels were detected in the present study for CRP and D-Dimer in patients admitted to



ICU compared to the ward those also observed in various infections for severe cases as well.

The main study parameter of the present study is liver function enzymes especially AST and GGT which were also elevated in ICU patients. Studies indicated an elevated level of GGT that is considered as the cholangiocyte-related enzyme. However, GGT is mainly located in the cell membranes of several tissues such as the heart, brain, kidneys, pancreas, bile duct, spleen, gall bladder and seminal vesicles. Therefore, merely a higher GGT level could be associated with drug-induced liver injury or damage in other organs (32). In contrast, an elevated AST level was more frequent in severe cases where higher level of AST is reported to be more than three-folds according to the normal range in 5.66% of the patients with COVID-19 during hospitalization. Similarly, a three-folds higher level of AST was found in ICU patients compared to the ward and 23.1% of the patients those also have elevated AST died from COVID-19. Limited data is available related to the variability of liver function tests. In this context, Chen et al. showed no changes in AST level were found in patients with COVID-19 (33). However, Cai et al. (34) revealed a three-folds increase in 12% of the patients with SARS-CoV-2 infection. Additionally, the authors also noted that medications including non-steroidal anti-inflammatory drugs, antibiotics, and herbal medications may induce liver injury and were related to disease severity in cases with abnormal liver test findings. Therefore, it is suggested to closely follow-up cases receiving these particular treatments, particularly in those who had impaired liver function test levels at admission to the hospital.

Consequently, the present study revealed an overall mortality rate of 8.8% with a number of 11 cases of those all in admitted to the ICU. The mortality rate of the ICU was 91.7%, unfortunately, one patient was merely discharged from ICU. Several studies reported dispersed mortality rates where a recent Chinese study noted 31.4% and it is 21% in a US study. Another study reported an apparent mortality rate of 4% in China, 13% in Italy, 11% in Spain and 15% in France (35). In relation, distinct outcomes have been obtained from several studies as the mortality rate of ICU in China is reported as 37.7%, Italy as 25.6%, the USA as 23.6%, Spain as 29.2% and Denmark as 41.2%, Germany as 24.3% and UK as 8% (36). The unexpected mortality rate in ICU observed in this study could highly be associated with the smaller sample size compared to the studies in other countries. It should be closer or lower after enrolling more cases in the study.

Previous studies included age, gender, and comorbid diseases as the risk factors of COVID-19, this is one of the reports to exhibit abnormal liver function test results those associated with severe disease. It can be speculated that SARS-CoV-2 may lead to severe multi-organ dysfunction in humans and the findings of the present study could support some part of this hypothesis.

The present study has some limitations. First, the biggest limitation of this study is the very small sample size thus inhibits the meaningful conclusions obtained from the

present study. Second, patients enrolled in this study were from a single, large city of Turkey restricting the generalizability of the results to the rural areas or other regions. Lastly, most of the patients in our cohort were Caucasian, hence study findings can not be extended to other ethnicities. Large prospective multiethnic studies are required to provide comprehensive data about the COVID-19 pandemic.

## Conclusions

We present a very first reported data on all cases infected with SARS-CoV-2 from a mid-size city of Turkey. Cough, dyspnea and fever are the most observed symptoms of the disease. Hemocytometric changes particularly elevated NLR in patients infected with SARS-CoV-2 might be a useful tool for the prediction of the disease progression. The remarkable outcome of the present study is that patients with elevated AST level were at great risk of progressing to severe disease those require close monitoring.

**Author Contributions:** HAD: Project design, Review of the literature, Data collection and statistical analyzes HAD: Writing and Revisions

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## References

1. Zhao B, Ni C, Gao R, Wang Y, Yang L, Wei J, et al. Recapitulation of SARS-CoV-2 infection and cholangiocyte damage with human liver ductal organoids. Vol. 11, Protein & cell. 2020. p. 771–5.
2. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet* (London, England). 2020 Feb;395(10223):497–506.
3. Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. *Lancet* (London, England). 2020 Feb;395(10223):507–13.
4. Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, et al. Clinical Characteristics of 138 Hospitalized Patients With 2019 Novel Coronavirus-Infected Pneumonia in Wuhan, China. *JAMA*. 2020 Mar;323(11):1061–9.
5. Xu Z, Shi L, Wang Y, Zhang J, Huang L, Zhang C, et al. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. Vol. 8, *The Lancet. Respiratory medicine*. 2020. p. 420–2.
6. Guan W, Ni Z, Hu Y, Liang W, Ou C, He J, et al. Clinical Characteristics of Coronavirus Disease 2019 in China. *N Engl J Med* [Internet]. 2020 Feb 28;382(18):1708–20. Available from: <https://doi.org/10.1056/NEJMoa2002032>
7. Hu Y, Sun J, Dai Z, Deng H, Li X, Huang Q, et al. Prevalence and severity of corona virus disease 2019 (COVID-19): A systematic review and meta-analysis. *J Clin Virol Off Publ Pan Am Soc Clin Virol*. 2020 Jun;127:104371.
8. Li B, Yang J, Zhao F, Zhi L, Wang X, Liu L, et al. Prevalence and impact of cardiovascular metabolic diseases on COVID-19 in China. *Clin Res Cardiol*. 2020 May;109(5):531–8.

9. Fadini GP, Morieri ML, Longato E, Avogaro A. Prevalence and impact of diabetes among people infected with SARS-CoV-2. Vol. 43, *Journal of endocrinological investigation*. 2020. p. 867–9.
10. Huang I, Lim MA, Pranata R. Diabetes mellitus is associated with increased mortality and severity of disease in COVID-19 pneumonia - A systematic review, meta-analysis, and meta-regression. *Diabetes Metab Syndr*. 2020;14(4):395–403.
11. Wu C, Chen X, Cai Y, Xia J, Zhou X, Xu S, et al. Risk Factors Associated With Acute Respiratory Distress Syndrome and Death in Patients With Coronavirus Disease 2019 Pneumonia in Wuhan, China. *JAMA Intern Med*. 2020 Jul;180(7):934–43.
12. Yang X, Yu Y, Xu J, Shu H, Xia J, Liu H, et al. Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study. *Lancet Respir Med*. 2020 May;8(5):475–81.
13. Bhatraju PK, Ghassemieh BJ, Nichols M, Kim R, Jerome KR, Nalla AK, et al. Covid-19 in Critically Ill Patients in the Seattle Region - Case Series. *N Engl J Med*. 2020 May;382(21):2012–22.
14. Guo W, Li M, Dong Y, Zhou H, Zhang Z, Tian C, et al. Diabetes is a risk factor for the progression and prognosis of COVID-19. *Diabetes Metab Res Rev*. 2020 Mar;e3319.
15. Ye Q, Wang B, Mao J. The pathogenesis and treatment of the 'Cytokine Storm' in COVID-19. *J Infect*. 2020 Jun;80(6):607–13.
16. Wu J, Li W, Shi X, Chen Z, Jiang B, Liu J, et al. Early antiviral treatment contributes to alleviate the severity and improve the prognosis of patients with novel coronavirus disease (COVID-19). *J Intern Med*. 2020 Jul;288(1):128–38.
17. Zhang J-J, Dong X, Cao Y-Y, Yuan Y-D, Yang Y-B, Yan Y-Q, et al. Clinical characteristics of 140 patients infected with SARS-CoV-2 in Wuhan, China. *Allergy*. 2020 Jul;75(7):1730–41.
18. Wang X, Fang J, Zhu Y, Chen L, Ding F, Zhou R, et al. Clinical characteristics of non-critically ill patients with novel coronavirus infection (COVID-19) in a Fangcang Hospital. *Clin Microbiol Infect Off Publ Eur Soc Clin Microbiol Infect Dis*. 2020 Aug;26(8):1063–8.
19. Gattinoni L, Chiumello D, Caironi P, Busana M, Romitti F, Brazzi L, et al. COVID-19 pneumonia: different respiratory treatments for different phenotypes? Vol. 46, *Intensive care medicine*. 2020. p. 1099–102.
20. Li K, Wu J, Wu F, Guo D, Chen L, Fang Z, et al. The Clinical and Chest CT Features Associated With Severe and Critical COVID-19 Pneumonia. *Invest Radiol [Internet]*. 2020;55(6):327–331. Available from: <https://europepmc.org/articles/PMC7147273>
21. Xu Y-H, Dong J-H, An W-M, Lv X-Y, Yin X-P, Zhang J-Z, et al. Clinical and computed tomographic imaging features of novel coronavirus pneumonia caused by SARS-CoV-2. *J Infect*. 2020 Apr;80(4):394–400.
22. Hu Z, Song C, Xu C, Jin G, Chen Y, Xu X, et al. Clinical characteristics of 24 asymptomatic infections with COVID-19 screened among close contacts in Nanjing, China. *Sci China Life Sci*. 2020 May;63(5):706–11.
23. Tan M, Liu Y, Zhou R, Deng X, Li F, Liang K, et al. Immunopathological characteristics of coronavirus disease 2019 cases in Guangzhou, China. *Immunology*. 2020 Jul;160(3):261–8.
24. Lo IL, Lio CF, Cheong HH, Lei CI, Cheong TH, Zhong X, et al. Evaluation of SARS-CoV-2 RNA shedding in clinical specimens and clinical characteristics of 10 patients with COVID-19 in Macau. *Int J Biol Sci*. 2020;16(10):1698–707.
25. Khartabil TA, Russcher H, van der Ven A, de Rijke YB. A summary of the diagnostic and prognostic value of hemocytometry markers in COVID-19 patients. *Crit Rev Clin Lab Sci [Internet]*. 2020 Aug 17;57(6):415–31. Available from: <https://doi.org/10.1080/10408363.2020.1774736>
26. Song R, Han B, Song M, Wang L, Conlon CP, Dong T, et al. Clinical and epidemiological features of COVID-19 family clusters in Beijing, China. *J Infect*. 2020 Aug;81(2):e26–30.
27. Feng Z, Yu Q, Yao S, Luo L, Duan J, Yan Z, et al. Early Prediction of Disease Progression in 2019 Novel Coronavirus Pneumonia Patients Outside Wuhan with CT and Clinical Characteristics. *medRxiv [Internet]*. 2020; Available from: <https://www.medrxiv.org/content/early/2020/02/23/2020.02.19.20025296>
28. Zhang B, Zhou X, Qiu Y, Song Y, Feng F, Feng J, et al. Clinical characteristics of 82 cases of death from COVID-19. *PLoS One*. 2020;15(7):e0235458.
29. de la Rica R, Borges M, Aranda M, del Castillo A, Socias A, Payaras A, et al. Low albumin levels are associated with poorer outcomes in a case series of COVID-19 patients in Spain: a retrospective cohort study. *medRxiv [Internet]*. 2020 Jan 1;2020.05.07.20094987. Available from: <http://medrxiv.org/content/early/2020/07/15/2020.05.07.20094987.abstract>
30. Ge H, Wang X, Yuan X, Xiao G, Wang C, Deng T, et al. The epidemiology and clinical information about COVID-19. *Eur J Clin Microbiol Infect Dis Off Publ Eur Soc Clin Microbiol*. 2020 Jun;39(6):1011–9.
31. Clinical findings in a group of patients infected with the 2019 novel coronavirus (SARS-Cov-2) outside of Wuhan, China: retrospective case series. *BMJ*. 2020 Feb;368:m792.
32. Dillon JF, Miller MH. Gamma glutamyl transferase "To be or not to be" a liver function test? *Ann Clin Biochem*. 2016 Nov;53(6):629–31.
33. Chen L, Liu HG, Liu W, Liu J, Liu K, Shang J, et al. [Analysis of clinical features of 29 patients with 2019 novel coronavirus pneumonia]. *Zhonghua jie he hu xi za zhi = Zhonghua jiehe hu xuzhi = Chinese J Tuberc Respir Dis*. 2020 Feb;43(0):E005.
34. Cai Q, Huang D, Yu H, Zhu Z, Xia Z, Su Y, et al. COVID-19: Abnormal liver function tests. *J Hepatol*. 2020 Sep;73(3):566–74.
35. Ceylan Z. Estimation of COVID-19 prevalence in Italy, Spain, and France. *Sci Total Environ*. 2020 Aug;729:138817.
36. Quah P, Li A, Phua J. Mortality rates of patients with COVID-19 in the intensive care unit: a systematic review of the emerging literature. Vol. 24, *Critical care (London, England)*. 2020. p. 285.

## Clinical characteristics and postoperative complications of patients who underwent laparoscopic sleeve gastrectomy due to obesity; A Single Center Experience

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### Abstract

**Objective:** Obesity, which is considered as a chronic disease today, has become an epidemic problem especially in developed countries. Laparoscopic sleeve gastrectomy (LSG) is currently one of the most common bariatric procedures in obesity treatment. This study aims to present our demographic, clinical, histopathological and postoperative early and late complications of patients who underwent LSG.

**Material and Methods:** Patients who underwent LSG due to obesity between March 2018 and December 2019 in our clinic were included in the study. The patients' age, gender, comorbid diseases, preoperative body mass index (BMI) and length of hospital stay, postoperative complications and pathology results were recorded retrospectively.

**Results:** Of the 278 patients included in the study, 201 (72.3%) were female, 77 (27.6%) were male and the mean age was 36.2 (min: 18-max: 60 years). The mean BMI of the patients was 46.4 (min: 35.2-max: 75.1). The average hospital stay of the patients was 4.2 days (3-13 days). When the early and late complications of the patients are examined; It was observed that 25 (8.9%) patients developed early postoperative complications, and 2 (0.7%) patients developed late postoperative complications.

**Conclusion:** LSG is a reliable surgical method with low mortality and morbidity rates. It is important that we do not have mortality and that our morbidity is within acceptable limits. Bariatric surgery is an effective and reliable application in the treatment of obesity and metabolic surgery today, in terms of its results, if the right patient is selected and performed in experienced centers.

**Keywords:** Obesity, Laparoscopic sleeve gastrectomy, Postoperative complication

### Introduction

Obesity has become an epidemic problem, especially in developed countries, and ranks second after smoking among preventable diseases that cause death. Obesity is a disease that must be fought due to the high cost of treatment and the diseases it brings. Obesity is accepted as a chronic disease that affects life expectancy and comfort negatively (1,2). Obesity is not just a weight problem, it is increasing rapidly especially in young people, and many chronic diseases appear at an earlier age. Venous circulation disorders, especially diabetes and hypertension, coagulopathies and cardiac disorders are the main ones (3). While 67% of the population of the United States of America (USA) is in the overweight or obese group, this rate is 40-50% in many European countries (4).

A sustainable diet has a very important place in the treatment of obesity, but it seems almost impossible to do this in severe obesity.

Surgical treatment emerges as an appropriate and effective option in the treatment of this endemic disease, which causes premature deaths together with the diseases it brings (5,6). These risks should be taken into consideration when planning surgical treatment. In the treatment of morbid obesity, the surgeon has the necessary consultant specialist physicians and special operating room equipment and equipment; needs adequate follow-up facilities. The multidisciplinary approach is important. The main goal in obesity surgery is to achieve the desired body weight in patients. Laparoscopic sleeve gastrectomy (LSG) is the most commonly used bariatric surgical technique today, and it is a restrictive procedure because it is an intervention in which gastric volume is reduced (7).

After surgery, ghrelin hormone produced from the gastric fundus is not produced, it plays an important role in weight loss and resolution of metabolic disorders.



During surgery, after the stomach fundus is resected along the main curve with the straight line up to the gastric-esophageal junction, this hormone cannot be secreted, which strongly reduces the desire to eat and consequently leads to efficient and continuous weight loss. LSG has taken its place among metabolic surgical procedures as in laparoscopic roux-en-y gastric bypass (LRYGB) and biliopancreatic diversion with duodenal switch (BPD-DS). With this study, we aimed to present our demographic, clinical, histopathological and postoperative early and late complications of our patients who underwent LSG in our center.

## Materials and methods

### Study design and patients

The data of patients who underwent bariatric surgery for obesity between March 2018 and December 2019 in the general surgery clinic of the Şanlıurfa Training and Research Hospital were recorded retrospectively. The study was conducted in accordance with the Helsinki Declaration. Patients who underwent LSG were included in the study. Patients with missing file and computer data, patients who underwent revision surgery with LRYGB, BPD-DS were excluded from the study. Patients' age, gender, comorbid diseases, preoperative body mass index (BMI) and length of hospital stay, postoperative complications and pathology results were recorded retrospectively through file and computer records. Determined as the criteria for commissioning; Patients with a BMI of 35 kg / m<sup>2</sup> and above, with medical problems such as hypertension, diabetes mellitus and sleep apnea, and patients under 65 years of age with a BMI of 40 kg / m<sup>2</sup> and above were included in the operation. All patients were consulted with psychiatry, chest diseases, cardiology, internal medicine or endocrinology clinics before surgery. An informed consent form was obtained from the patient and / or patient relative for the surgical intervention. Surgical time, postoperative hospital stay, intraoperative blood transfusion, postoperative blood transfusion, morbidity and complications in the first 30 days of the patients were also determined. Postoperative complications were graded according to Clavien-Dindo classification (8).

### Body mass index classification

BMI obtained from anthropometric measurements, body weight and height measurements, accepted by the World Health Organization in Obesity Studies, is the most common and valid standard height-weight index that can be applied to all individuals without gender discrimination. According to the BMI values accepted by the World Health Organization, individuals; The cases can be classified as underweight, normal, overweight and obese as well as obese individuals (9).

### Surgical technique

In patients we used LSG as a surgical method, the operation was performed under general anesthesia after 8 hours of fasting before surgery. 1x0.6 IU was administered to all patients before surgery and 1x0.6 IU low molecular weight heparin (Clexane® and / or Oksapar®) subcutaneously for 5 days postoperatively. The patients were positioned with

their legs open and fifteen mm-Hg intraabdominal pressure was used. Surgery was performed with the help of 5 trocars (Covidien®, Ethicon®) in the surgical procedure. A liver retractor was inserted with a subxiphoid 5mm trocar, an image trocar was inserted approximately 3-4 cm proximal from the umbilicus superior, and 12 mm from the right subcostal midclavicular line (for stapler entry), 10 mm from the left subcostal midclavicular line, and left subcostal mid-axillary line. 5 mm trocar inserts were made. The gastrocolic ligament was opened using ultrasonic energy devices (Harmonic, Ethicon®, Ligasure, Covidien®), adjacent to the greater curvature of the stomach. The gastrocolic ligament was cut from the pylorus approximately 3-4 cm proximal to the angle of His and the stomach was freed by the greater curvature. With the help of 38 F bougie laparoscopic linear staplers (Echelon 45 and 60 Endopath Stapler®, Ethicon Endosurgery, Cincinnati, OH) placed through the orogastric route, approximately 2-3 cm proximal to the fundus was included, and the stomach was resected on the vertical line up to the angle of sensation. Hemoclip was applied to bleeding points on the staple line for hemostasis. No intervention was applied to the stapler line for extra support. The gastric specimen was removed from its 12 mm trocar in the right midclavicular line. Leak test was done with methylene blue. A Jackson-Pratt drain was placed along the stapler line. In addition, bilateral laptop compression stockings were worn in the pre-operative ward until the patients were mobilized. After the leak test with methylene blue on the postoperative 2nd or 3rd day, liquid foods were started and the drains were removed one day later.

Statistical analysis: Statistical Package for the Social Sciences (SPSS 21 Inc., Chicago, IL, USA) computer software was used for bio-statistical analyses. When the data were presented as mean values their standard deviation values were given, when they were presented as median values their minimum-maximum values were also stated.

## Results

Total 278 patients included in the study, 201 (72.3%) were female, 77 (27.6%) were male and the mean age was 36.2 (min: 18-max: 60 years). The mean BMI of the patients was 46.4 (min: 35.2-max: 75.1). When the comorbid diseases of the patients were examined, hypertension in 44 (15.8%) patients, cardiac comorbidity in 6 (2.1%) patients, pulmonary comorbidity in 28 (10%) patients, diabetes mellitus in 41 (14.7%), hypothyroidism in 14 (5%) patients, 2 (%) patients. 0.7), the patient had hyperparathyroidism. In the intraoperative period, a mass lesion of approximately 2-3 cm in size was observed on the greater curvature of the stomach in one (0.35%) patient. Since the lesion was in the specimen, no extra intervention was made. The pathology result of the patient was reported as a benign peripheral nerve sheath tumor with myxoid features. When the histopathological results of all patients were examined, it was observed that 83 (29.8%) patients had normal gastric tissue, 195 (70.1%) patients had chronic gastritis, and 66 (23.7%) patients were helicobacter pylori positive. Perioperative hiatal hernia was detected in one patient and repair was performed during surgery.



The average hospital stay of the patients was 4.2 days (3-13 days). When the early and late complications of the patients are examined; It was observed that 25 (8.9%) patients developed early postoperative complications, and 2 (0.7%) patients developed late postoperative complications. In early period compositions; Postoperative hemorrhage was observed in 16 (5.7%) patients.

Despite close postoperative follow-up and blood replacement in 5 patients, relaparoscopy was performed due to hemodynamic instability. 11 (3.9%) patients were followed up conservatively and ES replacement was performed. Since there was no active bleeding from the drain and the patients' vital signs were stable, exploration was not performed.

Postoperative intraabdominal abscess (Figure-1) developed in one (0.35%) patient and percutaneous abscess drainage was performed. It was observed that pneumonia developed in one (0.35%) patient. Postoperative atelectasis developed in two (0.7%) patients.

One (0.35%) patient developed necrosis at the epigastric trocar site due to cautery burn and wound debridement was performed. Trocar site infection was observed in 3 (1%) patients, and postoperative pancreatitis was observed in 1 (0.35%) patient.

When early complications are classified according to Clavien-Dindo Classification; Three patients had grade I, 15 patients had grade II, 2 patients had grade IIIA, and 5 patients had grade IIIB. Late complications; Two (0.7%) patients were diagnosed with polyneuropathy as a result of electromyography (EMG) performed due to the symptoms of muscle weakness and myalgia at the 2nd and 5th months postoperatively.

Clinical improvement was observed in the patients after thiamine replacement. The postoperative complication rate was 9.7%. The major complication rate was 2.5% and the minor complication rate was 7.1%. No mortality due to surgery was observed in any patient in the postoperative period. The demographic, clinical, histopathological characteristics and postoperative complications of the patients are presented in table-1.

**Table 1:** Demographic, clinical, histopathological and postoperative complications of the patients

Variable			Numeric (Total:278)	Percentage (%)
Age (year) (median)			36,2 years	
Sex	Female		201	72,3
	Male		77	27,6
Co-morbidities	DM		41	14,7
	HT		44	15,8
	Pulmonary comorbidity		28	10
	Cardiac comorbidity		6	2,1
	Hypothyroidism		14	5
	Hyperparathyroidism		2	0,7
BMI (Body Mass Index)			46,4	
Hospitalization			4,2 days	
Postoperative early complications	Postoperative bleeding		16	5,7
	Abdominal abscess		1	0,35
	Postoperative pancreatitis		1	0,35
	Pneumonia		1	0,35
	Postoperative atelectasis		2	0,7
	Trocar site infection		3	1
	Necrosis due to cautery burn at the trocar site		1	0,5
Postoperative late complications	Polyneuropathy		2	0,7
Claviendindo classification (Postoperative complications)	Grade-1		3	1
	Grade-2		15	5,3
	Grade-3a		2	0,7
	Grade-3b		5	1,7
	Grade-4		-	-
	Grade-5		-	-
Histopathological results	pathology	Normal gastric tissue	83	29,8
		Chronic gastritis	195	70,1
		H.pylori positive	66	23,7
		Gastro intestinal stromal tumor	1	0,35





**Figure 1:** Intraabdominal abscess

## Discussion

In societies with a high socio-economic level, lack of physical activity and changes in nutritional habits together with technological advances constitute the most important environmental causes of obesity. In societies with low socio-economic levels, limited opportunities to find suitable food and consequently one-way nutrition of people lead to an increase in the incidence of obesity. Basal metabolic rate slows down with advanced age, energy consumption decreases, and obesity frequency increases. In this case, if energy intake is not reduced, body weight increases with age (10,11).

Since obesity and cholelithiasis are common, preoperative imaging of the gallbladder is important. It has been reported that the risk of gallstones formation in patients during the rapid weight loss period is 38% in the 6-month follow-up (12). Simultaneous cholecystectomy was performed in 4 patients. The possibility of cholelithiasis should be considered in addition to the effects of the surgery performed on patients' complaints of abdominal pain and bloating, dyspeptic. Routine ultrasonography control or even prophylaxis with cholesterol-sparing drugs is recommended in the 2nd and 5th years after bariatric surgery (13).

One of the issues that reduce the treatment success rate in obese patients is eating behavior and patient compliance. Publications investigating the relationship between obesity and neural circuits report a connection between pleasure and reward circuits and the success of the operation (14). Although obesity is not seen as a surgical problem, it will be difficult to achieve expected weight loss.

Although the concepts of successful and unsuccessful bariatric procedures have been recognized until recently, many researchers consider the 15–50% weight loss resulting from this procedure to be successful (15). In our series, this rate is 42.6% and it is seen that our transactions are generally successful.

The most important and early complications of LSG are bleeding and leakage (2%) and it is often seen in the area close to the suture angle. Placing the last stapler line close to the esophagus, incisura angularis stenosis, and twisting of the tubular stomach can be considered among the causes of leakage. Many methods have been tried to prevent this complication, but no technique has been defined to eliminate the possibility of complications. Stapler line disintegration occurs due to mechanical and ischemic reasons. Mechanical leaks occur within the first 2 days, while ischemia sources decomposition occurs between the 5th-7th days when the inflammatory and fibrotic response is most intense (16). It is recommended to suture the stapler line with 2/0 Vicryl or silk with a continuous technique to prevent bleeding and leak complications (16). They also reported that the use of absorbable polymer membranes with the stapler strengthened the stapler line and significantly prevented bleeding and leakage (17). In our patients, no routine suture was placed on the stapler line, and sutured in case of bleeding or perioperative suspicion. Omarov et al. (18) reported that there was 1.1% anastomotic leakage and 1.1% bleeding. Bleeding requiring relaparoscopy was observed in 5 (1.7%) patients. Conservative ES replacement was performed in 11 (3.9%)

patients without the need for surgery. Laparoscopic abscess drainage was performed in 1 patient due to intraabdominal abscess. The abscess seen in one of our patients was thought to be due to leakage from the specimen during the exit of the surgery specimen due to the presence of the abscess in the right quadrant and the passage of the contrast material administered to the small intestines during CT scan and the absence of extralumination from the stapler line. There was no leak in our cases.

The limitation of our study is that it is retrospective and single centered. In conclusion, LSG, which can be applied in obesity surgery, is a safe surgical method with low mortality and morbidity rates. However, LSG is a unique procedure that may have serious postoperative complications and postoperative morbidity is likely to be encountered. It is important that we do not have mortality and that our morbidity is within acceptable limits. Bariatric surgery is an effective and reliable application in the treatment of obesity and metabolic surgery today, in terms of its results, if the right patient is selected and performed in experienced centers.

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## References

- World Health Organization, Obesity and Overweight, WHO, 2018 (updated February 2015). Available from: <https://www.who.int/en/news-room/fact-sheets/detail/obesity-and-overweight>
- Ali M, El Chaar M, Ghiassi S, Rogers AM; American Society for Metabolic and Bariatric Surgery Clinical Issues Committee. American Society for Metabolic and Bariatric Surgery updated position statement on sleeve gastrectomy as a bariatric procedure. *Surg Obes Relat Dis*. 2017 Oct;13(10):1652-1657
- Fobi MA. Surgical treatment of obesity: a review. *J Natl Med Assoc* 2004;96:61-75
- World Health Organization. WHO global database on body mass index.
- Adams T, Gress M, Smith S, et al. Long-term mortality after gastric bypass surgery. *N Engl J Med* 2007;357:753-761.
- Schauer PR, Kashyap SR, Wolski K, Brethauer SA, Kirwan JP, Pothier CE, Thomas S, Abood B, Nissen SE, Bhatt DL. Bariatric surgery versus intensive medical therapy in obese patients with diabetes. *N Engl J Med* 2012;366:1567-1576.
- Lazzati A, Guy-Lachuer R, Delaunay V, et al. Bariatric surgery trends in France: 2005-2011. *Surg Obes Relat Dis* 2014;10(2):328-34.
- Guetta O, Vakhrushev A, Dukhno O, Ovnat A, Sebbag G. New results on the safety of laparoscopic sleeve gastrectomy bariatric procedure for type 2 diabetes patients. *World Journal of Diabetes*.2019; 10(2), 78.
- World Health Organization. "Obesity: preventing and managing the global epidemic: report of a WHO consultation on obesity, Geneva, 3-5 June 1997." (1998).
- Baysal A. Aksoy M, Bozkurt N ve ark. *Diyet El Kitabı*. Ankara: Hatiboğlu Yayınevi: 2011.
- Gagner M, Hutchinson C, Rosenthal R. Fifth International Consensus Conference: current status of sleeve gastrectomy. *Surg Obes Relat Dis* 2016;12:750-6.
- Bariyatrik Cerrahi Klavuzu. *Türkiye Endokrinoloji ve Metabolizma Derneği*. 2018
- Melmer A, Sturm W, Kuhnert B et al. Incidence of gallstone formation and cholecystectomy 10 years after bariatric surgery. *Obes Surg* 2015;25(7):1171-6.
- Larjani S, Spivak I, Hao Guo M, Aliarzadeh B, Wang W, Robinson S, et al. Preoperative predictors of adherence to multidisciplinary follow-up care postbariatric surgery. *Surg Obes Relat Dis* 2016;12:350-6.
- Benaiges D, Goday A, Pedro-Botet J, Más A, Chillarón JJ, Flores-Le Roux JA. Bariatric surgery: to whom and when? *Minerva Endocrinol* 2015;40:119-28.
- Baker RS, Foote J, Kemmeter P, et al. The science of stapling and leaks. *Obes Surg* 2004;14(10):1360-6
- Consten EC, Gagner M, Pomp A, et al. Decreased bleeding after laparoscopic sleeve gastrectomy with or without duodenal switch for morbid obesity using a stapled buttressed absorbable polymer membrane. *Obes Surg*. 2004 Nov-Dec;14(10):1360-6.
- Omarov T, Ibrahimova A, Abdullayev A, Bayramov N, Şahin TT. Bariatric surgery in Azerbaijan: a single center experience. *Laparoscopic Endoscopic Surgical Science (LESS)*. 2017; 24(4), 122-127.

## Idiopathic temporal bone encephalocele: Diagnosed and treated during cochlear implantation

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### Abstract

**Objective:** Meningoencephaloceles of the temporal bone are rare entities. There are two main categories, congenital and acquired. Acquired meningoencephaloceles are more common due to iatrogenic injury to the tegmen tympani which is a common complication during mastoidectomy.

**Case Presentation:** We present a case of an idiopathic meningoencephalocele that was diagnosed and treated successfully during cochlear implantation via transmastoid approach. Fascia lata graft was used to reconstruct the deficit of tegmen tympani. Audiological outcome has been improved.

**Conclusions:** Idiopathic Meningoencephaloceles present without specific clinical symptomatology and as a result diagnosis delay significant. In literature, there are various surgical approaches with mixed results.

**Keywords:** Temporal bone, Idiopathic Meningoencephalocele, Sensorineural Hearing Loss

### Introduction

Encephaloceles are classified into cranial and basal. Cranial encephaloceles are the most common and mostly involve the occipital and frontonasal regions. Basal encephaloceles exhibit herniations of the brain or meninges without disconnection of subarachnoid(1,2). Temporal lobe encephaloceles are types of basal encephaloceles and subcategorized into either lateral or midline lesions. Lateral temporal lobe encephaloceles are more common than midline encephaloceles. They may clinically present with clear otorrhea, rhinorrhea, conductive hearing loss, chronic otitis media, middle ear effusion, seizures, meningitis, and intracranial abscess; however, they may also be asymptomatic and incidentally discovered on imaging(1,3). Midline temporal lobe encephaloceles, such as temporosphenoidal encephaloceles, may present as a mass in sphenoid sinus or nasopharynx, with clear rhinorrhea, focal neurologic deficits, or any of the previously mentioned infectious etiologies. Spontaneous meningoencephaloceles are either congenital or idiopathic presenting(1). In idiopathic cases, they are usually located at the tegmen tympani or the tegmen mastoideum. The purpose of the present article is to present a case of meningoencephalocele discovered in temporal bone during cochlear implant surgery. We described our management to restore the deficit effectively.

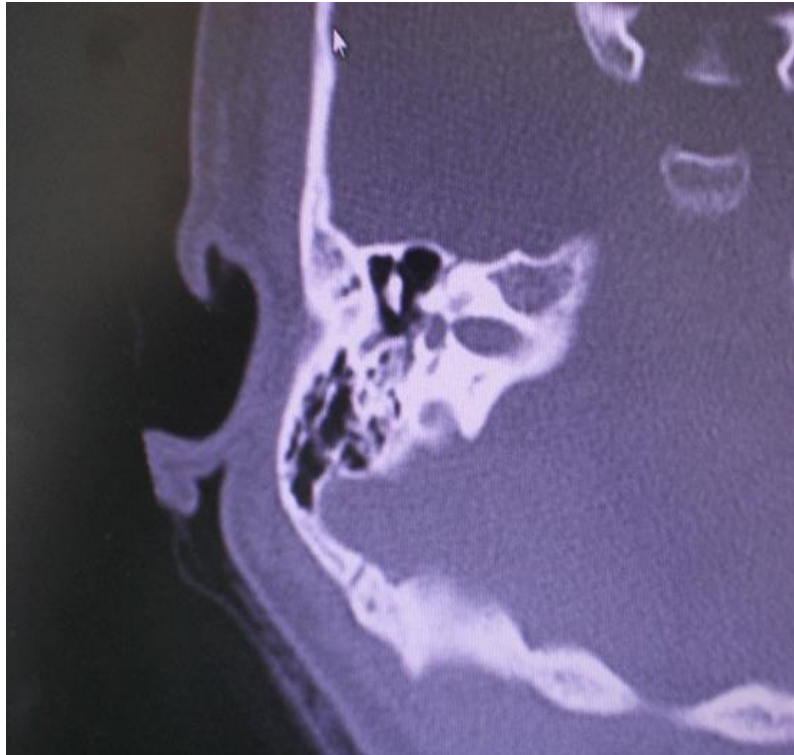
### Case Presentation

A 24-year-old female, teacher referred to our department with progressive right ear fullness started 12 months ago. She denied vertigo or purulent ear discharge. A history of multiple hearing loss episodes in the past 7 years was recorded. Intravenous treatment with corticosteroids was the treatment of choice because sudden hearing loss was considered the diagnosis. Due to social and professional reasons she was uncomfortable with the hearing aids. On clinical examination, cranial nerves function was normal. Nystagmus and fistula test were all absent, and endoscopic ear examination does not exhibit a pulsatile mass or other pathologic signs. There was no visible leak. Pure tone audiogram revealed mild to severe sensorineural hearing loss with right-sided 20 dB air-bone gap hearing loss. Magnetic Resonance Imaging (MRI) and Computed Tomography (CT) revealed no soft tissue in the middle ear cleft (Figure 1). Based on history and audiometry findings, cochlear implantation was indicated. A postauricular incision was made, and the mastoid cavity was exposed. As the tegmen tympani was revealed, we have found two small meningoencephaloceles, one of them was in contact with the ossicular ossicle (Figure 2). The masses were removed and the defect was closed with a cartilage graft locked in place, and two layers of Solvent-Dehydrated fascia lata were inserted as an underlay technique.

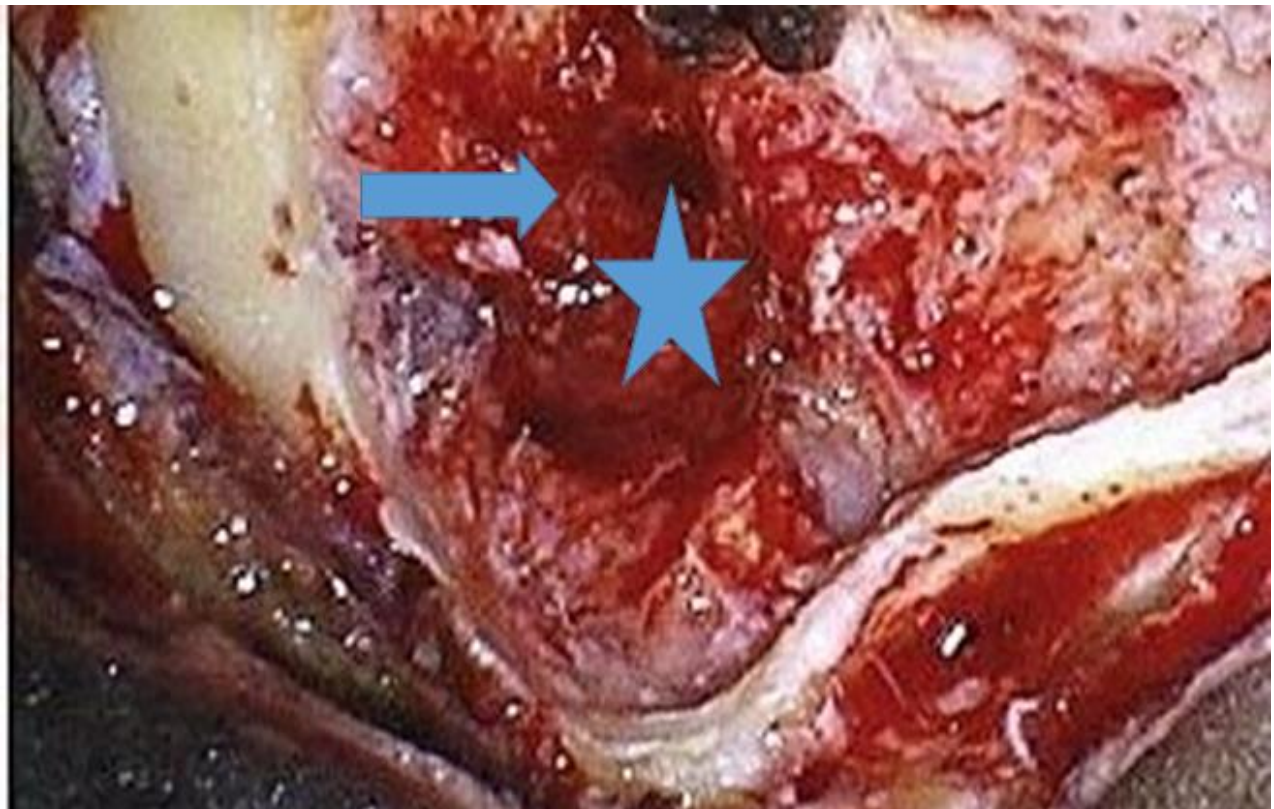


The whole mastoid cavity was finally filled up and obliterated with soft tissues taken from postauricular skin flap. Cochlear implantation was effectively completed.

There were no postoperative complications. The patient was discharged from the hospital 14 days after the surgery and follow-ups were normal. Finally, the audiological outcome was good.



**Figure 1:** Preoperative CT-Scan shows no pathological findings.



**Figure 2:** The intraoperative findings of the patient. As it can be seen, we had already contacted a sufficient mastoidectomy. The asterisk and the arrow show where the encephaloceles were found.



## Discussion

We presented a case of a young woman with the chief complaint of repeated hearing loss episodes. The decision to conduct a cochlear implantation was based both on her personal demands (professional, age, quality of life), and findings of hearing tests.

Encephalocele formation through the temporal bone has multiple etiologies and is broadly categorized as congenital or acquired. Congenital defects are less common than acquired ones and often are developed due to improper ossification of the petrosquamous junction or labyrinthine malformation. The occurrence of temporal meningoceles and/or meningoencephaloceles in the middle ear or mastoid is often insidious in onset and usually only occurs with ipsilateral ear fullness or CSF leak from a tympanic membrane perforation or through the eustachian tube(4,5).

Acquired nontraumatic etiologies of temporal bone encephaloceles include spontaneous perforation, chronic middle ear disease with or without cholesteatoma, and neoplasms. Traumatic etiologies are the most common acquired source and include temporal bone trauma, post-radiotherapy dehiscence, and iatrogenic causes(6-8).

Lateral temporal lobe encephalocele (LTE) is a rare entity, and it is a form of basal encephalocele. It has multiple etiologies and is classified into congenital and acquired(9-11). Due to its nonspecific presentation, imaging is crucial, and both CT and MRI are complementary. MRI is indicative of an interruption of dural line, and images appear as ‘‘tear-drop’’. MRI is also specific in differentiate brain tissues from cholesteatoma, cholesterol granuloma, or granulation tissues(11-13).

Diagnosis of temporal bone meningoencephaloceles may be challenging. Symptoms may include conductive hearing loss, otorrhea, and recurrent meningitis(10,11). Other less common presenting symptoms are facial nerve weakness, seizures(1,2,6,7) or pulsatile tinnitus(13). It is quite common for these patients to be misdiagnosed and treated as from chronic otitis media(10,11). There are no available guidelines for the management of temporal bone defects; most surgeons rely on their personal experience for the diagnosis and surgical closure of such defects. A variety of surgical techniques have been described in the literature(14,15).

Numerous surgical approaches involving grafting the bony defect have been described and found to be successful for managing meningoencephaloceles.

There have been proposed different surgical approaches. The most common are the transmastoid approach with subtotal petrosectomy and middle-ear obliteration, and the combined middle cranial fossa and transmastoid approach(16).

By the first approach, a post-aural incision is usually made 3–4 cm behind the retroauricular groove and mastoidectomy is conducted, to exenterate the air cells, remove any diseased mucosa and cholesteatoma if present, and expose the herniated brain tissue widely.

In most cases, the size of the dural deficit is smaller than the bony tegmen deficit(17,18). Fascia can be harvested and placed as underlay after lifting the dura from the surrounding bony edges.

By the transmastoid approach, mastoidectomy is conducted using a standard postaural approach. The meningoencephalic herniation can be identified and resected. By the combined middle cranial fossa and transmastoid approach, the meningoencephalic herniation can be initially managed through a transmastoid route(19,20). The post-aural incision was then extended superiorly as a reverse question mark and a posteriorly based skin flap can be elevated.

## Conclusions

A meningoencephalocele in the temporal bone may cause hearing loss. A CT scan or even MRI in some cases cannot detect very small meningoceles. A surgical exploration in such cases is indicated.

There are several surgical treatment approaches. The ideal one includes exposure and visualization of the encephalocele and its surgical resection. This approach should combine with the repair of the associated osseous or dural deficit.

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## References

1. Al-Nashar IS, Carrau RL, Herrera A, Snyderman CH: Endoscopic transnasal transpterygopalatine fossa approach to the lateral recess of the sphenoid sinus. *Laryngoscope* 2004;114:528–32.
2. Goldwing-Wood DG, Williams HO, Brookes GB. Tegmental dehiscence and brain herniation into the middle ear cleft. *J Laryngol Otol*. 1991;105:477-80.
3. Ting KN, Liew YT, Prepageran N. Lateral temporal lobe meningoencephalocele: Transmastoid repair with surgical and temporalis graft. *Indian J Otol*. 2019;25:219-22.
4. Papanikolaou V, Bibas A, Ferekidis E, Angnostopoulou A, Xenelis J. Idiopathic Temporal Bone Encephalocele. *Skull Base*. 2017;17:311-16.
5. Kutz JW Jr, Husain IA, Isaacson B, Roland PS. Management of spontaneous cerebrospinal fluid otorrhea. *Laryngoscope*. 2008;118:2195–9.
6. Nahas Z, Tatlipinar A, Limb CJ, Francis HW. Spontaneous meningoencephalocele of the temporal bone: clinical spectrum and presentation. *Arch Otolaryngol Head Neck Surg*. 2008;134:509–18
7. Jackson CG, Pappas DG Jr, Manolidis S, et al. Brain herniation into the middle ear and mastoid: concepts in diagnosis and surgical management. *Am J Otol*. 1997;18:198-205.
8. Mosnier I, Fiky LE, Shahidi A, Sterkers O. Brain herniation and chronic otitis media: diagnosis and surgical management. *Clin Otolaryngol Allied Sci*. 2000;25: 385-91.

9. Brown NE, Grundfast KM, Jabre A, Megerian CA, O'Malley BW Jr., Rosenberg SI. Diagnosis and management of spontaneous cerebrospinal fluid-middle ear effusion and Otorrhea. *Laryngoscope*. 2004;114: 800-5.
10. Glasscock Jr MD, Jackson GC, Wiet JR. Surgical management of brain tissue into the middle ear and mastoid. *Laryngoscope* 1979;89:1743-54
11. Iurato S, Ettore GC, Selvini C. Brain herniation into the middle ear: two idiopathic cases treated by a combined intracranial mastoid approach. *Laryngoscope* 1989;99:950-4
12. Jeevan DS, Ormond DR, Kim AH et al., Cerebrospinal fluid leaks and encephaloceles of temporal bone origin: nuances to diagnosis and management. *World Neurosurg*. 2015;83:560-6.
13. Sanna M, Paolo F, Russo A, Falcioni M. Management of meningoencephalic herniation of the temporal bone: personal experience and literature review. *Laryngoscope*. 2009;119:1579-85.
14. Sanna M, Fois P, Russo A, Falcioni M. Management of meningoencephalic herniation of the temporal bone: personal experience and literature review. *Laryngoscope* 2009;119:1579-85.
15. Jackson CG, Pappas DG Jr, Manolidis S et al. Brain herniation into the middle ear and mastoid: concepts in diagnosis and surgical management. *Am J Otol*. 1997;18:198-205
16. Kantas I, Tzindros G, Papadopoulou A, Marangos N: Midfacial degloving: the best alternative for treatment of transsphenoidal meningocele of the pterygopalatine fossa. *Skull Base*. 2006;16:117-22.
17. Lalwani A K. In: Jackler RK, Brackman DE, editor. *Neurotology*. St. Louis, MO: Mosby; 2004. Temporal bone encephalocele. pp. 1089-1095
18. Gubbels SP, Selden NR, Delashaw JB Jr, McMenomey SO: Spontaneous middle fossa encephalocele and cerebrospinal fluid leakage: diagnosis and management. *Otol Neurotol*. 2007;28:1131-9
19. Kale SU, Pfeiderer AG, Cradwick JC. Bilateral defects of the tegmen tympani associated with brain and dural prolapse in a patient with pulsatile tinnitus. *J Laryngol Otol*. 2000;114:861-3.
20. Kenning TJ, Willcox TO, Artz GJ, Sciffmacher P, Farrell CJ, Evans JJ: Surgical management of temporal meningoencephaloceles, cerebrospinal fluid leaks, and intracranial hypertension: treatment paradigm and outcomes. *Neurosurg Focus*. 2012;32:6

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