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LETTER TO THE EDITOR / PISMO UREDNIKU

The International Day of Persons with Disabilities (IDPD) has been celebrated on December 3 since 1992. According to data from the World Health Organization (WHO), people with disabilities make up 16% of the world's population. They often face obstacles that reduce their ability to get educated and take leadership positions in the health sector. This year's IDPD theme is "Amplifying the leadership of persons with disabilities for an inclusive and sustainable future". The WHO has recognized that strengthening the leadership of persons with disabilities is essential both for advancing the principles of equality for all and achieving global health goals.

Međunarodni dan osoba sa invaliditetom (MDOI) obeležava se 3. decembra od 1992. godine. Prema podacima Svetske zdravstvene organizacije (SZO), osobe sa invaliditetom čine 16% svetske populacije. Oni se često suočavaju sa preprekama koje im smanjuju mogućnost obrazovanja i zauzimanja vodećih pozicija u zdravstvenom sektoru. Tema ovogodišnjeg MDOI je „Jačanje liderstva osoba sa invaliditetom za inkluzivnu i održivu budućnost". SZO je prepoznala da je jačanje liderstva osoba sa invaliditetom od suštinskog značaja kako za unapređenje principa jednakosti za sve tako i za postizanje globalnih zdravstvenih ciljeva.

Dear authors, editors, peer reviewers, and readers of the *Vojnosanitetski pregled*,
I would like to thank you for your cooperation and support in the previous year and wish you all the best in the coming 2025!

Merry Christmas and Happy New Year!

Editorial staff of the *Vojnosanitetski pregled*

Poštovani autori, urednici, recenzenti i čitaoci časopisa *Vojnosanitetski pregled*,
uz zahvalnost na saradnji i podršci u protekloj godini,
želim vam sve najbolje u nastupajućoj 2025. godini!

Srećna Nova godina i božićni praznici!

Redakcija *Vojnosanitetskog pregleda*

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Characteristics of human immunodeficiency virus-1 influencing the development and efficacy of anti-HIV-1 vaccines

Karakteristike virusa humane imunodeficijencije-1 koje utiču na razvoj i efikasnost anti-HIV-1 vakcina

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Key words:
disease transmission, infectious; geography, medical; hiv; vaccines; treatment outcome.

Ključne reči:
infekcija, putevi širenja; geografija, medicinska; hiv; vakcine; lečenje, ishod.

Introduction

The human immunodeficiency virus (HIV) belongs to the family *Retroviridae*, subfamily *Orthoretrovirinae*, genus *Lentivirus*, characterized by a long incubation period of several months to several years and the outbreak of diseases with a chronic course and fatal outcome. HIV-1 and HIV-2 viruses, derived from primate lentiviruses, have a tropism for the cells of the immune system, leading to a depletion of $CD4^+$ T lymphocytes and a lack of cellular immune response, and the final stage of HIV infection is characterized by the development of acquired immunodeficiency syndrome (AIDS). Since the beginning of the AIDS pandemic, 88.4 million people have been infected with HIV, and 42.3 million have died from AIDS-related diseases. According to the latest data, in 2023, 39.9 million people worldwide were living with HIV infection (38.6 million adults and 1.4 million children), 1.3 million were newly infected, and around 630,000 people died from AIDS-related diseases. Of those infected, 53% were young women and girls [1]. HIV is currently one of the most serious public health problems, and understanding the mechanisms of replication and spread of HIV infection is crucial for the development of new drugs and vaccines to combat this disease. The antiretroviral drugs (AD) that have come to market over the last forty years have significantly improved the quality of life and life expectancy of HIV-positive individuals. Due to the great genetic and antigenic variability of the virus, no effective vaccine has yet been developed that could significantly reduce the incidence of HIV infections in risk groups.

HIV-1 is a ribonucleic acid (RNA) virus with a protein capsid of atypical symmetry and a lipid bilayer envelope. The capsid consists of the small basic nucleoproteins p7, p9, and the protein p24. Inside the core are viral enzymes reverse transcriptase (RT), integrase, and protease, which are involved in the synthesis of proviral deoxyribonucleic acid (DNA) and its integration into the host genome, as well as in the maturation of viral particles. The matrix protein p17 is located between the capsid and the lipid envelope, while the transmembrane glycoprotein gp41 and the surface glycoprotein gp120 are integrated into the envelope and contribute to the adsorption and penetration of HIV-1 into the target cell. The virus has a diploid RNA genome consisting of two identical molecules of linear single-stranded $^{(+)}$RNA. The genome of HIV-1 contains three genes found in all retroviruses – *GAG*, *POL*, and *ENV*, which code for the structural and functional proteins of HIV-1, and six regulatory genes – *TAT*, *REV*, *VIF*, *NEF*, *VPR*, and *VPU*, whose products regulate viral replication and are responsible for evading the host immune response [2].

Variability of HIV-1 and its impact on vaccine development

Groups and subtypes of HIV

HIV-1 is responsible for the global AIDS epidemic and infects 95% of HIV-infected individuals, while HIV-2 is less prevalent and less virulent [3–5] but causes similar clinical

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symptoms to HIV-1 [6]. HIV-2 is most widespread in West Africa, while only a few people are infected in Europe, India, and the United States of America. Compared to HIV-1, it is characterized by a longer asymptomatic phase and a slower course of the disease. There are four different groups within the HIV-1 type: M (main), O (outlier), N (non-M, non-O), and P (pending) [7–11]. These groups are genetically related but have a different geographical distribution, and all four lead to similar clinical symptoms of HIV infection [12].

Group M is the most widespread and accounts for approximately 90% of all HIV-1 infections. It emerged in the 1920s in Kinshasa, Democratic Republic of Congo, and its zoonotic origin is the simian immunodeficiency virus (SIV) from wild chimpanzees (SIV_{cpz}), which infects chimpanzees. There are nine subtypes within this group, designated by the letters A to J (A1, A2, A3, A4, A6, B, C, D, F1, F2, G, H, J, and K) [13–18]. The geographic distribution of group M circulating subtypes is presented in Figure 1.

Group N was originally identified in Central Africa but is relatively rare and causes only a few infections worldwide [17]. Like group M, it is derived from the SIV_{cpz} [16].

Group O causes infections in West and Central Africa and is rarely found outside this region [17]. In contrast to groups M and N, group O is derived from the SIVs infecting the western lowland gorillas (SIV_{gor}), which infects gorillas. For this reason, detection of this group had been difficult with the original HIV-1 diagnostic kits until newer generation tests were developed [19].

Group P was first isolated in Cameroon in 2009 and is the least widespread of all groups. It has a high degree of genetic similarity with SIV_{gor}.

Circulating recombinant forms and unique recombinant forms

Infection of an individual with two or more HIV-1 subtypes can lead to their recombination and the formation of a unique recombinant form (URF) of the virus. This URF can be detected in an infected person by sequencing the viral genome and has no major epidemiologic significance. When the URF is detected in three or more geographically distant individuals who are epidemiologically unrelated, this recombinant genome is referred to as the circulating recombinant form (CRF). It has epidemiologic significance for the spread of the M-group HIV-1 epidemic [18]. The CRFs are designated by numbers and are numbered in the order of their discovery. Approximately 150 different CRFs and several URFs have been identified [20]. The HIV-1 subtypes E and I no longer exist today as independent entities, as recombination of these subtypes with other subtypes has taken place over the years, and CRF01_AE (recombination of subtypes A and E) and CRF04_cpx (complex recombination

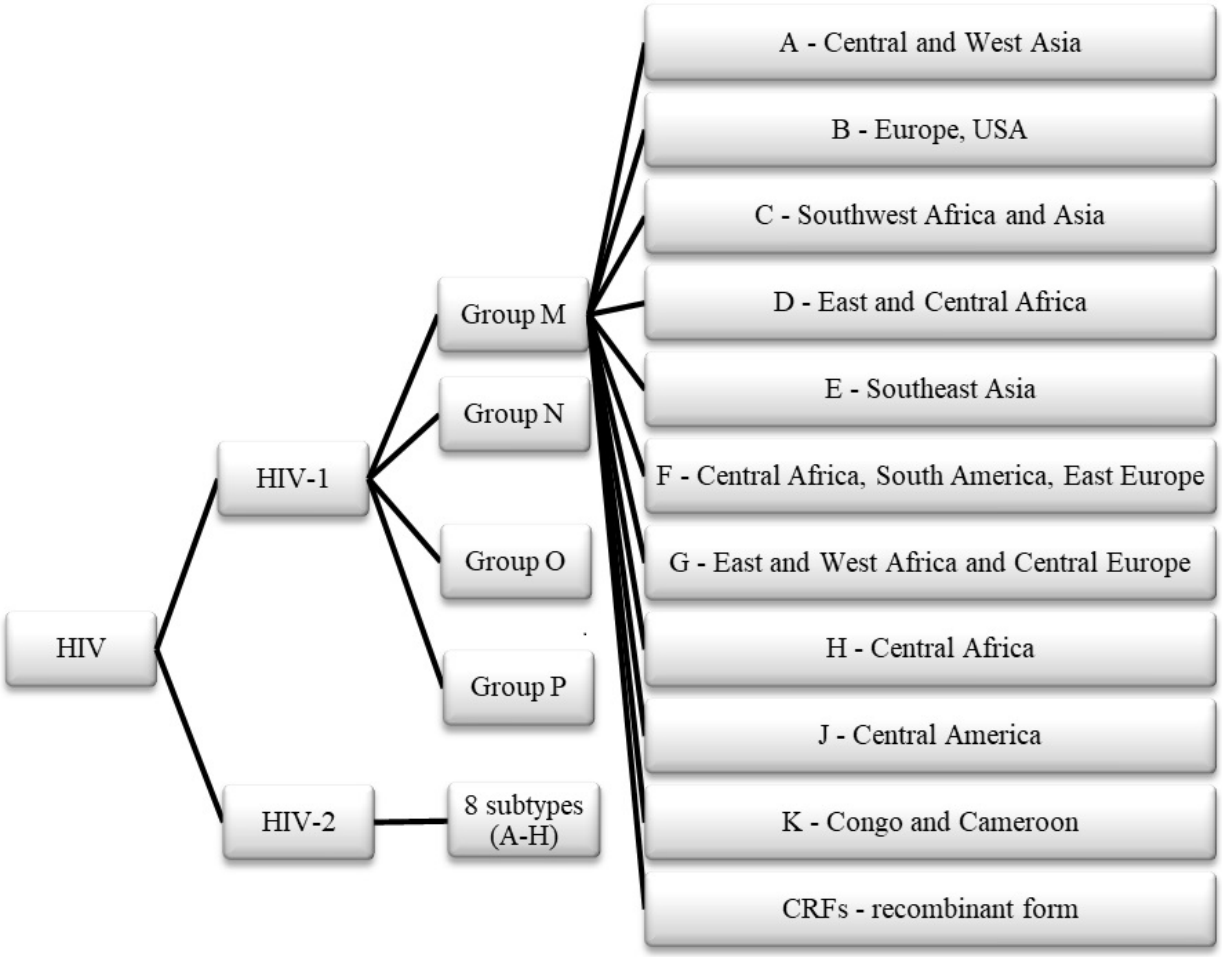

Fig. 1 – Geographic distribution of human immunodeficiency virus (HIV)-1 circulating subtypes.

of several subtypes) have taken subtypes E and I out of circulation. Most of the HIV-1 infections are caused by subtype C (46.6%), followed by subtype B (12.1%), subtype A (10.3%), CRF02_AG (7.7%), and CRF01_AE (5.3%). CRF01_AE is most widespread in Asia, and CRF02_AG in West Africa [11].

The geographical distribution of groups and subgroups of HIV-1, as well as CRFs, can change over time, and population migration and travel are the most important factors for the spread of different types of HIV-1 to geographically distant locations. In addition, most HIV-1 infections worldwide belong to group M and its subgroups, and different subgroups may predominate in various regions of the world. The Balkan Peninsula is characterized by the occurrence of different subtypes of HIV-1 of group M. Subtype B is predominant in Serbia, Slovenia, and Hungary, while subtype A is predominant in Albania, and subtype F in Romania [21, 22]. Determining viral subtypes is important for predicting therapeutic success, as certain HIV-1 subtypes may be resistant to some antiretroviral drugs. It is also of particular importance for the development of vaccines against HIV-1. An effective vaccine must protect against different types/subtypes of the HIV-1 virus and their recombinant forms. In addition, the diversity of HIV-1 subtypes also affects the accuracy of HIV diagnostic tests and viral load tests. The tests currently in use are able to identify and monitor all subtypes and recombinant forms that have been identified so far. However, it is expected that current recombination will lead to the emergence of new URFs and CRFs for which the tests have not yet been designed [19].

Genetic and antigenic variability of HIV-1

HIV-1 is characterized by a high rate of viral mutations that cause extreme genetic diversity of the virus. As a consequence, the virus adapts to the effector mechanisms of the immune response and evades it efficiently. It also develops resistance to antiretroviral therapy. However, some mutations are not beneficial for the virus, as they impede its survival and ability to cause the infection of the host (e.g., "fitness" of the virus) [23, 24]. The genetic diversity of HIV-1 is a consequence of a high rate of viral replication, the non-repairing function of the enzyme RT, and the recombinations that happen during the replication of the virus [25, 26].

High replication rate of HIV-1 and the reverse transcriptase enzyme

HIV-1 RT is a multifunctional enzyme that possesses both RNA-dependent DNA polymerase and DNA-dependent DNA polymerase activity as well as ribonuclease H activity, which specifically degrades the RNA strand of the resulting RNA/DNA hybrid. In contrast to other DNA polymerases, the HIV-1 RT does not have the function of correcting errors that occur during replication. The rate of nucleotide substitution introduced by the RT is approximately 10^{-4} *per* nucleotide *per* replication cycle, resulting in one nucleotide substitution *per* genome during a replication cycle [27]. The HIV-1 replicates daily at a high rate; it is estimated that an infected person produces about 10^9 virions *per* day. In contrast, the life span of the virus in plasma and virus-infected cells is quite short, with a half-life of about two days. Therefore, the wild-type strains that primarily infected humans are completely replaced by genetically similar variants of the same viruses (e.g., quasispecies) within two to four weeks [28]. This property of RT, together with the high replication rate of the virus *in vivo*, contributes to the continuous emergence of new viral variants [13, 29-31]. During viral replication, insertions, deletions, and duplications also occur, which also contribute to the genetic heterogeneity of HIV-1 [28].

Genetic recombination of HIV-1

Genetic recombination of HIV-1 occurs when two or more genetically distant viruses recombine during dual or multiple infections of a person, resulting in a new form of the virus with the original sequence (CRF or URF) [32]. Recombination increases the overall genetic complexity and diversity of the viral population, leading to faster viral adaptation, the emergence of resistance to AD, including multidrug resistance, evasion of the immune response, and disease progression [33-38].

Emergence of HIV-1 quasispecies

Quasispecies are a group of mutant viruses that develop during viral replication in an HIV-positive person. The HIV-1 can mutate into multiple quasispecies during infection, which reduces the ability of the immune response that has developed against the primary wild strain of the virus to control the infection and also leads to the emergence of HIV viral variants that are resistant to AD [39]. One of the main goals of early initiation of antiretroviral therapy during HIV infection is to reduce the rate of viral replication, thereby reducing the possibility of the emergence of quasispecies. For this reason, in several clinical trials investigating HIV vaccines, pre-exposure prophylaxis has been administered along with the vaccine to increase vaccine efficacy [40].

Antigenic epitopes of HIV-1 important for vaccine development

Antigenic epitopes that are potential candidates for HIV-1 vaccine development must be evolutionarily conserved and present in the majority of HIV-1 subtypes and their variants for the vaccine to be effective in a wider geographic area.

Antigenic epitopes of HIV-1 for B lymphocytes

Epitopes for B lymphocytes can be either linear or conformational [41]. Linear epitopes consist of a continuous (consecutive) sequence of amino acids on the antigen, whereas a conformational epitope is formed after the protein has folded and connects two or more non-consecutive linear sequences (e.g., discontinuous epitope). The epitopes of HIV for B-cell receptor (BCR) can be lipid, glycan and protein

antigens, or their combinations [42-45]. Numerous studies have shown that broadly neutralizing HIV-1 antibodies (bNAbs) bind more efficiently to conformational than linear epitopes [42-44, 46], which complicates their use in vaccine development, where single epitopes are used. HIV epitopes for bNAbs are thought to have an evolutionarily conserved sequence due to their broad affinity for multiple HIV-1 subtypes, making them cross-reactive [44, 47], but they may also contain highly variable segments [42]. Although these epitopes do not have the same amino acid sequence, the binding of bNAbs may result from the epitopes having a common conformation in the secondary or tertiary structure of the protein, which poses a challenge to the development of anti-HIV vaccines [48].

The presence of bNAbs was first detected in 20–50% of HIV-positive individuals who had had chronic HIV infection for more than 2–5 years [44, 49, 50]. Based on the study of bNAbs isolated from infected individuals, it was found that these antibodies mainly belong to different isotypes of the IgG class. They can bind to five different epitope regions of the HIV ENV protein: the CD4 binding site; the V2 proteoglycan motif at the top of the ENV trimer; the V3 proteoglycan motif with high mannose content; the membrane-proximal external region (MPER) of the ENV transmembrane domain; the gp120-gp41 protein junction with or without fusion protein [42-44]. Ideal vaccine antigens would be HIV-1 epitopes that induce bNAbs that bind to one or more of the above sites on the Env protein and induce the production of IgG. However, these have not been identified or developed to date [43, 44, 51]. An overview of BCR epitopes for bNAbs and their characteristics is given in Table 1.

Several studies have shown that bNAbs isolated from HIV-positive individuals with chronic infection share common features of paratopes [antigenic epitope binding sites in the complementarity-determining region (CDR)1, CDR2, and CDR3 of the immunoglobulin light and heavy chains], such as a high degree of somatic hypermutation in the genes of the V(D)J region, the presence of a long heavy chain in the CDR3, and polyreactivity or autoreactivity with self-antigens (proteins, glycans, and lipids), which can lead to autoimmune diseases [42-44, 52, 53].

In addition to the antigenic epitopes that lead to the formation of bNAbs, the HIV-1 also possesses epitopes that do not lead to the formation of neutralizing antibodies (i.e., eliciting non-neutralizing antibodies – nNAbs) but can lead to antibody-dependent cellular cytotoxicity (ADCC) or antibody-dependent cellular viral inhibition (ADCVI) [54-57]. ADCC and ADCVI are mediated by the Fcγ receptor on the surface of effector cells (NK cells, macrophages, dendritic cells, or neutrophils) that produce the effector molecules perforin and granzyme, resulting in cytolysis (ADCC) or β-chemokines to inhibit the virus-infected cell (ADCVI). In addition to the beneficial functions of nNAbs, they can interfere with the functions of bNAbs by competing for the same antigenic epitopes, thereby reducing the function of bNAbs. They can also lead to increased infection of cells with HIV, as they bind to Fcγ receptors on the surface of macrophages and dendritic cells, which can promote the entry of HIV into these cells [58].

Antigenic epitopes of HIV-1 for T lymphocytes

Epitopes for T lymphocytes are short linear peptides generated by the processing of protein antigens and displayed by the major histocompatibility complex (MHC) molecules. Evolutionarily conserved epitopes of HIV-1 for T lymphocytes have an identical amino acid sequence in HIV-1 isolates of the same subtype (i.e., type-specific epitopes) or for several different HIV-1 subtypes [59-61]. They are also evolutionarily conserved in other human lentiviruses, primate [primate immunodeficiency lentiviruse (PIV)] and cat lentiviruses [HIV, SIV, feline immunodeficiency virus

Table 1

Characteristics of HIV epitopes for the B-cell receptor that induce broadly neutralizing antibodies (bNAbs)

Structure of the epitope	Epitope regions on the HIV ENV protein	bNAbs	Antibody isotype
Conformational	CD4 binding site	VRCO1, CH103, b12	IgG1
		3BNC117	IgG1κ
		PGV04, 8ANC13, CH235	IgG
	V2 proteoglycan	PG9, CHO1	IgG1
		PGT145, VRC2609	IgG
	V3 proteoglycan	PGT121, PGT128	IgG1
		PGT135	IgG
	gp120-gp41	PGT151, VRC34.01, 35022, 8ANC195	IgG
Linear	MPER	10E8, 2F5	IgG3
		4E10	IgG3κ

HIV – human immunodeficiency virus; MPER – membrane-proximal external region; the antibodies VRCO1, 3BNC117, b12, PG9, PGT145, PGT121, 35022, 10E8, 4E10, and 2F5 bind to the Fcγ receptor on the surface of effector cells and induce antibody-dependent cellular cytotoxicity.

Table 2

Example of HIV-1 epitopes for T lymphocytes

Epitope	Protein	HXB2 location	Subprotein	HXB2 DNA	Subtype	Species	HLA
WEKIRLRP	GAG	15-23	p17(15-23)	832..858	B	human	A*02:05
EDEGKISKI	POL	197-205	RT(42-50)	2673..2699	B	human	B*51:01
VWKDAETTL	ENV	44-52	gp120(44-52)	6354..6380	B	human	B*38:01

HIV – human immunodeficiency virus; RT – reverse transcriptase; HLA – human leukocyte antigens.

(FIV)] [62, 63]. Mutations in epitopes for T lymphocytes occur regularly, both in conserved and non-conserved epitopes with variable sequences, leading to differential retention of variants that occur after mutations. It is hypothesized that highly conserved epitopes occur in protein regions essential for viral survival and that any significant mutation would compromise viral viability [64-66]. Therefore, variants that occur after these mutations are eliminated by the mechanisms of natural selection.

To date, a large number of HIV-1 epitopes have been discovered for T lymphocytes that are candidates for vaccine development. HIV epitopes can vary in length, but the variant with the shortest epitope length is thought to elicit the strongest immune response, so this epitope variant is considered "optimal". HIV-1 epitopes shorter than 21 amino acids are included in a list of optimal epitopes, the so-called A-list of HIV epitopes [20]. Each HIV-1 epitope has a unique identification number, the position of the defined epitope site in relation to the HXB2 protein sequence (viral reference genome HXB2, GenBank code K03455), the protein on which it is located and its subunit, the virus subtype, the epitope sequence, and the host and the human leukocyte antigens (HLA) restriction epitope element [20]. An example of an epitope for T lymphocytes can be found in Table 2.

Evolutionarily conserved epitopes of HIV-1 for T lymphocytes must be able to induce activation of $CD8^+$ cytotoxic T lymphocytes (CTLs) [44, 67], $CD8^+$ and $CD4^+$ T lymphocytes [68, 69], and possibly activation of a subset of cytotoxic $CD4^+$ CTL [70]. All of the above activities of T lymphocytes are important for the development of prophylactic vaccines.

HIV-1 epitopes for T lymphocytes are subdivided into epitopes for CTL/$CD8^+$ and epitopes for T helper lymphocytes (T helper/$CD4^+$). To date, 2,067 epitopes for CTL and 725 epitopes for $CD4^+$ T lymphocytes have been identified, which are mainly located on the GAG, POL, and ENV proteins. The largest number of identified epitopes belongs to HIV-1 virus subtype B. A list of HIV-1 epitopes for CTL/$CD8^+$ and $CD4^+$ T lymphocytes is available in the Los Alamos HIV database [20]. According to the information in this database, 569 epitope sequences belong to subtypes B (537), C (9), and G (7), and 16 additional subtypes (01_AE: 4; 01B: 2; 02_AG: 4; A: 2; A1: 3; F1: 1) were identified in Serbia (date of access July 1, 2024).

HIV-1 antigen epitopes for T lymphocytes bind to MHC class I and MHC class II molecules coded by HLA genes and their alleles. Various HLA allotypes can be associated with susceptibility or resistance to HIV infection [71-74]. Certain alleles for HIV resistance/susceptibility also differ in correlation with ethnicity or the endemic prevalence of HIV subtypes that circulate in the country. For instance, the resistance of European and North American Caucasians to HIV-1 subtype B and African populations from Kenya, Tanzania, and sub-Saharan Africa to subtypes A, C, and D are associated with HLA-A2 alleles such as HLA-A2, HLA-A*0205, and HLA-A*6802 [71-73, 75, 76]. In contrast, susceptibility to HIV-1 is associated with HLA-B7 alleles: susceptibility to subtype B in Europe and North America with HLA-B*3501, HLA-B*3502, and HLA-B*5303 alleles [75], and to subtypes A, C, and D in Kenya with HLA-B*0702 and HLA-B*4201 alleles [76].

Conclusion

The existence of a large number of groups and subgroups of HIV-1 with a high degree of mutation and recombination leading to the emergence of circulating and unique recombinant forms and quasispecies of HIV-1 makes it difficult to develop a single vaccine against HIV-1 that would be effective against all strains of the virus in all geographic areas.

Given the effector functions of HIV-1 antibodies (bNAbs and nNAbs) and the risk of interference of these antibodies with other functions in the body, antigenic epitopes that lead to the production of bNAbs and/or nNAbs must be carefully selected when choosing a potential vaccine antigen candidate. In the development of vaccines containing antigenic epitopes of HIV-1 for T lymphocytes, the distribution of HLA allotypes in a given endemic area should be considered in addition to the selection of the appropriate epitope to induce a protective immune response.

REFERENCES

1. *Joint United Nations Programme on HIV/AIDS (UNAIDS).* Global HIV & AIDS Statistics - 2023 Fact Sheet [Internet]. Switzerland: UNAIDS 2023; [cited 2024 July 4; accessed on 2024 Sept 2]. Available from: https://www.unaids.org/en/resources/fact-sheet
2. *Masenga SK, Mweene BC, Luwaya E, Muchaili L, Chona M, Kirabo A.* HIV-Host Cell Interactions. Cells 2023; 12(10): 1351.
3. *Clavel F, Guétard D, Brun-Vézinet F, Chamaret S, Rey MA, Santos-Ferreira MO*, et al. Isolation of a new human retrovirus from West African patients with AIDS. Science 1986; 233(4761): 343–6.
4. *Ariyoshi K, Schim van der Loeff M, Berry N, Jaffar S, Whittle H.* Plasma HIV viral load in relation to season and to Plasmodium falciparum parasitaemia. AIDS 1999; 13(9): 1145–6.

5. *Ariyoshi K, Jaffar S, Alabi AS, Berry N, Schim van der Loeff M, Sabally S*, et al. Plasma RNA viral load predicts the rate of CD4 T cell decline and death in HIV-2-infected patients in West Africa. AIDS 2000; 14(4): 339–44.
6. *Wilkins A, Ricard D, Todd J, Whittle H, Dias F, Paulo Da Silva A.* The epidemiology of HIV infection in a rural area of Guinea-Bissau. AIDS 1993; 7(8): 1119–22.
7. *Burke DS*. Recombination in HIV: an important viral evolutionary strategy. Emerg Infect Dis 1997; 3(3): 253–9.
8. *Plantier JC, Leoz M, Dickerson JE, De Oliveira F, Cordonnier F, Lemée V*, et al. A new human immunodeficiency virus derived from gorillas. Nat Med 2009; 15(8): 871–2.
9. *Robertson DL, Anderson JP, Bradac JA, Carr JK, Foley B, Funkhouser RK*, et al. HIV-1 nomenclature proposal. Science 2000; 288(5463): 55–7.
10. *Vallari A, Holzmayer V, Harris B, Yamaguchi J, Ngansop C, Makamche F,* et al. Confirmation of putative HIV-1 group P in Cameroon. J Virol 2011; 85(3): 1403–7.
11. *Giovanetti M, Ciccozzi M, Parolin C, Borsetti A.* Molecular Epidemiology of HIV-1 in African Countries: A Comprehensive Overview. Pathogens 2020; 9(12): 1072.
12. *Williams A, Menon S, Crowe M, Agarwal N, Biccler J, Bbosa N,* et al. Geographic and population distributions of Human Immunodeficiency Virus (HIV)-1 and HIV-2 circulating subtypes: a systematic literature review and meta-analysis (2010-2021). J Infect Dis 2023: 228(11): 1583−91.
13. *Perrin L, Kaiser L, Yerly S*. Travel and the spread of HIV-1 genetic variants. Lancet Infect Dis 2003; 3(1): 22–7.
14. *Lessells RJ, Katzenstein DK, De Oliveira T*. Are subtype differences important in HIV drug resistance? Curr Opin Virol 2012; 2(5): 636–43.
15. *Wainberg MA, Brenner BG*. The impact of HIV genetic polymorphisms and subtype differences on the occurrence of resistance to antiretroviral drugs. Mol Biol Int 2012; 2012: 256982.
16. *Goudsmit J*. Viral Sex; The Nature of AIDS. New York (NY): Oxford University Press. 1997. p. 260.
17. *Giovanetti M, Ciccozzi M, Parolin C, Borsetti A*. Molecular Epidemiology of HIV-1 in African Countries: A Comprehensive Overview. Pathogens 2020; 9(12): 1072.
18. *He L, Dong R, He RL, Yau SS*. A novel alignment-free method for HIV-1 subtype classification. Infect Genet Evol 2020; 77: 104080.
19. *Alexander TS*. Human Immunodeficiency Virus Diagnostic Testing: 30 Years of Evolution. Clin Vaccine Immunol 2016; 23(4): 249–53.
20. *Apetrei C, Hahn B, Rambaut A, Wolinsky S, Brister JR, Keele B,* et al. HIV Sequence Compendium 2021. Published by Theoretical Biology and Biophysics Group, Los Alamos National Laboratory, NM, LA-UR-23-22840 [Internet]. 2021 [cited 2024 June 20; accessed on 2024 Sept 3]. Available from: https://www.hiv.lanl.gov and https://www.hiv.lanl.gov/content/immunology/
21. *Stanojevic M, Alexiev I, Beshkov D, Gökengin D, Mezei M, Minarovits J,* et al. HIV-1 molecular epidemiology in the Balkans: a melting pot for high genetic diversity. AIDS Rev 2012; 14(1): 28−36.
22. *Siljic M, Salemovic D, Jevtovic D, Pesic-Pavlovic I, Zerjav S, Nikolic V,* et al. Molecular typing of the local HIV-1 epidemic in Serbia. Infect Genet Evol 2013; 19: 378−85.
23. *Ariën KK, Abraha A, Quiñones-Mateu ME, Kestens L, Vanham G, Arts EJ*. The replicative fitness of primary human immunodeficiency virus type 1 (HIV-1) group M, HIV-1 group O, and HIV-2 isolates. J Virol 2005; 79(14): 8979–90.
24. *Troyer RM, McNevin J, Liu Y, Zhang SC, Krizan RW, Abraha A,* et al. Variable fitness impact of HIV-1 escape mutations to cytotoxic T lymphocyte (CTL) response. PLoS Pathog 2009; 5(4): e1000365.
25. *Zhuang J, Jetzt AE, Sun G, Yu H, Klarmann G, Ron Y,* et al. Human immunodeficiency virus type 1 recombination: rate, fidelity, and putative hot spots. J Virol 2002; 76(22): 11273–82.
26. *Ramirez BC, Simon-Loriere E, Galetto R, Negroni M*. Implications of recombination for HIV diversity. Virus Res 2008; 134(1-2): 64–73.
27. *Nowak M*. HIV mutation rate. Nature 1990; 347(6293): 522.
28. *Hu WS, Temin HM*. Retroviral recombination and reverse transcription. Science 1990; 250(4985): 1227–33.
29. *Mamrosh JL, Sharon E, Fung D, Korber TMB, Brander C, Barouch D,* et al. HIV Molecular Immunology 2022 [Internet]. New Mexico: Los Alamos National Laboratory, Theoretical Biology and Biophysics; 2023 [accessed on 2024 Sept 6]. Available from: https://www.hiv.lanl.gov/content/immunology/howto-cite.html
30. *Coffin JM*. HIV population dynamics in vivo: implications for genetic variation, pathogenesis, and therapy. Science 1995; 267(5197): 483–9.
31. *Wei X, Ghosh SK, Taylor ME, Johnson VA, Emini EA, Deutsch P,* et al. Viral dynamics in human immunodeficiency virus type 1 infection. Nature 1995; 373(6510): 117–22.
32. *Song H, Giorgi EE, Ganusov VV, Cai F, Athreya G, Yoon H,* et al. Tracking HIV-1 recombination to resolve its contribution to HIV-1 evolution in natural infection. Nat Commun 2018; 9(1): 1928.
33. *Nishimura Y, Shingai M, Lee WR, Sadjadpour R, Donau OK, Willey R,* et al. Recombination-mediated changes in coreceptor usage confer an augmented pathogenic phenotype in a nonhuman primate model of HIV-1-induced AIDS. J Virol 2011; 85(20): 10617–26.
34. *Nora T, Charpentier C, Tenaillon O, Hoede C, Clavel F, Hance AJ*. Contribution of recombination to the evolution of human immunodeficiency viruses expressing resistance to antiretroviral treatment. J Virol 2007; 81(14): 7620–8.
35. *Moutouh L, Corbeil J, Richman DD*. Recombination leads to the rapid emergence of HIV-1 dually resistant mutants under selective drug pressure. Proc Natl Acad Sci USA 1996; 93(12): 6106–11.
36. *Ritchie AJ, Cai F, Smith NM, Chen S, Song H, Brackenridge S,* et al. Recombination-mediated escape from primary CD8+ T cells in acute HIV-1 infection. Retrovirology 2014; 11: 69.
37. *Streeck H, Li B, Poon AF, Schneidewind A, Gladden AD, Power KA,* et al. Immune-driven recombination and loss of control after HIV superinfection. J Exp Med 2008; 205(8): 1789−96.
38. *Liu SL, Mittler JE, Nickle DC, Mulvania TM, Shriner D, Rodrigo AG,* et al. Selection for human immunodeficiency virus type 1 recombinants in a patient with rapid progression to AIDS. J Virol 2002; 76(21): 10674−84.
39. *Smyth RP, Davenport MP, Mak J*. The origin of genetic diversity in HIV-1. Virus Res 2012; 169(2): 415−29.
40. *McNicholl JM*. Combining biomedical preventions for HIV: Vaccines with pre-exposure prophylaxis, microbicides or other HIV preventions. Hum Vaccin Immunother 2016; 12(12): 3202–11.
41. *Nielsen M, Marcatili P*. Prediction of Antibody Epitopes. Methods Mol Biol 2015; 1348: 23–32.
42. *Wu X, Kong XP*. Antigenic landscape of the HIV-1 envelope and new immunological concepts defined by HIV-1 broadly neutralizing antibodies. Curr Opin Immunol 2016; 42: 56–64.
43. *McCoy LE, Burton DR*. Identification and specificity of broadly neutralizing antibodies against HIV. Immunol Rev 2017; 275(1): 11–20.
44. *Korber B, Hraber P, Wagh K, Hahn BH*. Polyvalent vaccine approaches to combat HIV-1 diversity. Immunol Rev 2017; 275(1): 230–44.
45. *Cerutti N, Loredo-Varela JL, Caillat C, Weissenhorn W*. Antigp41 membrane proximal external region antibodies and the art of

using the membrane for neutralization. Curr Opin HIV AIDS 2017; 12(3): 250–6.
46. *Yoon H, Macke J, West AP Jr, Foley B, Bjorkman PJ, Korber B,* et al. CATNAP: a tool to compile, analyze and tally neutralizing antibody panels. Nucleic Acid Res 2015; 43(W1): W213–9.
47. *Burton DR, Mascola JR.* Antibody responses to envelope glycoproteins in HIV-1 infection. Nat Immunol 2015; 16(6): 571–6.
48. *Landais E, Moore PL.* Development of broadly neutralizing antibodies in HIV-1 infected elite neutralizers. Retrovirology 2018; 15(1): 61.
49. *Hraber P, Seaman MS, Bailer RT, Mascola JR, Montefiori DC, Korber BT.* Prevalence of broadly neutralizing antibody responses during chronic HIV-1 infection. AIDS 2014; 28(2): 163–9.
50. *Gray ES, Madiga MC, Hermanus T, Moore PL, Wibmer CK, Tumba NL,* et al. The neutralization breadth of HIV-1 develops incrementally over four years and is associated with CD4+ T cell decline and high viral load during acute infection. J Virol 2011; 85(10): 4828–40.
51. *Burton DR, Hangartner L.* Broadly Neutralizing Antibodies to HIV and Their Role in Vaccine Design. Annu Rev Immunol 2016; 34: 635–59.
52. *Kelsoe G, Haynes BF.* Host controls of HIV broadly neutralizing antibody development. Immunol Rev 2017; 275(1): 79–88.
53. *Wibmer CK, Moore PL, Morris L.* HIV broadly neutralizing antibody targets. Curr Opin HIV AIDS 2015; 10(3): 135–43.
54. *Boesch AW, Brown EP, Ackerman ME.* The role of Fc receptors in HIV prevention and therapy. Immunol Rev 2015; 268(1): 296–310.
55. *Pollara J, Bonsignori M, Moody MA, Pazgier M, Haynes BF, Ferrari G.* Epitope specificity of human immunodeficiency virus-1 antibody dependent cellular cytotoxicity [ADCC] responses. Curr HIV Res 2013; 11(5): 378–87.
56. *Girard MP, Picot V, Longuet C, Nabel GJ.* Report of the Cent Gardes HIV Vaccine Conference: The B-cell response to HIV. Part 2: Non-neutralizing antibodies: Fondation Mérieux Conference Center, Veyrier du Lac, France 5–7 November 2012. Vaccine 2013; 31(29): 2984–7.
57. *Forthal DN, Moog C.* Fc receptor-mediated antiviral antibodies. Curr Opin HIV AIDS 2009; 4(5): 388–93.
58. *Sahay B, Nguyen CQ, Yamamoto JK.* Conserved HIV Epitopes for an Effective HIV Vaccine. J Clin Cell Immunol 2017; 8(4): 518.
59. *Létourneau S, Im EJ, Mashishi T, Brereton C, Bridgeman A, Yang H,* et al. Design and pre-clinical evaluation of a universal HIV-1 vaccine. PLoS One 2007; 2(10): e984.
60. *Ondondo B, Murakoshi H, Clutton G, Abdul-Jawad S, Wee EG, Gatanaga H,* et al. Novel Conserved-region T-cell Mosaic Vaccine With High Global HIV-1 Coverage Is Recognized by Protective Responses in Untreated Infection. Mol Ther 2016; 24(4): 832–42.
61. *Fischer W, Perkins S, Theiler J, Bhattacharya T, Yusim K, Funkhouser R,* et al. Polyvalent vaccines for optimal coverage of potential T-cell epitopes in global HIV-1 variants. Nat Med 2007; 13(1): 100–6.
62. *Sanou MP, Roff SR, Mennella A, Sleasman JW, Rathore MH, Yamamoto JK,* et al. Evolutionarily conserved epitopes on human immunodeficiency virus type 1 (HIV-1) and feline immunodeficiency virus reverse transcriptases detected by HIV-1-infected subjects. J Virol 2013; 87(18): 10004–15.
63. *Roff SR, Sanou MP, Rathore MH, Levy JA, Yamamoto JK.* Conserved epitopes on HIV-1, FIV and SIV p24 proteins are recognized by HIV-1 infected subjects. Hum Vaccin Immunother 2015; 11(6): 1540–56.
64. *Ferguson AL, Mann JK, Omarjee S, Ndung'u T, Walker BD, Chakraborty AK.* Translating HIV sequences into quantitative fitness landscapes predicts viral vulnerabilities for rational immunogen design. Immunity 2013; 38(3): 606–17.
65. *Leslie AJ, Pfafferott KJ, Chetty P, Draenert R, Addo MM, Feeney M,* et al. HIV evolution: CTL escape mutation and reversion after transmission. Nat Med 2004; 10(3): 282–9.
66. *Borthwick N, Ahmed T, Ondondo B, Hayes P, Rose A, Ebrahimsa U,* et al. Vaccine-elicited human T cells recognizing conserved protein regions inhibit HIV-1. Mol Ther 2014; 22(2): 464–75.
67. *Hanke T.* Conserved immunogens in prime-boost strategies for the next-generation HIV-1 vaccines. Expert Opin Biol Ther 2014; 14(5): 601–16.
68. *Samri A, Bacchus-Souffan C, Hocqueloux L, Avettand-Fenoel V, Descours B, Theodorou I,* et al. Polyfunctional HIV-specific T cells in Post-Treatment Controllers. AIDS 2016; 30(15): 2299–302.
69. *De Souza MS, Ratto-Kim S, Chuenarom W, Schuetz A, Chantakulkij S, Nuntapinit B*, et al. The Thai phase III trial (RV144) vaccine regimen induces T cell responses that preferentially target epitopes within the V2 region of HIV-1 envelope. J Immunol 2012; 188(10): 5166–76.
70. *Soghoian DZ, Jessen H, Flanders M, Sierra-Davidson K, Cutler S, Pertel T,* et al. HIV-specific cytolytic CD4 T cell responses during acute HIV infection predict disease outcome. Sci Transl Med 2012; 4(123): 123ra25.
71. *MacDonald KS, Fowke KR, Kimani J, Dunand VA, Nagelkerke NJ, Ball TB,* et al. Influence of HLA supertypes on susceptibility and resistance to human immunodeficiency virus type 1 infection. J Infect Dis 2000; 181(5): 1581–9.
72. *MacDonald KS, Embree JE, Nagelkerke NJ, Castillo J, Ramhadin S, Njenga S,* et al. The HLA A2/6802 supertype is associated with reduced risk of perinatal human immunodeficiency virus type 1 transmission. J Infect Dis 2001; 183(3): 503–6.
73. *Koehler RN, Walsh AM, Saathoff E, Tovanabutra S, Arroyo MA, Currier JR,* et al. Class I HLA-A*7401 is associated with protection from HIV-1 acquisition and disease progression in Mbeya, Tanzania. J Infect Dis 2010; 202(10): 1562–6. Erratum in: J Infect Dis 2011; 203(5): 749.
74. *Peterson TA, Kimani J, Wachihi C, Bielawny T, Mendoza L, Thavaneswaran S,* et al. HLA class I associations with rates of HIV-1 seroconversion and disease progression in the Pumwani Sex Worker Cohort. Tissue Antigens 2013; 81(2): 93–107.
75. *Liu C, Carrington M, Kaslow RA, Gao X, Rinaldo CR, Jacobson LP,* et al. Association of polymorphisms in human leukocyte antigen class I and transporter associated with antigen processing genes with resistance to human immunodeficiency virus type 1 infection. J Infect Dis 2003; 187(9): 1404–10.
76. *Farquhar C, Rowland-Jones S, Mbori-Ngacha D, Redman M, Lohman B, Slyker J,* et al. Human leukocyte antigen (HLA) B*18 and protection against mother-to-child HIV type 1 transmission. AIDS Res Hum Retroviruses 2004; 20(7): 692–7.

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Changing panorama in risk factors of cerebral palsy

Promenljiva slika faktora rizika od cerebralne paralize

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Abstract

Background/Aim. The etiology of cerebral palsy (CP) is multifactorial and not yet fully understood. The aim of this study was to determine the changes in CP etiological factors over the past 30 years. **Methods.** A retrospective study analyzed a database of 296 individuals with CP. Risk factors (RFs) were divided into preconception, antenatal, intrapartum, and neonatal. Patients on the register were divided into three cohorts: those born before 2000, the ones born between 2000 and 2010, and those born after 2010. The changes in CP RFs were investigated at ten-year intervals. **Results.** The five RFs with the highest total frequency were low birth weight (46.62%), prematurity (44.25%), advanced maternal age – over 35 years (37.16%), emergency cesarean section (33.78%), and birth asphyxia (25.33%). The consanguineous marriage rate was 22.29%. **Conclusion.** Low birth weight and prematurity rates, which are the most frequently identified RFs, are gradually increasing. The rate of birth asphyxia has decreased in the last ten years. The rate of advanced maternal age is increasing, and consanguineous marriage is still an important RF in Turkey.

Key words:
birth weight; causality; cerebral palsy; infant, premature; risk factors.

Apstrakt

Uvod/Cilj. Etiologija cerebralne paralize (CP) je multifaktorijalna i još uvek nije potpuno razjašnjena. Cilj rada bio je da se utvrde promene u etiološkim faktorima CP tokom poslednjih 30 godina. **Metode.** Retrospektivnom studijom analizirana je baza podataka 296 bolesnika sa CP. Faktori rizika (FR) podeljeni su u prekonceptualne, antenatalne, intrapartalne i neonatalne. Bolesnici u registru podeljeni su u tri kohorte: na bolesnike rođene pre 2000. godine, one rođene između 2000. i 2010. godine i bolesnike rođene posle 2010. godine. Promene u FR od CP istraživane su u intervalima od po deset godina. **Rezultati.** Najveću učestalost imalo je sledećih pet FR: niska porođajna težina (46,62%), prevremenost (44,25%), životno doba majke iznad 35 godina starosti (37,16%), hitni carski rez (33,78%) i asfiksija pri porođaju (25,33%). Stopa brakova između srodnika iznosila je 22,29%. **Zaključak.** Stope niske porođajne težine i prevremenost, koji su najčešće označeni FR od CP, postepeno se poboljšavaju. Stopa asfiksije pri porođaju smanjena je u poslednjih deset godina. Stopa 'odmakla starost majke' raste, a brak između srodnika i dalje predstavlja važan FR u Turskoj.

Ključne reči:
telesna masa, rođenje; etiološki faktori; cerebralna paraliza; nedonošče; faktori rizika.

Introduction

Cerebral palsy (CP) is a group of permanent disorders affecting the development of movement and posture, causing a limitation of activity, attributed to nonprogressive disturbances that occur in the developing fetal or infant brain. The motor disorders of CP are often accompanied by disturbances of sensation, perception, cognition, communication, and behavior associated with epilepsy and secondary musculoskeletal problems [1].

The birth prevalence of CP has been calculated as 1.2–2.5/1,000 [2]. In a study conducted in Turkey, the prevalence of CP was found to be 4.4 *per* 1,000 live births [3]. The etiology of CP is multifactorial and not yet fully understood. While CP may be the result of exposure to a single etiological factor such as perinatal asphyxia, irreversible

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brain damage may also occur as a result of consecutive exposure to many factors [4]. Fahey et al. [5] reported that genetic mutations may be responsible for a substantial proportion of CP cases. At the same time, an environmental second hit those who suffer from a genetic susceptibility to CP may also be an important factor [6]. It has been reported that polymorphism in the catalase antioxidant gene is associated with decreased defense capacity against reactive oxygen species and causes higher susceptibility to CP in infants with perinatal hypoxic-ischemic encephalopathy [7]. Furthermore, no specific etiological factor can be currently identified in more than 50–75% of CP cases [8]. Foremost risk factors (RFs) are prematurity and low birth weight (LBW), but the precise etiology of most cases of CP remains obscure.

Identifying a single and clear cause of CP is not always possible. Therefore, the terminology of RF is used to describe the etiology of the disease. Since the *per* pregnancy number for each etiological factor is not known, although the factors mentioned do not fully reflect RFs, these etiological factors will be referred to as RFs in this article as a generally accepted view. It would be accurate to evaluate RFs as a predisposition for the development of CP and the etiology of CP as a process that includes an integrated form of causal evidence [8]. Various RFs change over time, such as changes in pregnancy healthcare and neonatal care. Therefore, CP surveillance should be reviewed frequently in all countries.

In the last 30 years, there have been radical changes in etiological factors and our understanding of them. The factors shown in the etiology of CP will change over time due to the decrease in stillbirth and neonatal mortality rates and the increase in the survival rate of premature babies.

The aim of this study was to investigate the changes in CP RFs by screening individuals with CP who were followed up in a pediatric rehabilitation clinic in the last 30 years and to compare these RFs by CP subtype.

Methods

Participants

This retrospective study was carried out by reviewing the database of 296 individuals with CP followed in the university pediatric rehabilitation clinic. CP RFs and demographic information were obtained from the database. Twenty-two cases with missing information in the register were excluded. No parent declined to be involved in the study.

Research ethics approval was obtained from the Istanbul Medeniyet University Goztepe Ethics Committee (No. 2021/0006, from January 13, 2021). All patients' caregivers gave informed consent with the approval of the Ethics Committee.

Measures

CP subtypes are classified according to the Surveillance for Cerebral Palsy in Europe classification into four groups: spastic (unilateral and bilateral), dyskinetic (dystonic and choreoathetoic), ataxic, and non-classifiable [9].

CP RFs were divided into four groups: preconception, antenatal, intrapartum, and neonatal RFs with reference to the work of McIntyre et al. [10]. Then, RFs in the data were distributed to the appropriate RF group. Findings comprised four preconception, nine antenatal, eight intrapartum, and six neonatal RFs.

Risk factors

The RFs in the first, preconception group, are the following: consanguine marriage, blood incompatibility, maternal age, previous maternal miscarriage, and stillbirths. In the second, antenatal group, the following RFs are present: prematurity, LBW, plurality, prenatal infection, third-trimester hemorrhage, gestational diabetes, preeclampsia, maternal morbidity, and placental/amniotic abnormalities. In the intrapartum group, the RFs are as follows: birth asphyxia (BA), emergency cesarean section (C/S), home birth, premature rupture of membranes, vacuum and forceps, abnormal presentation, cord around neck, and meconium stained. In the final, neonatal group, these RFs are present: postnatal seizure, hypoglycemia, postnatal infection, metabolic and developmental diseases, postnatal cerebrovascular accident (CVA), and prolonged jaundice.

Babies born under 2,500 g were accepted as LBW, and babies born alive before the 37th gestational week were accepted as preterm [11]. Being over 35 years old was accepted as an advanced maternal age. Babies who were diagnosed with neonatal hypoglycemia by the neonatologist in the national health system were included as hypoglycemia. Miscarriage and stillbirths refer to miscarriage and stillbirths in the same woman who has given birth to a child with CP.

The data were classified according to date of birth and divided into three cohorts: those born before 2000, those born between 2000 and 2010, and those born after 2010. Changes in CP RFs were investigated at ten-year intervals. Secondly, the distribution and variation of RFs according to the CP subtype were investigated.

Data analysis

All statistical analyses were performed with SPSS version 25.0 software (IBM, Chicago, IL). Descriptive statistics were presented as frequencies with percentages, for categorical variables and mean ± standard deviation (minimum-maximum) for continuous variables. In order to investigate the relationship between two categorical variables, Pearson's Chi-square and Fisher's exact tests were used. Likewise, multinominal logistic regression was used to predict the prevalence of more than two categories. Statistical significance was determined as $p < 0.05$.

Results

The study included 296 patients, 135 (45.6%) female and 161 (54.4%) male. Their mean age was 11.85 ± 7.20 (min. =

1.00, max. = 38) years; 136 (45.9%) patients were born between 2010 and 2020, 126 (42.6%) were born between 2000 and 2010, and 34 (11.5%) patients were born before 2000.

The distribution according to CP subtypes is shown in Figure 1. The Chi-squared test result shows no significant difference in CP subtypes between cohorts [$\chi^2(6.\ 296) = 8.59$, $p = 0.20$].

Among the 296 patients, 277 (93.6%) had RFs, and the frequencies of RFs are presented in Table 1.

Overall, the five RFs with the highest total frequency were LBW (46.62%), prematurity (44.25%), advanced maternal age (37.16%), emergency C/S (33.78%), and BA (25.33%). The consanguineous marriage rate was 22.29%. When classified according to ten-year groups, the occurrence

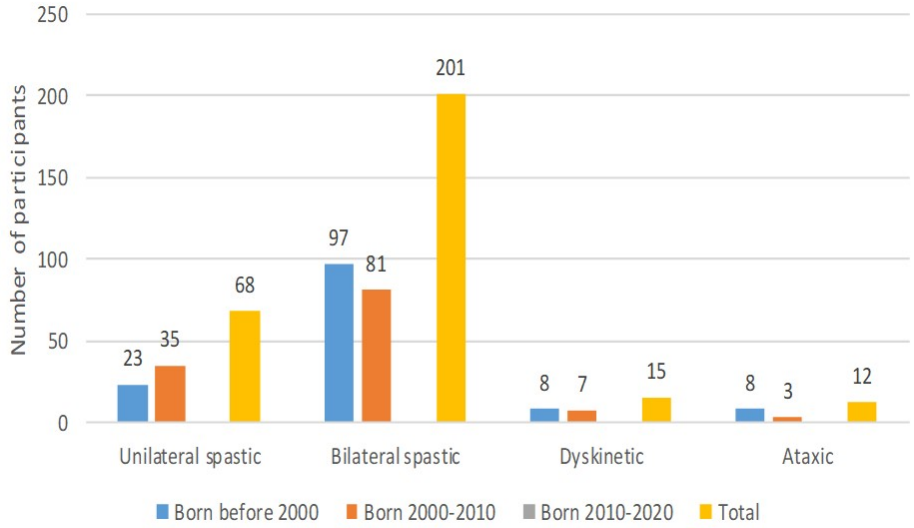

Fig. 1 – Distribution of cerebral palsy subtypes according to age groups.

Table 1

Frequencies of group/particular risk factors for cerebral palsy depending on the age group of patients

Risk factors	Born 2010–2020	Born 2000–2010	Born before 2000	Total
Preconception	79 (46.20)	71 (41.52)	21 (12.28)	171 (57.8)
consanguineous marriage	28 (42.42)	31 (46.97)	7 (10.61)	66 (38.6)
blood incompatibility	1 (8.33)	7 (58.33)	4 (33.33)	11 (6.4)
advanced maternal age	61 (55.45)	38 (34.54)	11 (10.00)	110 (64.3)
miscarriage and stillbirths	14 (27.45)	23 (45.10)	14 (27.45)	51 (29.8)
Antenatal	91 (50.00)	74 (40.66)	17 (9.34)	182 (61.5)
prematurity	77 (58.78)	45 (34.35)	9 (6.87)	131 (72.0)
low birth weight	82 (59.42)	46 (33.33)	10 (7.25)	138 (75.8)
plurality	10 (62.50)	5 (31.25)	1 (6.25)	16 (8.8)
prenatal infection	1 (11.11)	5 (55.56)	3 (33.33)	9 (4.9)
3rd trimester hemorrhage	12 (37.50)	17 (53.13)	3 (9.37)	32 (17.6)
gestational diabetes	2 (25.00)	5 (62.50)	1 (12.50)	8 (4.4)
preeclampsia	13 (41.94)	13 (41.94)	5 (16.12)	31 (17.0)
maternal morbidity	5 (62.5)	3 (37.5)	0 (0.00)	8 (4.4)
placental/amniotic abnormalities	5 (100.00)	0 (0.00)	0 (0.00)	3 (1.6)
Intrapartum	73 (51.05)	59 (41.26)	11 (7.69)	143 (48.3)
birth asphyxia	26 (34.67)	39 (52.00)	10 (13.33)	75 (52.4)
emergency C/S	67 (67.00)	32 (32.00)	1 (1.00)	100 (69.9)
home birth	2 (50.00)	0 (0.00)	2 (50.00)	4 (2.8)
premature rupture of membranes	1 (14.29)	6 (85.71)	0 (0.00)	7 (4.9)
vacuum and forceps	2 (50.00)	1 (25.00)	1 (25.00)	4 (2.8)
abnormal presentation	0 (0.00)	3 (75.00)	1 (25.00)	4 (2.8)
cord around neck	0 (0.00)	3 (60.00)	2 (40.00)	5 (3.5)
meconium stained	0 (0.00)	3 (75.00)	1 (25.00)	4 (2.8)
Neonatal	34 (37.78)	40 (44.44)	16 (17.78)	90 (30.4)
postnatal seizure	10 (27.78)	17 (47.22)	9 (25.00)	36 (40.0)
hypoglycemia	0 (0.00)	3 (75.00)	1 (25.00)	4 (4.4)
postnatal infection	8 (36.36)	11 (50.00)	3 (13.64)	22 (24.4)
metabolic and developmental diseases	9 (56.25)	5 (31.25)	2 (12.50)	16 (17.8)
postnatal CVA	4 (50.00)	3 (37.50)	1 (12.50)	8 (8.9)
prolonged jaundice	6 (46.15)	6 (46.15)	1 (7.70)	13 (14.4)

C/S – cesarean section; CVA – cerebrovascular accident; Chi-square test used.
All values are given as numbers (percentages).

of premature birth in groups was 26.47%, 35.71%, and 56.61% (born between 2010 and 2020, born between 2000 and 2010, and born before 2000, respectively), and LBW in groups was 29.41%, 36.50%, and 60.30% (born between 2010 and 2020, born between 2000 and 2010, and born before 2000, respectively).

A Chi-square test for independence indicated no significant association between cohorts and RF groups of preconception, antenatal, and intrapartum, but a significant association was found between cohorts and neonatal RF group (Table 2).

Statistically significant associations for independence results between cohorts and RFs are shown in Table 3 (using a Chi-square test).

A Chi-square test for independence indicated no significant association between CP subtypes and RF groups of intrapartum and neonatal but a significant association between CP subtypes and preconception and antenatal RF groups (Table 4). *Post-hoc* analysis (adjusted residual analysis) was conducted to investigate which subtype of CP was significantly different. According to the Bonferroni adjustment, the critical *p*-value was determined as 0.006 (0.05/8 = 0.006). The association between antenatal RFs unilateral and bilateral spastic CP was statistically significant, $p < 0.006$.

Fisher's exact test results showed a significant association between preconception RFs and CP subtypes in patients born between 2010 and 2020, $p < 0.05$. Moreover, there was a significant association between antenatal RFs and CP subtypes in those born between 2000 and 2010, $p < 0.01$.

The association between placental/amniotic anomalies and ataxic CP was statistically significant, $p < 0.006$. For premature rupture of membranes, it was not possible to talk about a univariate significance.

Table 2

Frequency of group of risk factors for cerebral palsy, depending on the age group of patients

Risk factors	Born 2010–2020	Born 2000–2010	Born before 2000	*p*-values
Preconception	79 (26.7)	71 (24.0)	21 (7.1)	0.85
Antenatal	91 (30.7)	74 (25.0)	17 (5.7)	0.14
Intrapartum	73 (24.7)	59 (19.9)	11 (3.7)	0.08
Neonatal	34 (11.5)	40 (13.5)	16 (5.4)	0.04[a]

All values are given as numbers (percentages). [a]Statistically significant at level $p < 0.05$; Chi-square test used.

Table 3

Relation between the presence of particular/group cerebral palsy risk factors and age group of patients

Risk factors	Born 2010–2020		Born 2000–2010		Born before 2000		*p*-values
	present	absent	present	absent	present	absent	
Preconception							
blood incompatibility*	1 (0.3)	135 (45.6)	7 (2.4)	119 (40.2)	4 (1.4)	30 (10.1)	0.004[b]
advanced maternal age	61 (20.6)	75 (25.3)	38 (12.8)	88 (29.7)	11 (3.7)	23 (7.8)	0.04[a]
miscarriage and stillbirths	14 (4.7)	122 (41.2)	23 (7.8)	103 (34.8)	14 (4.7)	20 (6.8)	< 0.001[c]
Antenatal							
prematurity	77 (26.0)	59 (19.9)	45 (15.2)	81 (27.4)	9 (3.0)	25 (8.4)	< 0.001[c]
low birth weight	82 (27.7)	54 (18.2)	46 (15.5)	80 (27.0)	10 (3.4)	24 (8.1)	< 0.001[c]
prenatal infection*	1 (0.3)	135 (45.6)	5 (1.7)	121 (40.9)	3 (1.0)	31 (10.5)	0.04[a]
Intrapartum							
emergency C/S	67 (22.6)	69 (23.3)	32 (10.8)	94 (31.8)	1 (0.3)	33 (11.1)	< 0.001[c]
home birth*	2 (0.7)	134 (45.3)	0 (0.0)	126 (42.6)	2 (0.7)	32 (10.8)	0.04[a]
cord around neck*	0 (0.0)	136 (45.9)	3 (1.0)	123 (41.6)	2 (0.7)	32 (10.8)	0.02[a]
Neonatal							
postnatal seizure*	10 (3.4)	126 (42.6)	17 (5.7)	109 (36.8)	9 (3.0)	25 (8.4)	0.01[b]

C/S – cesarean section. All values are given as numbers (percentages).
Statistically significant at level: [a]$p < 0.05$, [b]$p < 0.01$, [c]$p < 0.001$; *Fisher's exact test.

Table 4

Relation between particular/group cerebral palsy risk factors and subtypes

Parameters	US	BS	D	A	*p*-values
Preconception	43 (14.5)	110 (37.2)	7 (2.4)	11 (3.7)	0.04[a]
Antenatal*	32 (10.8)	136 (45.9)	7 (2.4)	7 (2.4)	0.01[b]
prematurity	18 (6.1)	105 (35.5)	3 (1.0)	5 (1.7)	< 0.001[c]
low birth weight	21 (7.1)	105 (35.5)	5 (1.7)	7 (2.4)	0.01[b]
placental/amniotic abnormalities*	1 (0.3)	2 (0.7)	0 (0.0)	2 (0.7)	0.03[a]
Intrapartum	33 (11.1)	99 (33.4)	4 (1.4)	7 (2.4)	0.34
premature rupture of membranes	3 (1.0)	2 (0.7)	1 (0.3)	1 (0.3)	0.04[a]
Neonatal*	19 (6.4)	64 (21.6)	5 (1.7)	2 (0.7)	0.71

US – unilateral spastic; BS – bilateral spastic; D – dyskinetic; A – ataxic.
All values are given as numbers (percentages).
Statistically significant at level: [a]$p < 0.05$, [b]$p < 0.01$, [c]$p < 0.001$; *Fisher's exact test; Chi-square test used.

Regarding the association between CP subtypes and listed RFs investigated by cohorts, it was found that Fisher's exact test results showed that there was a significant association between prematurity and CP subtypes in cohorts born between 2000 and 2010, $p < 0.01$. Furthermore, there was a significant association between LBW and CP subtypes in cohorts born between 2000 and 2010, $p < 0.05$.

Discussion

This study is the most recent on CS RFs in Turkey. In this study, the changes in CS RFs were investigated at ten-year intervals. The distribution of CP subtypes is compatible with the literature [2, 12]. The foremost RFs were found to be LBW, prematurity, advanced maternal age, emergency C/S, and BA.

No significant change was detected in the distribution of preconception, antenatal, and intrapartum RFs at ten-year intervals. However, neonatal RFs were found to be significantly higher in the group born before 2000. Among the neonatal RFs, the postnatal seizure rate has decreased significantly. There was no patient with a history of cord around the neck among patients born within the last ten years. These findings can be explained by the fact that neonatal RFs are now better controlled.

In literature, preterm delivery is still one of the main RFs for CP [13, 14], with more than 40% of individuals with CP born preterm [15]. The prematurity rate was found to be 45% in the United States of America and 24.8% to 48.4% in studies in Turkey [3, 16–18]. In the study by Tosun et al. [17], the LBW rate was found to be approximately 30%. In the literature, intrauterine growth restriction, which results in LBW, is one of the most important RFs in term babies [19, 20]. Premature birth in groups 26.47%, 35.71%, 56.61%, and LBW in groups 29.41%, 36.50%, and 60.30%, respectively, gradually, increase with each decade in accordance with the literature [19, 20]. However, rates of prematurity and LBW in the literature found in the last ten years were even much higher than the data in older literature. In other words, in addition to the increase reported in the literature, the rates continue to increase. This may be the result of increasing technology and intensive care services and keeping premature and LBW babies alive.

Although prematurity is the most frequently accused etiology when the most common RFs in term babies were investigated, babies with neonatal encephalopathy and perinatal stroke were reported as high risk, and babies with congenital disabilities, septicemia, meningitis, and small for gestational age were reported as moderate risk [20]. In our study, according to the total number of cases, the frequency of postnatal factors such as septicemia, meningitis, and perinatal stroke was much less than antenatal and intrapartum causes. This can be attributed to several factors. Firstly, antenatal and intrapartum factors are often more controllable and preventable through medical interventions during pregnancy and childbirth. This includes timely detection and treatment of prenatal bacterial infections or complications during labor and delivery. Additionally, advancements in postnatal care and medical technology have significantly reduced the impact of postnatal complications. Modern neonatal intensive care units and perinatology expertise allow for prompt diagnosis and management of postnatal issues. Moreover, underreporting or challenges in diagnosing and recording postnatal factors may contribute to their seemingly lower frequency in the study. Lastly, genetic or biological factors could also play a role, with postnatal effects being less pronounced or identifiable compared to pre-existing conditions. These combined factors underscore the importance of a comprehensive assessment and interpretation of study findings regarding the frequency of different causal factors in CP.

It was observed that preconception and antenatal RFs were associated with subtypes. In further analysis, only antenatal RFs were found to be significantly associated with unilateral and bilateral CP. It can be concluded that antenatal RFs (among which prematurity and LBW are the most common) are still the most important RFs. Managing antenatal RFs, mainly by preventing preterm births, becomes important in the prevention of spastic CP, which is the most common subtype.

The maternal age at first birth has increased significantly both in Europe and America, as in Turkey [21, 22]. In many countries, women postpone their first pregnancy to older ages. Advanced maternal age, which is one of the preconception RFs, was found to be statistically significantly higher in CP cases born in the last ten years. However, advanced maternal age brings with it many pregnancy risks [23]. Wu et al. [19] reported that independent RFs for CP include maternal age over 35, black race, and intrauterine growth restriction. Among the intrapartum RFs, the rate of emergency C/S was found to be significantly higher in the last decade. This may be due to the increase in older-age pregnancies.

The consanguineous marriage rate was found to be 22.29% in this study, and there was no significant difference between the groups. In their study, Tosun et al. [17] compared RFs of individuals with CP followed in the period of 1972–1994 and 1995–2006 and determined consanguineous marriage rates as 16.4% and 10.4%, respectively. Consanguineous marriage was reported as 21.4% in the study by Yılmaz Yalcinkaya et al. [24] and 23.8% in the study by Erkin et al. [18]. Consanguineous marriage rate is still very high in Turkey.

The rate of BA, which is one of the most accused factors in CP etiology in the past years, was found to be 25.33% in this study. McIntyre et al. [10] reported BA strongest and the most consistent intrapartum RF in children born at term in developed countries in their systematic review. Likewise, in Turkey, the perinatal asphyxia rate was reported as 66.3% and 71% in the studies by Tosun et al. [17] and 34.6% in the studies by Erkin et al. [18], much higher than in this study. BA ratio seems to be stable in the first two decades in this study, but a significant decrease is observed in the last ten years. With it still counting among the top-ranked five RFs, it is promising that it tends to decrease gradually.

In this study, ataxic CP was found to be associated with placental anomalies. The relationship of subtypes with BA has been specifically looked into. Although the relationship between BA and CP types was not significant, in addition to the placental anomalies, BA was the most associated RF for ataxic CP. In the studies by Erkin et al. [18], in accordance with this study, BA was observed at a much higher rate in hypoxic and dyskinetic CP compared to the spastic type.

Stillbirth history and blood incompatibility are decreasing RFs. Maternal history of stillbirth was defined as RF for all CP cases [10]. However, the causes of stillbirths are not fully understood. Stillbirths after 24 weeks of pregnancy are primarily due to pregnancy/delivery-related causes such as placental abnormalities, birth defects, and infection [25].

Limitations of the study

One of the limitations of this study is that the results could not be generalized since this was a single-center study. Moreover, since the study was conducted with patients followed up in the university rehabilitation unit, mild cases that were not followed up by a doctor may have been overlooked. One of the limitations is the fact that the study is retrospective and the database analysis structure may have caused many methodological issues, such as recording errors and missings and subjective assessments of different recorders. Although the small sample size in the third cohort (n = 34) may impact the statistical power and generalizability of the results, the observed trends provide valuable insights into the changes in CP RFs over time. Future studies with larger sample sizes across all cohorts are needed to confirm these findings. In addition, since the decrease of some RFs may be the reason for the increase of some, individual evaluation of RFs may not always give accurate results. For instance, the decrease in neonatal mortality is associated with an increase in LBW and premature babies, which may be the most important determinant of CP.

Conclusion

Cerebral palsy is a heterogeneous disability. Although it is expected that many different causal pathways play a role in its etiology, it is important to evaluate these etiological risk factors both individually and interrelated and, more importantly, at regular intervals at the national level. The right intervention should be done at the right time to ensure prevention. Therefore, we need to continuously monitor and report changes in the frequency of these specific risk factors.

Conflict of interest

The authors declare no conflict of interest.

R E F E R E N C E S

1. *Richards CL, Malouin F.* Cerebral palsy: definition, assessment and rehabilitation. Handb Clin Neurol 2013; 111: 183–95.
2. *Johnson A.* Prevalence and characteristics of children with cerebral palsy in Europe. Dev Med Child Neurol 2002; 44(9): 633–40.
3. *Serdaroğlu A, Cansu A, Ozkan S, Tezcan S.* Prevalence of cerebral palsy in Turkish children between the ages of 2 and 16 years. Dev Med Child Neurol 2006; 48(6): 413–6.
4. *Nelson KB.* Causative factors in cerebral palsy. Clin Obstet Gynecol 2008; 51(4): 749–62.
5. *Fahey MC, Maclennan AH, Kretzschmar D, Gecz J, Kruer MC.* The genetic basis of cerebral palsy. Dev Med Child Neurol 2017; 59(5): 462–9.
6. *Esih K, Goričar K, Dolžan V, Rener-Primec Z.* The association between antioxidant enzyme polymorphisms and cerebral palsy after perinatal hypoxic-ischaemic encephalopathy. Eur J Paediatr Neurol 2016; 20(5): 704–8.
7. *Platt MJ, Panteliadis CP, Häusler M.* Aetiological Factors. In: *Panteliadis CP*, editor. Cerebral Palsy: A Multidisciplinary Approach. Cham: Springer International Publishing; 2018. pp. 49–58.
8. *Dammann O.* Philosophy, Epidemiology, and Cerebral Palsy Causation. In: *Panteliadis CP*, editor. Cerebral Palsy. Cham: Springer International Publishing; 2018. pp. 29–33.
9. *Cans C.* Surveillance of cerebral palsy in Europe: a collaboration of cerebral palsy surveys and registers. Surveillance of Cerebral Palsy in Europe (SCPE). Dev Med Child Neurol 2000; 42(12): 816–24.
10. *McIntyre S, Taitz D, Keogh J, Goldsmith S, Badawi N, Blair E.* A systematic review of risk factors for cerebral palsy in children born at term in developed countries. Dev Med Child Neurol 2013; 55(6): 499–508.
11. *Quinn JA, Munoz FM, Gonik B, Frau L, Cutland C, Mallett-Moore T,* et al. Preterm birth: Case definition & guidelines for data collection, analysis, and presentation of immunisation safety data. Vaccine 2016; 34(49): 6047–56.
12. *Sellier E, Platt MJ, Andersen GL, Krägeloh-Mann I, De La Cruz J, Cans C.* Decreasing prevalence in cerebral palsy: a multi-site European population-based study, 1980 to 2003. Dev Med Child Neurol 2016; 58(1): 85–92.
13. *Oskoui M, Coutinho F, Dykeman J, Jetté N, Pringsheim T.* An update on the prevalence of cerebral palsy: a systematic review and meta-analysis. Dev Med Child Neurol 2013; 55(6): 509–19. Erratum in: Dev Med Child Neurol 2016; 58(3): 316.
14. *Himpens E, Van den Broeck C, Oostra A, Calders P, Vanhaesebrouck P.* Prevalence, type, distribution, and severity of cerebral palsy in relation to gestational age: a meta-analytic review. Dev Med Child Neurol 2008; 50(5): 334–40.
15. *Smithers-Sheedy H, McIntyre S, Gibson C, Meehan E, Scott H, Goldsmith S,* et al. A special supplement: findings from the Australian Cerebral Palsy Register, birth years 1993 to 2006. Dev Med Child Neurol 2016; 58 Suppl 2: 5–10.
16. *Allen MC.* Neurodevelopmental outcomes of preterm infants. Curr Opin Neurol 2008; 21(2): 123–8.
17. *Tosun A, Gökben S, Serdaroğlu G, Polat M, Tekgül H.* Changing views of cerebral palsy over 35 years: the experience of a center. Turk J Pediatr 2013; 55(1): 8–15.
18. *Erkin G, Delialioglu SU, Ozel S, Culha C, Sirzai H.* Risk factors and clinical profiles in Turkish children with cerebral palsy: analysis of 625 cases. Int J Rehabil Res 2008; 31(1): 89–91.
19. *Wu YW, Croen LA, Shah SJ, Newman TB, Najjar DV.* Cerebral palsy in a term population: risk factors and neuroimaging findings. Pediatrics 2006; 118(2): 690–7.

20. *Morgan C, Fahey M, Roy B, Novak I.* Diagnosing cerebral palsy in full-term infants. J Paediatr Child Health 2018; 54(10): 1159–64.
21. *Shadyab AH, Gass ML, Stefanick ML, Waring ME, Macera CA, Gallo LC,* et al. Maternal Age at Childbirth and Parity as Predictors of Longevity Among Women in the United States: The Women's Health Initiative. Am J Public Health 2017; 107(1): 113–9.
22. *Tromp M, Ravelli AC, Reitsma JB, Bonsel GJ, Mol BW*. Increasing maternal age at first pregnancy planning: health outcomes and associated costs. J Epidemiol Community Health 2011; 65(12): 1083–90.
23. *Lampinen R, Vehviläinen-Julkunen K, Kankkunen P.* A review of pregnancy in women over 35 years of age. Open Nurs J 2009; 3: 33–8.
24. *Yilmaz Yalçinkaya E, Hüner B, Dinçer Ü, Diraçoğlu D, Aydin R, İçağasioğlu A,* et al. Demographic and Clinical Findings of Cerebral Palsy Patients in Istanbul: A Multicenter Study. Turk J Phys Med Rehab 2014; 60: 134–8.
25. *Stillbirth Collaborative Research Network Writing Group.* Causes of death among stillbirths. JAMA 2011; 306(22): 2459–68.

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Risk factors and preventive measures for abnormal blood pressure during hemodialysis filtration for patients with sepsis

Faktori rizika i mere prevencije poremećaja krvnog pritiska tokom hemodijafiltracije kod bolesnika sa sepsom

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Abstract

Background/Aim. Hemodialysis filtration (HDF) plays an extremely important role in treating patients with sepsis and subsequent acute renal failure. However, abnormal blood pressure (ABP) during HDF badly influences the prognosis and increases all-cause mortality in patients with sepsis. The aim of the study was to investigate risk factors and preventive measures of ABP during HDF for patients with sepsis. **Methods.** A total of 145 patients with sepsis undergoing HDF were included in this study, and they were divided into two groups: the normal blood pressure (NBP) group (n = 89) and the ABP group (n = 56). Their clinical data were collected, and the independent influencing factors for ABP during HDF were assessed by univariate and multivariate logistic regression analyses. A nomogram model for prediction was constructed based on the results of multivariate analysis, and its discrimination and consistency were assessed using receiver operating characteristic and calibration curves. The Kaplan-Meier method was used to plot the survival curve to evaluate the prognosis 28 days after HDF. **Results.** Multivariate logistic regression analysis revealed that age, blood calcium, fasting plasma glucose, intact parathyroid hormone, ultrafiltration volume, and ultrafiltration rate were independent risk factors, whereas albumin was a protective factor for ABP during HDF ($p < 0.05$). The nomogram model exhibited a good fitting effect, with high discrimination and accuracy. Kaplan-Meier survival analysis showed that the NBP group had a significantly higher 28-day survival rate than that of ABP (88.76% vs. 73.21%) ($p < 0.05$). **Conclusion.** The constructed risk model is suitable for identifying high-risk groups and provides a reference for effective prevention and treatment, to lower the incidence rate of ABP and improve the prognosis.

Key words:
acute kidney injury; blood pressure; blood pressure determination; dialysis; prognosis; risk factors; sepsis.

Apstrakt

Uvod/Cilj. Hemodijafiltracija (HDF) ima izuzetno važnu ulogu u lečenju bolesnika sa sepsom i posledičnom akutnom bubrežnom slabošću. Međutim, poremećaj krvnog pritiska (PKP) tokom HDF loše utiče na prognozu i povećava smrtnost od svih uzroka kod bolesnika sa sepsom. Cilj rada bio je da se ispitaju faktori rizika i mere prevencije PKP tokom HDF kod bolesnika sa sepsom. **Metode.** U ovu studiju je ukupno bilo uključeno 145 bolesnika sa sepsom koji su bili podvrgnuti HDF, i bili su podeljeni u dve grupe: grupu sa normalnim krvnim pritiskom (NKP) (n = 89) i grupu sa PKP (n = 56). Prikupljeni su njihovi klinički podaci, a nezavisni faktori uticaja na PKP tokom HDF procenjeni su univarijantnom i multivarijantnom logističkom regresionom analizom. Na osnovu rezultata multivarijantne analize konstruisan je model nomograma za predviđanje i njegova diskriminacija i konzistencija bile su procenjene korišćenjem *receiver operating characteristic* i *cacalibration* krive. Da bi se procenila prognoza 28 dana posle HDF, za crtanje krive preživljavanja korišćen je Kaplan-Majerov metod. **Rezultati.** Multivarijantnom logističkom regresionom analizom otkriveno je da su životno doba, kalcijum u krvi, glukoza u plazmi natašte, intaktni paratireoidni hormon, volumen ultrafiltracije i brzina ultrafiltracije bili nezavisni faktori rizika, dok je albumin bio faktor protekcije od PKP tokom HDF ($p < 0,05$). Model nomograma pokazao je dobar efekat uklapanja, sa visokom diskriminacijom i tačnošću. Kaplan-Majerova analiza preživljavanja pokazala je da je grupa sa NKP imala statistički značajno višu 28-dnevnu stopu preživljavanja u odnosu na grupu sa PKP (88,76% vs. 73,21%) ($p < 0,05$).

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Zaključak. Konstruisani model rizika je pogodan za identifikaciju visokorizičnih grupa i pruža preporuku za efikasnu prevenciju i lečenje, u cilju snižavanja stope incidencije PKP i poboljšanja prognoze.

Ključne reči:
bubreg, akutna insuficijencija; krvni pritisak; krvni pritisak, merenje; hemodijaliza; prognoza; faktori rizika; sepsa.

Introduction

Sepsis is an infection-induced systemic inflammatory response syndrome in humans. Its pathogenesis is that the overloaded inflammatory mediators in the body trigger responses to infections, which often damage multiple organs or tissues, thus easily resulting in the death of patients [1, 2]. Kidney injury, a common complication of sepsis in patients, can give rise to a plunge in the renal function of patients within a short time, which deteriorates the disease, greatly shortens the lifespan, and leads to poor prognosis of the patients [3]. Hemodialysis filtration (HDF) is an effective treatment method with high safety and reliability, which can sustain the life of patients with sepsis and relieve their disease [4]. The action mechanism is that HDF removes harmful substances from blood through extracorporeal blood circulation, regulates the patients' immune function, and provides solutions to high catabolism and capacity overload, thus extending life and improving patients' quality of life [5]. As one of the common complications during HDF, abnormal blood pressure (BP) – ABP is a vital factor influencing the therapeutic effect of HDF [6]. It has been pointed out that ABP triggers physical pain and pain-related fear [7], and even leads to mesenteric ischemia and thrombosis in severe cases. Clarifying factors influencing ABP in the HDF process is conducive to early prevention and treatment, thus decreasing the incidence rate of ABP and increasing the survival rate of patients with sepsis.

The aim of this study was to investigate the risk factors and corresponding preventive measures of ABP during HDF for patients with sepsis. This study was expected to provide clinical evidence for treating sepsis in patients clinically.

Methods

General data

This study was approved by the Ethics Committee of the Taizhou People's Hospital, China (from June 4, 2019). and performed according to the Declaration of Helsinki. Written informed consent was obtained from all subjects. For this retrospective nested case-control study, 145 patients with sepsis undergoing HDF in our Hospital from June 2019 to December 2021 were selected, including 78 males and 67 females, aged 21–68 years (with an average age of 61.47 ± 10.89 years). The clinical course of sepsis in these patients was 3–8 months, with an average of 5.27 ± 1.34 months. The flow chart of subject selection is shown in Figure 1.

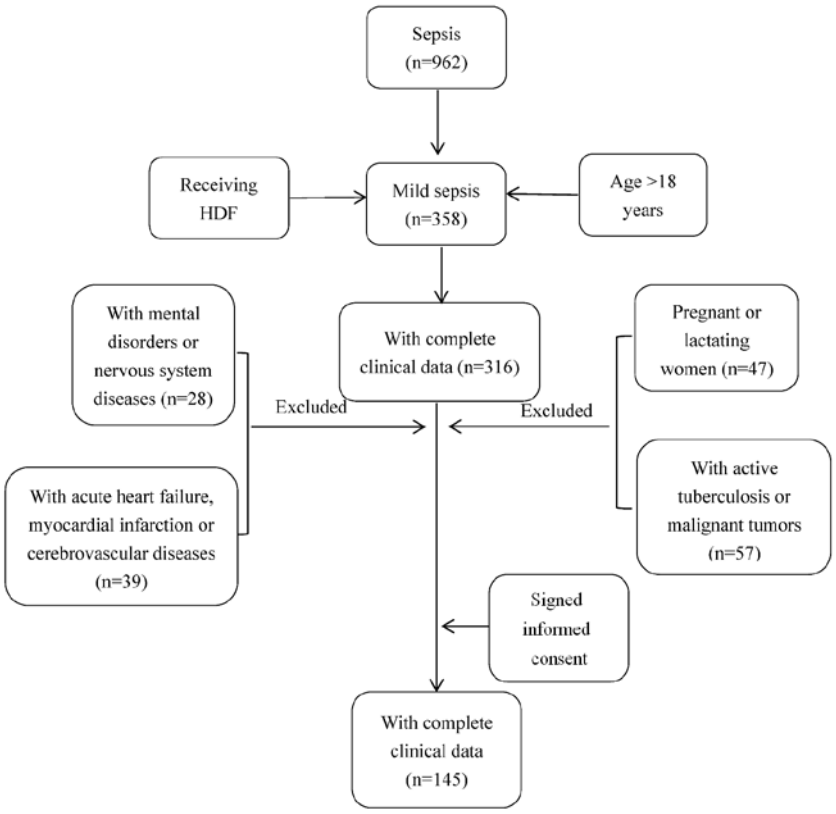

Fig. 1 – Flow chart of subject selection.
HDF – hemodialysis filtration.

Inclusion and exclusion criteria

Inclusion criteria involved: patients over 18 years of age; those meeting the diagnostic criteria for mild sepsis specified in the International Guidelines for the Management of Sepsis and Septic Shock [8]; those receiving HDF; those who received glucocorticoid therapy during dialysis; those with complete clinical data.

Exclusion criteria were set as follows: pregnant or lactating women; patients with active tuberculosis or malignant tumors; those with mental disorders or nervous system diseases; those complicated with acute heart failure, myocardial infarction, or cerebrovascular diseases; those with incomplete clinical data.

Hemodialysis filtration methods

HDF of all patients was performed using Fresenius 4008B hemodialysis machine (Germany) and polysulfone dialysis membranes using the post-dilution method. Blood flow was 200–300 mL/min. HDF was conducted twice to three times a week, 3–4 hrs each time. The dialysate flow was 500 mL/min, the filtration coefficient was 5.5 mL/(h × mmHg), and the surface area of the dialysis membrane was 1.4 m^2.

Blood pressure measurement methods

Invasive BP testing was conducted using the radial artery and *unit* artery as puncture sites. Allen's test was performed before the radial artery puncture; the artery was punctured strictly aseptically and fixed properly. The entire measurement device (BSX-516, Changsha Sinocare Inc., China) included a pressure measurement tube, three-way cannula, pressure sensor, extension tube, heparin saline (1,250 units of heparin sodium in 500 mL of water), and a pressurized bag. During the connection of the arterial indwelling needle, the whole set of tubing was filled with heparin saline, without air leakage or bubbles. After the monitor was connected, the sensor was subjected to zero calibration. After successful calibration, invasive BP monitoring was carried out. Daytime BP (8:00–20:00) and nighttime BP (20:00–8:00) were recorded every 30 min. The effective BP readings throughout the day should be > 80%. The daytime and nighttime BP waveforms and values within 24 hrs were observed.

Diagnostic criteria for abnormal blood pressure

All patients were divided into normal BP (NBP) and ABP groups according to whether they had BP abnormality during HDF. BP abnormality included hypertension and hypotension. The patients were diagnosed with hypotension if the decrease of systolic BP was ≥ 20 mmHg (or if the decrease of the mean arterial pressure was ≥ 10 mmHg) during HDF [9]. The patients were diagnosed with hypertension if the BP rose sharply during HDF or immediately after HDF, and the average arterial pressure increased by at least 15 mmHg [10].

Observational indices

Through literature review of the possible influencing factors for ABP, the clinical data of patients were collected, including 1) basic data: age, gender, body mass index (BMI), complications (hypertension, diabetes mellitus, cerebrovascular and cardiovascular diseases), and primary diseases (diabetic nephropathy, chronic glomerulonephritis, hypertensive renal damage, etc.); 2) blood laboratory indices before HDF: hemoglobin (Hb), albumin, phosphorus, calcium, sodium, total cholesterol (TC), creatinine (Cr), blood urea nitrogen (BUN), fasting plasma glucose (FPG), low-density lipoprotein cholesterol (L-DLC), high-density lipoprotein cholesterol (H-DLC), and intact parathyroid hormone (iPTH); 3) HDF-related data: ultrafiltration volume (UV), ultrafiltration rate (UR), and blood flow; 4) the survival of patients 28 days after HDF observed during the short-term follow-up.

Statistical analysis

Statistical analysis was conducted using SPSS software version 22.0. The normality test was carried out for continuous variables. The measurement data were expressed as mean ± standard deviation, and the count data were represented as numbers (percentages). Intergroup comparison of measurement data conforming to normal distribution was performed by the independent samples *t*-test, and the comparison of count data was conducted with the χ^2 test. With the occurrence of ABP as the dependent variable (Yes = 1, No = 0) and the influencing factors for ABP during HDF in patients with sepsis as the independent variable, stepwise multivariate logistic regression (MLR) analysis was conducted. The values were assigned according to the description in Table 1 (values were assigned to continuous variables after they were converted into binary variables, and then binary variables were converted into numerical variables). A nomogram model for risk prediction was constructed based on the obtained risk factors. The discrimination of the model was examined by plotting the receiver operating characteristic (ROC) curve, and its consistency was evaluated using the

Table 1

Value assignment in multivariate logistic regression analysis

Variable	Values	
	1	0
Dependent		
blood pressure	abnormal	normal
Independent		
gender	female	male
age, years	> 60	> 60
Hemoglobin, g/L	> 110	≤ 60
Albumin, g/L	< 28	≥ 28
Calcium, mmol/L	> 2.30	≤ 2.30
FPG, mmol/L	> 7.9	≤ 7.9
iPTH, pg/mL	> 400	≤ 400
Ultrafiltration volume, mL	> 2,200	≤ 2,200
Ultrafiltration rate, mL/min	> 10	≤ 10

For abbreviations, see Figure 2.

calibration curve. The Kaplan-Meier method was utilized to plot survival curves. The value of $p < 0.05$ represented a statistically significant difference.

Results

Among 145 patients with sepsis, 56 patients were in the ABP (with ABP) and 89 in the NBP group (with no related symptoms). According to the results of univariate analysis of clinical data of patients in the two groups, statistically significant differences were detected concerning gender, age, Hb, albumin, blood calcium, FPG, iPTH, UV, and UR between the two groups of patients ($p < 0.05$). However, no statistically significant differences were observed concerning BMI, complications, primary diseases, TC, Cr, BUN, L-DLC, and H-DLC between the two groups of patients ($p > 0.05$) (Table 2).

MLR analysis revealed that age, blood calcium, FPG, iPTH, UV, and UR were independent risk factors, whereas albumin was a protective factor against ABP during HDF in patients with sepsis ($p < 0.05$) (Table 3).

Table 2

Univariate logistic regression analysis results of clinical data

Variable	ABP group (n = 56)	NBP group (n = 89)	*t*-value/χ^2 value	*p*-value
Male, n (%)	24 (42.86)	54 (60.67)	4.390	0.036
Age, years	63.91 ± 11.32	59.93 ± 10.32	2.068	0.040
BMI, kg/m^2	24.66 ± 4.89	25.57 ± 4.28	1.179	0.240
Complication, n (%)				
hypertension	16 (28.57)	18 (20.22)	1.334	0.248
diabetes mellitus	15 (26.79)	13 (14.61)	3.272	0.070
cerebrovascular disease	15 (26.79)	17 (19.10)	1.180	0.277
cardiovascular disease	16 (28.57)	21 (23.60)	0.448	0.503
Primary disease, n (%)				
diabetic nephropathy	13 (23.21)	16 (17.98)	0.589	0.443
chronic glomerulonephritis	11 (19.64)	13 (14.61)	0.631	0.427
hypertensive renal injury	5 (8.93)	9 (10.11)	0.055	0.814
other	4 (7.14)	12 (13.48)	1.408	0.235
Hemoglobin, g/L	108.76 ± 10.65	115.31 ± 10.94	3.546	< 0.001
Albumin, g/L	27.25 ± 4.94	29.55 ± 5.17	2.653	0.009
Phosphorus, mmol/L	1.81 ± 0.56	1.91 ± 0.59	1.013	0.313
Calcium, mmol/L	2.34 ± 0.15	2.25 ± 0.26	2.354	0.020
Sodium, mmol/L	136.74 ± 2.89	137.42 ± 2.61	1.465	0.145
Total cholesterol, mmol/L	4.56 ± 1.31	4.39 ± 1.02	0.874	0.384
Serum creatinine, μmol/L	663.82 ± 79.54	652.73 ± 81.49	0.805	0.422
BUN, mmol/L	19.22 ± 6.15	17.65 ± 4.93	1.695	0.092
FPG, mmol/L	9.32 ± 1.59	6.92 ± 1.42	9.458	< 0.001
L-DLC, mmol/L	2.87 ± 0.63	2.76 ± 0.55	1.108	0.270
H-DLC, mmol/L	1.25 ± 0.32	1.14 ± 0.43	1.648	0.102
iPTH, pg/mL	470.33 ± 152.42	365.26 ± 120.17	4.614	< 0.001
Ultrafiltration volume, mL	2769.48 ± 429.48	2019.65 ± 478.13	9.556	< 0.001
Ultrafiltration rate, mL/min	9.85 ± 2.98	8.56 ± 2.71	2.685	0.008
Blood flow, mL/min	235.47 ± 24.33	241.29 ± 24.75	1.388	0.167
Type of vascular access			0.342	0.559
autologous arteriovenous fistula	50 (89.29)	82 (92.13)		
tunnel-cuffed catheter	6 (10.71)	7 (7.87)		

ABP – abnormal blood pressure; NBP – normal blood pressure; BMI – body mass index; BUN – blood urea nitrogen; FPG – fasting plasma glucose; L-DLC – low-density cholesterol; H-DLC – high-density cholesterol; iPTH – intact parathyroid hormone.
All values are given as numbers (percentages) or mean ± standard deviation.

Table 3

Multivariate logistic regression analysis results of abnormal blood pressure during hemodialysis filtration

Item	OR	95% CI	*p*-value
Gender	2.353	1.044~3.259	0.069
Age	1.096	1.008~1.935	0.005
Hemoglobin	1.854	1.192~2.048	0.057
Albumin	0.691	0.142~0.973	0.012
Calcium	2.814	2.101~4.075	0.010
FPG	2.208	1.340~3.858	0.004
iPTH	2.762	1.549~4.511	0.011
Ultrafiltration volume	1.824	1.029~3.133	0.024
Ultrafiltration rate	3.415	1.483~5.348	0.003

OR – odds ratio; CI – confidence interval.
For other abbreviations, see Figure 2.

The risk nomogram prediction model of ABP during HDF in patients with sepsis was constructed based on the results of MLR analysis (Figure 2). The results revealed that the risk of ABP rose with the increase of age, blood calcium, FPG, iPTH, UV, and UR, but patients with a higher albumin level before HDF had a relatively small risk of ABP.

The discrimination of the nomogram model was evaluated *via* the ROC curve, and the results showed that the area under the curve (AUC) was 0.877 [95% confidence interval (CI): 0.817–0.956]. The maximum likelihood index of the ROC curve was 0.646, with the corresponding sensitivity and specificity of 79.5% and 85.1%, respectively (Figure 3A). The internal validation of the model by the Bootstrapping method (1,000 samples) illustrated that the C-index of the risk prediction model was 0.865, which represented that the nomogram model had a high overall discrimination (Figure 3B).

The calibration curve of the prediction model was plotted. It was found that the model probability curve in predicting ABP during HDF in patients with sepsis had a good fit with the reference probability curve. In addition, there was no statistically significant difference in the Hosmer-Lemeshow test results ($p > 0.05$), and the prediction index of the model was 0.873, representing the high accuracy of the model (Figure 4).

This study's follow-up rate was 100% (145/145), with no lost cases of patients with sepsis. The 28-day survival of the two groups was recorded, and the survival curves of the two groups of patients were plotted using the Kaplan-Meier method. The results manifested that the 28-day survival rate in the ABP group [73.21% (41/56)] was significantly lower than that in the NBP group [88.76% (79/89)] ($p < 0.05$) (Figure 5).

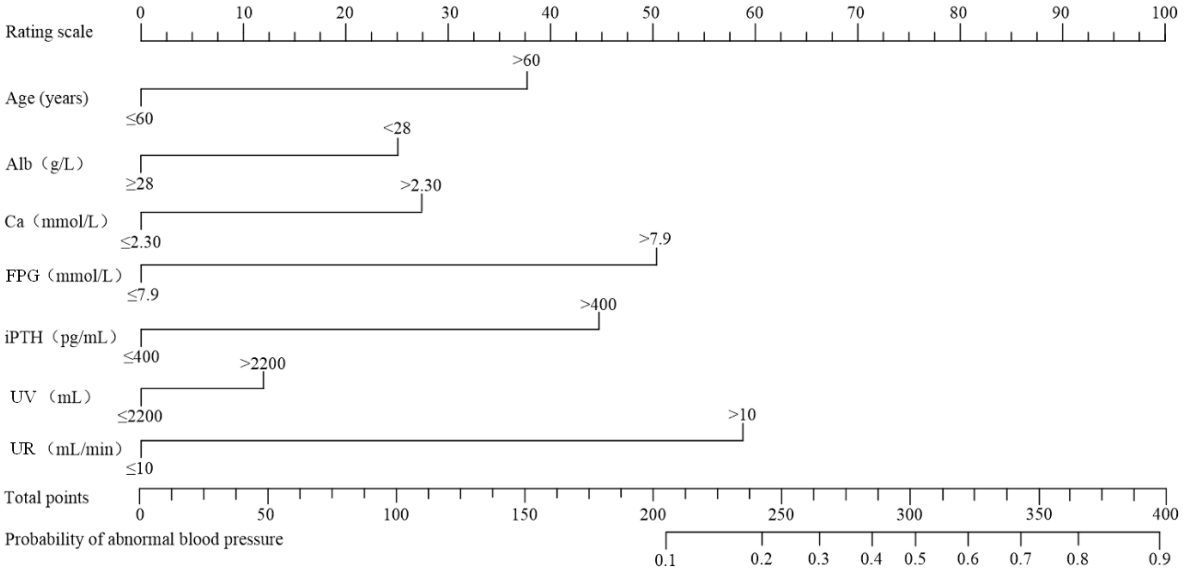

Fig. 2 – Nomogram model for risk prediction of abnormal blood pressure during hemodialysis filtration.
Alb – albumin; Ca – calcium; FPG – fasting plasma glucose; iPTH – intact parathyroid hormone; UV – ultrafiltration volume; UR – ultrafiltration rate.

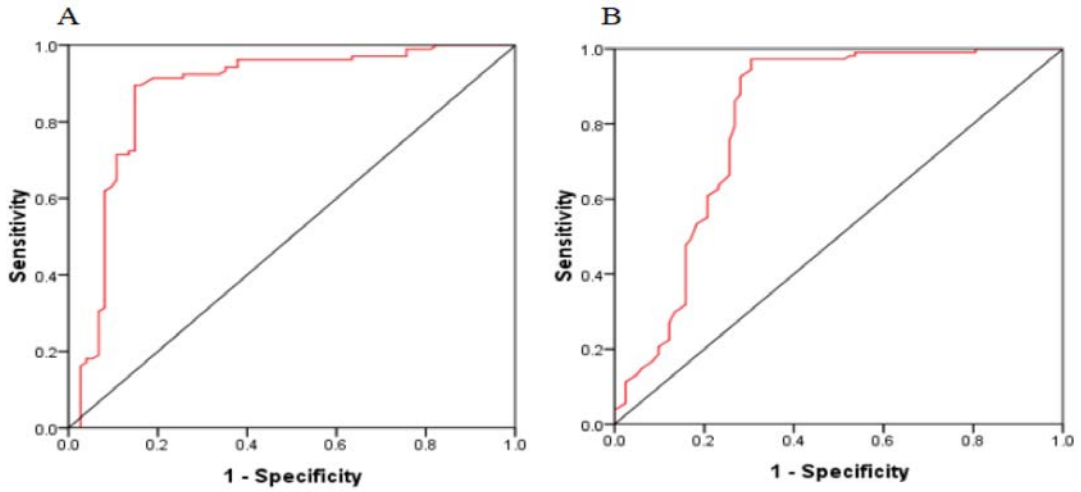

Fig. 3 – Receiver operating characteristic (ROC) curve of the nomogram prediction model.
A) Before internal calibration; B) after internal correction.

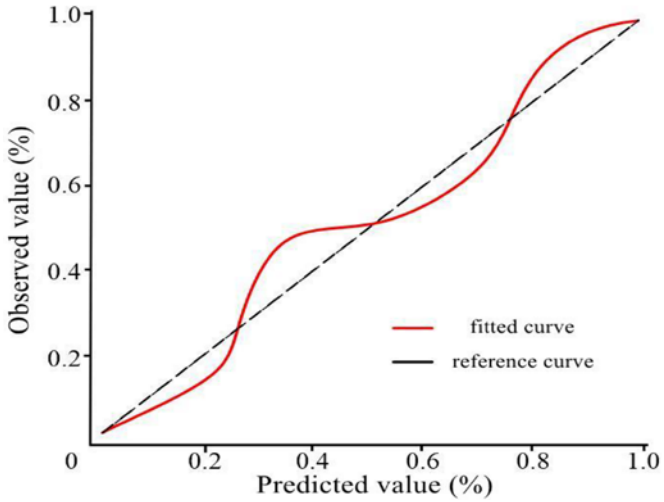

Fig. 4 – Calibration curve of nomogram model for prediction.

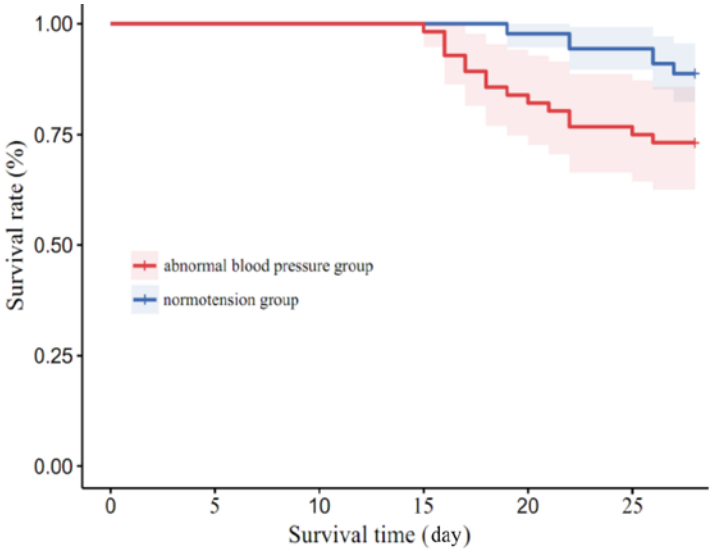

Fig. 5 – Survival curve analysis results.

Discussion

Sepsis is an infection-induced systemic response syndrome featured with high morbidity and mortality rates. Failure to take timely and accurate intervention measures causes organ dysfunction and circulatory disturbance, accompanied by multiple complications, finally leading to aggravation [11, 12]. Hemodialysis, an effective treatment for patients with sepsis, can clear inflammatory cytokines in the body, improve renal function, and prolong the life of patients. It can also adjust the electrolyte balance and acid-base state of the body, so it has been widely used in clinical practice [13]. ABP during HDF in patients is a common complication, which makes the patients more uncomfortable, affects the smooth progress of HDF, and induces other diseases by influencing the adequacy of HDF, thus interfering with the prognosis of patients [14]. ABP during HDF is an independent risk factor for patients' mortality, which remarkably affects the survival rate of patients [15]. Early appropriate measures to interfere with ABP among high-risk patients with sepsis can effectively decrease the incidence rate of ABP and elevate the survival rate of patients with sepsis. In the present study, therefore, the independent influencing factors for ABP during HDF in patients with sepsis were explored to provide a theoretical reference for increasing the survival rate of patients.

Various factors can lead to ABP during HDF in patients. Patients with ABP are older than those with NBP, reflecting that age is a crucial factor affecting BP during HDF [16, 17]. This is consistent with the results of this study which imply that higher age is an independent risk factor for ABP during HDF. The analysis of this study manifested that BP was associated with the function and metabolism of elderly patients. As the patients grow older, the fragility of blood vessels rises. Therefore, hypertension and cardiovascular and cerebrovascular diseases are prone to coincide. This leads to a poor ability to regulate sharp variations in blood volume and increases the probability of ABP during HDF. Albumin is associated with the human body's ability to respond to stress. The stress response declines with decreasing

albumin level, which reduces the effective circulating blood volume and HDF tolerance, thereby increasing the probability of ABP [18]. Blood calcium, a vital index for maintaining body circulation, affects the secretion of anti-inflammatory cytokines by mediating the exocytosis of macrophages. The upregulated calcium^{2+} concentration triggers the occurrence of atherosclerosis and influences the occurrence and development of plaques [19]. Atherosclerosis is an independent influencing factor for cardiovascular disease and death in patients [20], so there is a correlation between a high level of blood calcium and ABP during HDF in patients. This speculation is validated by observation in this study that the blood calcium level of patients with ABP before HDF was remarkably higher than that of patients with NBP, so the blood calcium before HDF was regarded as an independent influencing factor for ABP. Roszkowska-Blaim et al. [21] reported that the probability of hypotension was higher in patients undergoing HDF with a higher FPG level, so the process of HDF and the curative effect on patients were affected. Presumably, the patients with kidney injury and a higher FPG level have a wider variation range of autonomic nerve and vascular diseases, which weakens vascular adaptability during HDF and increases the probability of ABP. With the main function of regulating the blood calcium level, iPTH exerts a significant vasodilating effect. It is able to suppress the effects of multiple hormones such as angiotensin and impede smooth muscle contraction, thereby inducing ABP in patients [22].

The results of this study revealed that the UV and UR of patients in the ABP group were significantly higher than those in the NBP group. The reason is that a larger UV and a higher UR represent a larger liquid clearance volume in the HDF process, leading to a higher tendency to an exceeding level of capillary refilling, reducing the effective circulation volume and increasing the possibility of hypotension. Volume overload between dialysis sessions can aggravate pre-existing hypertension and negatively affect cardiovascular health. This often leads to a higher UR to manage the excess fluid, which can induce adverse outcomes such as abnormal ventricular remodeling and heart failure [23]. Therefore, UV and UR are independent indicators for predicting ABP during HDF in patients with sepsis, and effective control of UV and UR can reduce the incidence rate of ABP.

Based on the above multivariate analysis results, a nomogram model for risk prediction was established, and the discrimination and accuracy of the prediction model were assessed using the ROC and calibration curve. It was found that the constructed prediction model could accurately predict the risk of ABP during HDF in patients with sepsis, which provides a reference for the clinical screening of high-risk septic patients with ABP during HDF. Moreover, the death of patients 28 days after HDF was taken as the endpoint event, and the survival curves of the two groups of patients were drawn and compared in this study. It was discovered that a significant difference in the survival curve was found between the two groups of patients, indicating that ABP during HDF greatly influences the prognosis of patients with sepsis.

In light of the above analysis results of influencing factors for ABP in patients with sepsis, the corresponding nursing measures were proposed to reduce ABP during HDF. Specifically, before HDF, emphasis should be on clinical indices such as age, albumin, blood calcium, FPG, and iPTH of patients with sepsis, especially those with a higher risk of ABP. Besides, exact handovers and records should be guaranteed. Additionally, according to the patient's condition, a reasonable dialysis scheme should be formulated with strict control of the UV and UR. If necessary, drug therapy should be combined or HDF terminated to prevent adverse outcomes.

Limitations of the study

The study had several limitations. First, the research subjects were selected from a single center, with a small sample size. Second, the predictive score in this study was established through retrospective nested case-control analysis of limited clinical data, which was not further verified in a prospective cohort. Third, ABP in patients was only predicted with the presence of abnormality as the dependent variable of the model. In further study, the model can be optimized by considering the occurrence of hypotension or hypertension. Moreover, prospective, multi-center research with enlarged sample sizes can be conducted in the future.

Conclusion

In summary, elderly septic patients with higher levels of blood calcium, fasting plasma glucose, intact parathyroid hormone, ultrafiltration volume, and ultrafiltration rate are prone to abnormal blood pressure during hemodialysis filtration. In contrast, those with a higher albumin level before hemodialysis filtration have a relatively low risk. The constructed risk model is suitable for identifying high-risk groups and provides a reference for reasonable and effective prevention and treatment measures by clinicians to decrease the incidence rate of abnormal blood pressure and improve the prognosis of the patients.

Conflict of interest

The authors declare no conflict of interest.

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R E F E R E N C E S

1. *Addissouky TA, El Tantawy El Sayed I, Ali MMA, Wang Y, El Baz A, Khalil AA,* et al. Molecular Pathways in Sepsis Pathogenesis: Recent Advances and Therapeutic Avenues. J Cell Immunol 2023; 5(6): 174–83.
2. *Rose N, Matthäus-Krämer C, Schwarzkopf D, Scherag A, Born S, Reinhart K,* et al. Association between sepsis incidence and regional socioeconomic deprivation and health care capacity in Germany - an ecological study. BMC Public Health 2021; 21(1): 1636.
3. *Poston JT, Koyner JL.* Sepsis associated acute kidney injury. BMJ 2019; 364: k4891.
4. *Xu J.* A review: continuous renal replacement therapy for sepsis-associated acute kidney injury. All Life 2023; 16: 2163305.
5. *Rajdev K, Leifer L, Sandhu G, Mann B, Pervaiz S, Habib S,* et al. Fluid resuscitation in patients with end-stage renal disease on hemodialysis presenting with severe sepsis or septic shock: A case control study. J Crit Care 2020; 55: 157–62.
6. *Chen Z, Sun F, Shen Y, Ma L, Liu J, Zhou Y.* Impact of Dialysate Sodium Concentration Lowering on Home Blood Pressure Variability in Hemodialysis Patients. Ther Apher Dial 2019; 23(2): 153–9.
7. *Guo L, Ji Y, Sun T, Liu Y, Jiang C, Wang G,* et al. Management of Chronic Heart Failure in Dialysis Patients: A Challenging but Rewarding Path. Rev Cardiovasc Med 2024; 25(6): 232.
8. *Kuipers J, Verboom LM, Ipema KJR, Paans W, Krijnen WP, Gaillard CAJM,* et al. The prevalence of intradialytic hypotension in patients on conventional hemodialysis: a systematic review with meta-analysis. Am J Nephrol 2019; 49(6): 497–506.
9. *Mennuni S, Rubattu S, Pierelli G, Tocci G, Fofi C, Volpe M.* Hypertension and kidneys: unraveling complex molecular mechanisms underlying hypertensive renal damage. J Hum Hypertens 2014; 28(2): 74–9.
10. *Bellomo R, Kellum JA, Ronco C, Wald R, Martensson J, Maiden M,* et al. Acute kidney injury in sepsis. Intensive Care Med 2017; 43(6): 816–28.
11. *Hunt A.* Sepsis: an overview of the signs, symptoms, diagnosis, treatment and pathophysiology. Emerg Nurse 2019; 27(5): 32–41.
12. *Maneta E, Aivalioti E, Tual-Chalot S, Emini Veseli B, Gatsiou A, Stamatelopoulos K,* et al. Endothelial dysfunction and immunothrombosis in sepsis. Front Immunol 2023; 14: 1144229.
13. *Tsevi YM, Dolaama B, Tona KG, Tevi AA, Affanou EC, Amede AD,* et al. Chronic renal failure and hemodialysis in Lomé: are patients on haemodialysis and their entourage well informed? Pan Afr Med J 2021; 39: 85. (French)
14. *Latha Gullapudi VR, White K, Stewart J, Stewart P, Eldehni MT, Taal MW*, et al. An analysis of frequency of continuous blood pressure variation and haemodynamic responses during haemodialysis. Blood Purif 2022; 51(5): 435-49.
15. *Jeong HY, Kim HJ, Han M, Seong EY, Song SH.* Dialysis unit blood pressure two hours after hemodialysis is useful for predicting home blood pressure and ambulatory blood pressure in maintenance hemodialysis patients. Ther Apher Dial 2022; 26(1): 103–14.
16. *Chen TK, Knicely DH, Grams ME.* Chronic Kidney Disease Diagnosis and Management: A Review. JAMA 2019; 322(13): 1294–304.
17. *Okpa HO, Effa EE, Oparah SK, Chikezie JA, Bisong EM, Mbu PN,* et al. Intradialysis blood pressure changes among chronic kidney disease patients on maintenance haemodialysis in a tertiary hospital south - south Nigeria: a 2 year retrospective study. Pan Afr Med J 2019; 33: 91.
18. *Pedreros-Rosales C, Jara A, Lorca E, Mezzano S, Pecoits-Filho R, Herrera P.* Unveiling the clinical benefits of high-volume hemodiafiltration: Optimizing the removal of medium-weight uremic toxins and beyond. Toxins 2023; 15: 531.
19. *Tajbakhsh A, Kovanen PT, Rezaee M, Banach M, Sahebkar A.* Ca2+ Flux: Searching for a Role in Efferocytosis of Apoptotic Cells in Atherosclerosis. J Clin Med 2019; 8(12): 2047.
20. *Iida M, Harada S, Takebayashi T.* Application of Metabolomics to Epidemiological Studies of Atherosclerosis and Cardiovascular Disease. J Atheroscler Thromb 2019; 26(9): 747-57.
21. *Roszkowska-Blaim M, Skrzypczyk P, Jander A, Tkaczyk M, Bałasz-Chmielewska I, Żurowska A, et al.* Effect of hypertension and antihypertensive medications on residual renal function in children treated with chronic peritoneal dialysis. Adv Med Sci 2015; 60(1): 18–24.
22. *Zhou X, Guo Y, Luo Y.* The optimal range of serum intact parathyroid hormone for a lower risk of mortality in the incident hemodialysis patients. Ren Fail 2021; 43(1): 599–605.
23. *Kim TW, Chang TI, Kim TH, Chou JA, Soohoo M, Ravel VA,* et al. Association of ultrafiltration rate with mortality in incident hemodialysis patients. Nephron 2018; 139(1): 13–22.

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A multidisciplinary collaborative care model involving family members in the treatment of type 2 diabetes mellitus and associated diabetes distress in young and middle-aged patients

Multidisciplinarni model nege koji uključuje saradnju porodice u lečenju dijabetesa melitusa tipa 2 i pridruženog dijabetesnog distresa kod mladih i sredovečnih bolesnika

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Abstract

Background/Aim. Diabetes mellitus (DM) represents a significant and enduring health concern and, due to various complications arising from inadequate management, leads to disability and mortality. The aim of this study was to examine the impact of the family-engaged multidisciplinary collaborative care (FEC) model on the management of type 2 DM (T2DM) in young and middle-aged patients, considering the presence of DM distress. **Methods.** The study included 98 patients aged 18 to 59 diagnosed with T2DM and experiencing DM distress. The patients were admitted to the Department of Endocrinology of The First People's Hospital of Jingzhou, in Hubei province, China, between February and December 2023. Using the random number table method, the patients were randomly assigned to either the intervention group (IG) or control group (CG), each consisting of 49 patients. While both groups received standard care, IG additionally received FEC. We assessed and compared glycated hemoglobin (HbA1c) levels, Diabetes Distress Scale (DDS) scores, Summary of Diabetes Self-care Activities (SDSCA) scores, and body mass index (BMI) between the two groups before the intervention and three months after. **Results.** Three months post-intervention, IG exhibited lower HbA1c levels (6.02 ± 0.63 vs. 6.81 ± 0.85) and DDS scores (25.27 ± 2.70 vs. 34.24 ± 4.46) while demonstrating higher SDSCA scores (30.69 ± 1.91 vs. 25.03 ± 2.13) compared to CG. Additionally, the BMI of patients in IG measured 23.83 ± 2.51 kg/m^2, which, compared to the BMI of CG (25.64 ± 3.68 kg/m^2), was statistically significant ($p < 0.05$). **Conclusion.** The FEC model demonstrated efficacy in lowering HbA1c levels and BMI, mitigating DM distress, and enhancing self-care capabilities among young and middle-aged patients with T2DM experiencing DM-related distress.

Key words:
adult; diabetes mellitus, type 2; family; outcome assessment, health care; stress, physiological.

Apstrakt

Uvod/Cilj. Dijabetes melitus (DM) predstavlja značajan i trajan zdravstveni problem koji zbog raznih komplikacija koje nastaju usled neadekvatne kontrole stanja, dovodi do invaliditeta i mortaliteta. Cilj rada bio je da se ispita uticaj multidisciplinarnog modela nege koji uključuje saradnju porodice (MNSP) na lečenje tipa 2DM (T2DM) kod mladih i sredovečnih bolesnika, uzimajući u obzir prisustvo DM distresa. **Metode.** Istraživanjem je obuhvaćeno 98 bolesnika uzrasta od 18 do 59 godina sa dijagnozom T2DM i koji su ispoljavali distres povezan sa DM . Bolesnici su primljeni na Odeljenje za endokrinologiju, Prve narodne bolnice Jingdžou, u provinciji Hubei, Kina, između februara i decembra 2023. godine. Korišćenjem metode tabele slučajnih brojeva, bolesnici su nasumično raspoređeni ili u

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interventnu grupu (IG) ili u kontrolnu grupu (KG), a u svakoj je bilo po 49 bolesnika. Dok su ispitanici obe grupe primili standardnu negu, ispitanici IG su dodatno dobili i MNSP. Procenjivani su i upoređivani nivoi glikoziliranog hemoglobina (HbA1c), *Diabetes Distress Scale* (DDS) skorovi, *Summary of Diabetes Self-care Activities* (SDSCA) skorovi i indeks telesne mase (ITM) između dve grupe, pre intervencije i tri meseca kasnije. **Rezultati.** Tri meseca posle intervencije, kod IG su utvrđeni niži nivoi HbA1c (6,02 ± 0,63 vs. 6,81 ± 0,85) i DDS skorovi (25,27 ± 2,70 vs. 34,24 ± 4,46), a viši SDSCA skorovi (30,69 ± 1,91 vs. 25,03 ± 2,13) u poređenju sa KG. Dodatno, ITM bolesnika u IG iznosio je 23,83 ± 2,51 kg/m², što je u odnosu na ITM bolesnika u KG (25,64 ± 3,68 kg/m²) bilo statistički značajno ($p < 0,05$). **Zaključak.** Multidisciplinarni MNSP je pokazao efikasnost u snižavanju nivoa HbA1c i ITM, ublažavanju distres tegoba povezanih sa DM i poboljšanju sposobnosti samopomoći kod mladih i sredovečnih bolesnika sa T2DM i prisutnim distres tegobama povezanim sa DM.

Ključne reči:
odrasle osobe; dijabetes melitus, tip 2; porodica; zdravstvena zaštita, procena ishoda; stres.

Introduction

Diabetes mellitus (DM) is a metabolic disorder characterized by persistent hyperglycemia resulting from a combination of genetic and environmental factors. It represents a prevalent and enduring health concern, marked by various complications arising from inadequate management and substantial rates of disability and mortality [1]. In the 21st century, DM has emerged as a prominent global health challenge [2, 3]. A recent survey conducted by the International Diabetes Federation (IDF) revealed that approximately 537 million adults (aged 20–79 years) worldwide experienced DM in 2021, with the prevalence expected to escalate to 643 million by 2030 and 783 million by 2045 [1]. The IDF Diabetes Atlas, 10th edition [4], reported that in 2021, nearly half of adults with DM were unaware of their condition, and a significant portion (46.2%) of global deaths attributed to DM occurred in individuals under the age of 60 [5].

China, in particular, has witnessed a surge in DM subjects, claiming the title of the country with the highest number of patients, reaching 141 million adults in 2021 and projected to rise to 174 million by 2045 [1]. Type 2 DM (T2DM) constitutes over 90% of all DM subjects [6]. The Guidelines for the Prevention and Treatment of Type 2 Diabetes Mellitus in China (2020 Edition), issued by the Chinese Diabetes Section of the Chinese Medical Association, reported a prevalence of 11.2% among adults aged 18 years and above [7]. Notably, the onset of T2DM is concentrated in adults aged 40–59 years, with 59% of subjects aged 18–59 years [8, 9]. The prevalence of DM is escalating rapidly among young and middle-aged (YMA) adults, and the age of onset is decreasing [10, 11].

Another study conducted in China highlighted that DM distress affects 42.50−77.23% of patients with DM, with a 90.82% prevalence in young patients with T2DM. Among them, 57.14% exhibited severe DM and 33.67% had moderate DM. Younger patients are particularly susceptible to DM-related psychological distress [12–14]. Despite being integral to families and society [15], YMA patients with T2DM receive less attention compared to their older counterparts [16]. This demographic faces lower awareness and treatment adherence levels due to work and educational commitments [17]. Furthermore, a lack of understanding about DM prevention and treatment may lead some middle-aged patients with T2DM to self-discontinue medication upon achieving satisfactory blood glucose (BG) control or due to personal preferences [18], resulting in disease progression. Although self-care is a common strategy for managing chronic diseases, YMA patients with T2DM exhibit poorer adherence to diet, medication, and exercise compared to their older counterparts [19–24].

Given the distinctive characteristics of YMA patients with T2DM, including their substantial numbers, elevated mortality rates, susceptibility to DM distress, low diagnosis rates, poor self-care practices, and limited attention, the aim of this study was to assess the health outcomes of this demographic through the implementation of a family-engaged multidisciplinary collaborative care (FEC) model.

Methods

Participants

This research study included patients aged between 18 and 59 years diagnosed with T2DM and experiencing DM distress. The patients participated in this study voluntarily and without any compensation. At the initial outpatient screening, all participants had poorly controlled glycated hemoglobin A1c (HbA1c) levels ≥ 6.5% and a Diabetes Distress Scale (DDS) score ≥ 3. Due to their condition, these patients agreed to be hospitalized. The study recruited both newly diagnosed patients and those with a previous diagnosis of T2DM who had consistently poorly controlled glycemic levels. The participants were hospitalized in the Department of Endocrinology at The First People's Hospital of Jingzhou in Jingzhou City, Hubei province, China, from February to December 2023. The sample size was determined using the formula for two sample means: $\mathbf{n_1 = n_2 2\left[\frac{(\mu_\alpha+\mu_\beta)}{\delta/\sigma}\right]^2 + \frac{1}{4}\mu_\alpha^2}$. Employing a two-sided test with a significance level of $\alpha = 0.05$ and $\beta = 0.1$ corresponded to u0.05/2 = 1.96 and u0.01 = 1.282, which aligns with previous reports [24], where δ was 8.4 and σ was 11.89. Consequently, each group required a minimum of 42 subjects, and given a dropout rate of 20%, 50 subjects were eventually included in each group. Random codes were generated using the random number table method, assigned to groups, and placed in sequentially marked black sealed envelopes. Patients who met the inclusion and exclusion criteria randomly selected envelopes, which, based

on the assigned number, administered them to the control group (CG) or the intervention group (IG) (50 subjects *per* group). One case from the IG was lost to follow-up during the intervention (the patient was referred to another hospital), and one case from the CG was lost to follow-up at the third month of follow-up (the patient refused the follow-up due to distance). Consequently, 49 patients in each group completed the study.

The inclusion criteria for patients were as follows: individuals diagnosed with T2DM as *per* the diagnostic criteria outlined in the Guidelines for the Prevention and Treatment of Type 2 Diabetes Mellitus in China (2020 Edition) [25], with the additional requirement of family involvement; individuals aged between 18 and 59 years [26]; individuals with a DDS score ≥ 3 [27]; individuals who willingly volunteered for participation in the study and provided informed consent by signing a consent form; individuals proficient in using WeChat.

Exclusion criteria for participants were as follows: individuals experiencing severe or acute complications of DM; individuals with severe primary diseases affecting the heart, brain, or kidneys; individuals with a history of psychiatric disorders or cognitive deficits, such as dementia; individuals facing language communication barriers.

The study received an approval from the Ethics Committee of the The First People's Hospital of Jingzhou (No. KY202348, from March 16, 2023). General information between the two groups was comparable with no statistically significant difference ($p > 0.05$) (Table 1).

Table 1

General information on the two groups of patients with type 2 diabetes mellitus

Parameter	Intervention group (n = 49)	Control group (n = 49)	t/χ^2	p
Sex			0.049[a]	0.825
male	34 (69.4)	35 (71.4)		
female	15 (30.6)	14 (28.6)		
Age (years)	44.18 ± 9.462	44.78 ± 7.531	0.135[b]	0.733
BMI			0.228[a]	1.000
< 18.5	2 (4.1)	2 (4.1)		
18.5~23.9	15 (30.6)	15 (30.6)		
24~27.9	18 (36.7)	17 (34.7)		
≥ 28	14 (28.6)	15 (30.6)		
Marital status			0.3462[b]	0.841
married	40 (81.6)	41 (83.7)		
single	7 (14.3)	7 (14.3)		
divorced	2 (4.1)	1 (2.0)		
Education level			0.460[a]	0.928
primary school and below	6 (12.2)	7 (14.3)		
junior high school	19 (38.8)	18 (36.7)		
senior high school and middle special school	8 (16.3)	10 (20.4)		
college degree or above	16 (32.7)	14 (28.6)		
Monthly income, RMB			0.313[a]	1.000
1,000~1,999	3 (6.1)	4 (8.2)		
2,000~2,999	5 (10.2)	5 (10.2)		
3,000~3,999	12 (24.5)	11 (22.4)		
≥ 4,000	29 (59.2)	29 (59.2)		
Medical payment methods			1.181[a]	0.881
out-of-pocket payment	0 (0)	1 (2.0)		
new rural cooperative medical system	17 (34.7)	16 (32.7)		
employee insurance	15 (30.6)	15 (30.6)		
commercial insurance	4 (8.2)	5 (10.2)		
medical insurance for residents	13 (26.5)	12 (24.5)		
Living style			1.054[a]	0.788
with children	7 (14.3)	7 (14.3)		
with spouse	36 (73.5)	38 (77.6)		
alone	3 (6.1)	3 (6.1)		
other	3 (6.1)	1 (2.0)		
Occupation			0.541[a]	0.991
farmer	10 (20.4)	11 (22.4)		
worker	12 (24.5)	10 (20.4)		
public servant/institution staff/formal work	14 (28.6)	15 (30.6)		
unemployed	4 (8.2)	4 (8.2)		
retired	3 (6.1)	2 (4.1)		
other	6 (12.2)	7 (14.3)		

BMI – body mass index; RMB – Renminbi (Chinese yuan).
Values are given as numbers (percentages) or mean ± standard deviation.
Note: a – χ^2 value; b – *t*-value.

Control group

Patients received standard care during their hospital stay and underwent regular follow-ups *via* telephone and WeChat after discharge. The specific interventions implemented are presented in Table 2 (see Supplementary Material for the full content of standard care).

Intervention group – Establishment of a collaborative care team

The collaborative care team comprised eight members: an endocrinologist, a psychologist, a therapist, a dietitian, an endocrinology nurse practitioner, two DM-specialized nurses, and a graduate nursing student. With the active participation of patients' family members, the multidisciplinary nursing team comprehensively evaluated patients, identified issues, set goals, and formulated personalized, holistic nursing plans. They determined the implementation methods, provided ongoing evaluations of the interventions, and offered discharge guidance and feedback. In contrast, patients in the CG received routine clinical nursing, which included regular monitoring of BG, medication guidance, and health education. The specific responsibilities of each team member are outlined in Table 3.

Table 2

Specific interventions for the control group

Care in the hospital			Care after discharge		
Day 1 from admission	Day 2 from admission to 1 day prior to discharge	Day of discharge	Telephone follow-up	WeChat follow-up	Regular follow-up
Instruction was imparted on self-care practices for T2DM, encompassing scientifically tailored dietary measures, judicious exercise routines, appropriate medication adherence, hypoglycemia prevention, and psychological counseling.	Specialist nurses provided personalized instructions based on the specific medical conditions of individual patients.	Specialized nurses delivered centralized instruction on blood glucose monitoring and insulin injection using instructional models within the education room. Subsequently, patients were invited to participate in the "TT WeChat group chat for young and middle-aged individuals" for ongoing follow-up.	The physical and mental statuses of the patients were comprehensively assessed, and they were instructed to adhere to the prescribed medication regimen, including responding to any inquiries posed by the medical professionals.	In the "WeChat group chat for young and middle-aged individuals with TT" information regarding TT was disseminated through textual content, images, videos, and audio files, accompanied by interactive question-and-answer sessions.	During the third month of the intervention, the patient underwent a telephonic notification for the purpose of the follow-up.

T2DM – type 2 diabetes mellitus; **TT – textual toolbar.**

Table 3

The duties of each member of the family-involved multidisciplinary collaborative care team

Members	Duties
Endocrinology nurse practitioner	Assume the role of team leader, supervise the development of diverse management programs, team training and assessment initiatives, and workflow processes. Conduct regular meetings to consolidate, rectify, and control the ongoing tasks.
Graduate nursing student	Design intervention programs, conduct statistical data analysis, and compose research papers. Formulate general information questionnaires, and select survey scales.
Specialist	Tasked with diagnosing, treating, and modifying treatment regimens based on evolving patient conditions. Offer guidance on pertinent medical information.
Specialist nurse	Offer guidance on professional nursing knowledge and skills. Engage in the distribution and retrieval of questionnaires. Contribute to the comprehensive care of the research program.
Psychologist	Evaluate the psychological condition of patients. Deliver expert psychological counseling.
Dietitian	Compute BMI for patients. Evaluate the nutritional status of patients and offer appropriate nutritional counseling.
Therapist	Evaluate the patient's motor status. Prescribe exercises tailored to patients with DM, based on their BMI.

Table 3 (continued)

Members	Duties
Families of patients	Oversee patients in the timely completion of daily tasks. Offer emotional support and encouragement to patients. Engage in standard care practices and health education for individuals with DM. Administer nutritional support to patients based on the guidance of a registered dietitian. Aid patients in mobility and exercise routines.
Patients	Fulfill the program requirements with both qualitative and quantitative excellence. Adhere diligently to prescribed dietary, exercise, and blood glucose monitoring regimens. Collaborate with the psychologist to actively engage in the conscious adjustment of psychological well-being.

BMI – body mass index; DM – diabetes mellitus.

Intervention group – Implementation program

IG patients were administered care through the FEC model rooted in conventional practices. The FEC approach encompassed comprehensive assessment, problem identification, goal establishment, formulation of an individualized and holistic care plan, selection of implementation methods, evaluation of effects, and provision of discharge guidance and feedback (Table 4).

Evaluation indicators

The assessment criteria encompassed DDS scores, Summary of Diabetes Self-care Activities (SDSCA) scores, body mass index (BMI) values, and HbA1c levels. Developed by Polonsky et al. [13] in 2005, the DDS comprises 17 entries across four dimensions, utilizing a 6-point Likert scale to assign scores ranging from 1 (no problem) to 6 (a serious problem) for each item, resulting in a total score of 102. A higher score indicates increased distress. The Cronbach's coefficient for the DDS was calculated as 0.951, affirming its internal consistency.

SDSCA, devised by Toobert et al. [28] for assessing self-care behavior in diabetes patients over the preceding seven days, has undergone several revisions. This scale encompasses 11 scoring entries categorized into five dimensions: diet (overall and specific), exercise, BG monitoring, foot care, and medication. Each entry is scored on a scale from 0 to 7, culminating in a total score ranging from 0 to 77 points. Li et al. [29] evaluated the reliability and validity of the SDSCA, reporting Cronbach's α of 0.84 for the entire scale and Cronbach's α values ranging from 0.71 to 0.93 for each subscale, confirming the scale's overall reliability and validity.

Statistical analysis

The data analysis was performed using SPSS 19.0 software. Descriptive statistics for qualitative data included counts and percentages, with between-group comparisons conducted using the Chi-squared test. Quantitative data exhibiting normal distribution are presented as mean ± standard deviation. Between-group comparisons for quantitative data were carried out using the two independent samples *t*-tests, while intragroup comparisons utilized the paired *t*-test. Statistical significance was determined at a threshold of $p < 0.05$.

Results

Analysis of HbA1c levels before and after the interventions conducted in two distinct groups

Following the interventions, statistically significant reductions in HbA1c levels were observed in both groups, with a $p < 0.001$. The *t*-values for the CG and IG were 0.422 ($p = 0.674$) and -5.249 ($p < 0.001$), respectively. These values suggest that the reduction in HbA1c levels was notably more pronounced in the IG post-intervention, highlighting the efficacy of the intervention in this group (Table 5).

Comparisons of DDS scores conducted before and after interventions in both study groups

Following the intervention in the IG, there was a substantial decrease in DDS scores ($p < 0.001$). In contrast, the CG exhibited a comparatively lower reduction in DDS scores after the intervention, with mean differences (MD) changing from 47.71 ± 3.33 to 34.24 ± 4.46 ($t = 16.951$, $p < 0.001$). The *t*-values for the IG and CG were -0.316 and -12.07 ($p < 0.001$), respectively. These results indicate significant enhancements in diabetes distress among patients in the IG, while the CG showed less improvement (Table 6).

Comparisons of pre- and post-intervention BMI variations within the two study groups

BMI disparity between the two groups reached statistical significance following the interventions ($p < 0.001$). Conversely, there was no notable alteration in the BMI of patients within the CG post-intervention ($p > 0.05$) (Table 7).

A comparative analysis of the SDSCA scores before and after interventions in the two groups

Following the interventions, a significant reduction in the SDSCA scores was observed in the IG ($p < 0.001$), while the CG experienced a relatively modest decrease (MD = 25.03 ± 2.13). The CG exhibited a modest enhancement in the SDSCA score (MD = 25.03 ± 2.13). In contrast, the IG demonstrated a substantial rise in the total SDSCA score, progressing from an initial MD of 15.07 ± 4.50 to a final MD of 30.69 ± 1.91 ($t = 13.864$, $p < 0.001$). These findings indicate a notable enhancement in patients' self-care ability within the IG (Table 8).

Table 4

Specific interventions for the intervention group

Usual care in the hospital			Care after discharge		
Days 1−2 from admission	Day 3 from admission to 1 day prior to discharge	Day of discharge	Telephone follow-up	WeChat follow-up	Regular follow-up
Apart from standard care, the following elements are incorporated: Extend invitations to patients and their families to participate in the "Collaborative Care Communication Group," providing an introduction to the components of the family-engaged multidisciplinary collaborative care model. Thoroughly evaluate patients' conditions and formulate individualized care plans for each patient. Collaboratively devise a professional intervention plan for patients' conditions involving all team members.	Within the interdisciplinary team, each professional undertakes individualized interventions to address patient issues, employing a hybrid approach that integrates both online and offline methodologies. Offline nurses instruct patients and their families on the preparation of a DM diet in accordance with the dietitian's plan, utilizing the "Boohee Dietitian" app. The therapist educates patients on appropriate exercise regimens, while the psychologist conducts bi-weekly psychological interventions. Families play a vital role in supporting and overseeing patients to ensure timely completion of tasks. Additionally, the online "Collaborative Care Communication Group" sporadically disseminates videos featuring psychological interventions and healthy exercises for individuals with T2DM. The specialized nurse transmits the intervention outcomes of patients to the "Collaborative Care Communication Group," where team members formulate a new plan informed by the outcomes of their deliberations.	The specialist nurses provide individualized guidance on monitoring blood glucose levels and administering insulin through one-on-one sessions. Patients' families undergo assessment through a test paper, and patients scoring > 80 are deemed satisfactory. Individuals failing to meet the examination criteria receive personalized guidance focused on areas of insufficient comprehension and mastery, incorporating the examination results. Subsequently, they undergo reevaluation until achieving qualification.	Understand the physical and mental status of patients, advise them to adhere to prescribed medication regimens as directed by physicians, and address any inquiries they may have.	Enhance the understanding of DM by participating in the "Collaborative Care Communication Group," addressing inquiries and resolving uncertainties related to health knowledge. Gather the findings from the self-monitoring blood glucose card weekly *via* the "Collaborative Care Communication Group," ensuring submission by 9:00 p.m. each Sunday. Foster patient engagement by encouraging the sharing of their results and experiences within the group.	During the third month of the intervention, patients were contacted *via* phone for a scheduled follow-up, and a lecture took place in the endocrinology department's demonstration classroom. The lecture focused on elucidating the significance and appropriate methodologies for engaging T2DM patients and their families in the effective management of DM.

T2DM – type 2 diabetes mellitus (DM).

Table 5

Comparisons of HbA1c levels before and after interventions in the intervention and control groups

Parameter	HbA1c (%)		*t*	*p*
	before interventions	three months after interventions		
Intervention group (n = 49)	11.00 ± 1.92	6.02 ± 0.63	17.24	< 0.001
Control group (n = 49)	10.84 ± 1.79	6.81 ± 0.85	14.24	< 0.001
t	0.422	-5.249		
p	0.674	< 0.001		

HbA1c – glycated hemoglobin.
Values are given as mean ± standard deviation.

Table 6

Comparisons of DDS scores before and after interventions in the two groups

Parameter	DDS		*t*	*p*
	before interventions	three months after interventions		
Intervention group (n = 49)	47.51 ± 3.06	25.27 ± 2.70	38.195	< 0.001
Control group (n = 49)	47.71 ± 3.33	34.24 ± 4.46	16.951	< 0.001
t	-0.316	-12.07		
p	0.753	< 0.001		

DDS – diabetes distress scale.
Values are given as mean ± standard deviation.

Table 7

Comparisons of BMI before and after interventions in the two groups

Parameter	BMI (kg/m^2)		*t*	*p*
	before interventions	three months after interventions		
Intervention group (n = 49)	26.13 ± 4.12	23.83 ± 2.51	3.346	0.001
Control group (n = 49)	25.81 ± 3.97	25.64 ± 3.68	0.213	0.832
t	0.397	-2.86		
p	0.692	0.005		

BMI – body mass index.
Values are given as mean ± standard deviation.

Table 8

Comparisons of SDSCA scores before and after interventions in the two groups

Parameter	IG (n = 49)	CG (n = 49)	*t/z*	*p*
Diet management				
before	2.36 ± 0.70	5.78 ± 0.81	0.116	0.908
after	2.35 ± 0.70	4.84 ± 0.91	5.398	<0.001
t/z	0.212	-15.209		
p	< 0.001	< 0.001		
Exercise management				
before	2.45 ± 0.83	2.40 ± 0.84	0.302	0.763
after	5.64 ± 0.83	3.80 ± 0.99	9.99	< 0.001
t/z	0.870	-7.512		
p	< 0.001	< 0.001		
Blood glucose monitoring				
before	3 (1.4)	3 (2.4)	-0.87	0.930
after	6 (5.7)	4 (4.5)	-5.169	< 0.001
t/z	-7.754	-5.533		
p	< 0.001	< 0.001		
Foot care				
before	1 (1.4)	2 (1.4)	-0.080	0.936
after	7 (6.7)	5 (4.6)	-5.548	< 0.001
t/z	-7.905	-6.509		
p	< 0.001	< 0.001		

Table 8 (continued)

Parameter	IG (n = 49)	CG (n = 49)	*t/z*	*p*
Medication				
before	7 (1.7)	7 (3.7)	-0.211	0.833
after	7 (7.7)	7 (6.7)	-3.279	< 0.001
t/z	-4.644	-3.094		
p	< 0.001	0.002		
Total score				
before	15.07 ± 4.50	15.01 ± 3.94	0.69	0.945
after	30.69 ± 1.91	25.03 ± 2.13	13.864	< 0.001
t/z	-22.369	-15.661		
p	< 0.001	< 0.001		

SDSCA – Summary of Diabetes Self-care Activities; IG – intervention group; CG – control group.
Values are given as mean ± standard deviation or numbers (percentages).

Discussion

T2DM is a lifelong metabolic disorder characterized by persistent hyperglycemia, with no definitive cure [30], necessitating continuous and lifelong therapeutic interventions. Managing this condition in China, a country facing a significant diabetes burden, is particularly challenging. These challenges are compounded by the diverse family dynamics of patients, many of whom belong to families with varying socioeconomic statuses, responsibilities for both elderly care and child-rearing, and differing educational backgrounds. In the IG, the educational levels of patients and their family members ranged from primary school to advanced degrees, making health education a complex and time-consuming task. Given these differences, ensuring that patients and their families understand and apply the necessary health education further complicates diabetes management and increases the overall cost of care.

The imperative lies in identifying novel, effective, and feasible strategies to enhance diabetes care and therapeutic outcomes. The FEC model addresses nursing assessment issues, employing information technology for case-specific analysis. This model strategically directs healthcare resources toward bolstering comprehensive physical and psychological care, ensuring continuous support from admission to discharge for YMA patients with T2DM experiencing DM distress.

Through the coordinated efforts, distinct roles, collective wisdom, and synergistic strengths of multidisciplinary medical staff and family involvement, this model establishes a comprehensive and standardized long-term care mechanism for hospitals catering to the needs of YMA patients with T2DM with DM distress. Our study revealed a significant disparity in HbA1c levels, DDS scores, BMI values, and SDSCA scores between the CG and IG three months post-intervention ($p < 0.001$), aligning with the findings of Abdulrhim et al. [3], highlighting the positive impact of the FCE model on patient satisfaction, quality of life (QoL), and health status.

Furthermore, additional research indicated a correlation between DM distress and poor DM control [31]. Consequently, the FEC model emerges as clinically crucial in the comprehensive care of YMA patients with T2DM experiencing DM distress.

As reported [32], DM distress is higher and more common in younger patients than in older patients, which may be related to the family, social, and work responsibilities of younger patients. Owens-Gary et al. [33] stated that DM distress increased the risk of death, poor disease management, DM-related complications, and poor QoL and that FEC could improve DM distress in patients with DM. Likewise, our findings indicated significantly lower DDS scores in the FEC model group compared to the CG ($p < 0.001$). This underscores the heightened effectiveness of the FEC model in alleviating the condition of YMA patients with T2DM experiencing DM distress. It has been reported that family support and involvement in adult DM care elevate the motivation and self-efficacy of patients in managing DM [34]. Jiang et al. [35] found through a Mate analysis that family-engaged education was beneficial to BG control in T2DM patients. Meanwhile, another study indicated that disease management with family involvement ameliorated the anxiety and depression of patients and enhanced their QoL [36]. Accordingly, it is vital to actively encourage the patients' families to participate in managing chronic diseases and to work together with patients and medical staff to combat the disease.

In recent years, there has been a notable increase in the prevalence of DM, coinciding with a rise in the incidence of obesity and overweight conditions. The risk of developing T2DM shows a twofold increase with a weight gain of 5–8 kg and a fourfold increase with a weight gain of 20 kg or more [37]. Obesity has emerged as a significant factor contributing to the onset of T2DM [38]. According to survey data, 41.0% of patients with DM in China are overweight, and 24.3% are classified as obese [39].

Fisher et al. [40] highlighted that rates of DM distress tend to be higher in specific demographic groups, including women, relatively young adults, insulin-using patients, those with poorer BG control, individuals with a high BMI, those with a prolonged duration of DM, and those with significant comorbidities associated with DM.

Consequently, it can be inferred that the effective management of BMI is crucial for individuals with DM. In our study, the FEC model demonstrated a more significant im-

pact on BMI compared to the conventional care approach ($p < 0.001$). Conversely, BMI in the CG exhibited no statistically significant difference before and after the intervention ($p > 0.05$).

As the prevalence of DM continues to rise, self-care has emerged as a pivotal element in DM management [41]. In this study, we focused on a self-care intervention tailored for YMA patients with T2DM experiencing DM distress. Following a comprehensive regimen encompassing dietary adjustments, exercise, medication adherence, BG monitoring, and psychological support, patients exhibited a significant elevation in their SDSCA scores ($p < 0.001$). The multifaceted nature of the intervention underscores the intricate landscape of DM management, intensifying the challenges associated with self-care.

In concordance with our findings, the study by Nielsen et al. [42] demonstrated a greater impact of interventions on T2DM compared to type 1 DM (T1DM). This was associated with a reduction in HbA1C levels among patients with T2DM, aligning with outcomes observed in our study. The variance in intervention outcomes suggests that the effectiveness of self-care strategies may differ between T2DM and T1DM populations, emphasizing the importance of tailored approaches.

The crucial link between self-care and QoL in patients with DM is well-established [43], underscoring the necessity of maintaining sustained self-care practices. The results of our study affirm that educating patients on proper dietary habits and incorporating exercise into their routines can positively influence unhealthy lifestyle patterns, thereby enhancing their QoL and exerting favorable control over their condition. Consequently, healthcare professionals bear the responsibility of imparting relevant information and skills to empower patients in their self-care journey.

In this study, potential biases may exist due to the varied family backgrounds and socioeconomic statuses of the patients, as well as differences in self-care experience between newly diagnosed and previously diagnosed DM patients. In future research, efforts will be made to better control these potential influencing factors by targeting larger and more diverse samples and conducting long-term follow-ups to assess the durability of the intervention's benefits. However, a notable limitation of this study is the relatively brief intervention duration (three months). Consequently, future research endeavors should consider extending the intervention period to provide more robust insights.

Conclusion

The family-engaged multidisciplinary collaborative care model proves advantageous for comprehensive care and both the physical and mental development of young and middle-aged patients with type 2 diabetes mellitus experiencing diabetes distress. Furthermore, it facilitates the implementation of ongoing care, enhances the quality of care, optimizes healthcare resources, and improves the self-care capabilities of patients.

REFERENCES

1. *Sun H, Saeedi P, Karuranga S, Pinkepank M, Ogurtsova K, Duncan BB*, et al. IDF Diabetes Atlas: Global, regional, and country-level diabetes prevalence estimates for 2021 and projections for 2045. Diabetes Res Clin Pract 2022; 183: 109119. Erratum in: Diabetes Res Clin Pract 2023; 204: 110945.
2. *Kumah E, Afriyie EK, Abuosi AA, Ankomah SE, Fusheini A, Otchere G.* Influence of the Model of Care on the Outcomes of Diabetes Self-Management Education Program: A Scoping Review. J Diabetes Res 2021; 2021: 2969243.
3. *Abdulrhim S, Sankaralingam S, Ibrahim MIM, Diab MI, Hussain MAM, Al Raey H,* et al. Collaborative care model for diabetes in primary care settings in Qatar: a qualitative exploration among healthcare professionals and patients who experienced the service. BMC Health Serv Res 2021; 21(1): 192.
4. *International Diabetes Federation.* IDF Diabetes Alas. 10th ed. Brussels: International Diabetes Federation; 2021. p. 135.
5. *Saeedi P, Salpea P, Karuranga S, Petersohn I, Malanda B, Gregg EW,* et al. Mortality attributable to diabetes in 20-79 years old adults, 2019 estimates: Results from the International Diabetes Federation Diabetes Atlas, 9th edition. Diabetes Res Clin Pract 2020; 162: 108086.
6. *Weng J, Ji L, Jia W, Lu J, Zhou Z, Zou D,* et al. Standards of care for type 2 diabetes in China. Diabetes Metab Res Rev 2016; 32(5): 442−58.
7. *Chinese Medical Association Diabetes Branch.* Guidelines for the prevention and treatment of type 2 diabetes mellitus (2020 edition). Chin J Diabetes 2021; 13(04): 315−409.
8. *Cho NH, Shaw JE, Karuranga S, Huang Y, da Rocha Fernandes JD, Ohlrogge AW,* et al. IDF Diabetes Atlas: Global estimates of diabetes prevalence for 2017 and projections for 2045. Diabetes Res Clin Pract 2018; 138: 271−81.
9. *He YW, Ge YH, Li XY, Ye ZF, Kong LP.* Self-care activities and their influencing factors among young and middle-aged patients with type 2 diabetes mellitus. Chin J Prev Med 2022; 34(3): 258−62.
10. *Wang J, Xiao L, Yu C, Wang P, Li Z.* The current status and influencing factors of post-traumatic growth in middle-aged and young patients with type 2 diabetes. Chin J Pract Nurs 2022; 38(21): 1663−8.
11. *Ogurtsova K, da Rocha Fernandes JD, Huang Y, Linnenkamp U, Guariguata L, Cho NH,* et al. IDF Diabetes Atlas: Global estimates for the prevalence of diabetes for 2015 and 2040. Diabetes Res Clin Pract 2017; 128: 40−50.
12. *Hu Y, Li L, Zhang J.* Diabetes Distress in Young Adults with Type 2 Diabetes: A Cross-Sectional Survey in China. J Diabetes Res 2020; 2020: 4814378.
13. *Polonsky WH, Fisher L, Earles J, Dudl RJ, Lees J, Mullan J,* et al. Assessing psychosocial distress in diabetes: development of the diabetes distress scale. Diabetes Care 2005; 28(3): 626−31.
14. *Joensen LE, Almdal TP, Willaing I.* Associations between patient characteristics, social relations, diabetes management, quality of life, glycaemic control and emotional burden in type 1 diabetes. Prim Care Diabetes 2016; 10(1): 41−50.
15. *Li X, Liu S, Wu Q.* Disease adaptability and related factors in young and middle-aged patients with type 2 diabetes. Chin J Ment Health 2021; 35(8): 628−30.
16. *Li M.* Intervention of young and middle-aged patients with type 2 diabetes based on cross-theoretical model [M.S. Thesis]. China: North Sichuan Medical College; 2018. (Chinese)

17. *Zhou G, Xu R, Bian X, Zheng W*. Development of health literacy scale for middle-aged and young T2DM patients and its validation. Chin J Nurs 2023; 30(14): 73−8.
18. *Li XH, Cheng L, Long JH*. Research progress of self-management evaluation tools for diabetic patients. Chin J Nurs Sci 2015; 30(21): 97−100. (Chinese)
19. *Thewjitcharoen Y, Chotwanvirat P, Jantawan A, Siwasaranond N, Saetung S, Nimitphong H*, et al. Evaluation of Dietary Intakes and Nutritional Knowledge in Thai Patients with Type 2 Diabetes Mellitus. J Diabetes Res 2018; 2018: 9152910.
20. *Yang X, Yuan L, Guo X, Lou Q, Zhao F, Shen L*, et al. Analysis and countermeasures of dietary self-management in Chinese patients with type 2 diabetes. Chin J Diabetes 2013; 5(11): 666−9.
21. *Weinstock RS, Trief PM, Burke BK, Wen H, Liu X, Kalichman S*, et al. Antihypertensive and Lipid-Lowering Medication Adherence in Young Adults With Youth-Onset Type 2 Diabetes. JAMA Netw Open 2023; 6(10): e2336964.
22. *Zhong DY, Li LH, Li H, Ma R, Deng YH*. Study on the mechanism and molecular docking verification of buyang huanwu decoction in treating diabetic foot. World J Tradit Chin Med 2023; 9: 178−90.
23. *Yao WY, Han MG, De Vito G, Fang H, Xia Q, Chen Y*, et al. Physical Activity and Glycemic Control Status in Chinese Patients with Type 2 Diabetes: A Secondary Analysis of a Randomized Controlled Trial. Int J Environ Res Public Health 2021; 18(8): 4292.
24. *Kang S, Zhao XJ, Gao JX, Tian SZ, Qiu JJ, Wang ZN*, et al. Effect of the self-management support based on mobile APP on the self-efficacy and psychological distress of young and middle-aged diabetic patients. J Bengbu Med Uni 2021; 46(1): 115−9.
25. *Chinese Diabetes Society*. Guideline for the prevention and treatment of Type 2 Diabetes Mellitus in China (2020 edition) (Part 1). Chin J Pract Internal Med 2021; 41(8): 668−95.
26. *World Health Organization*. Health topics: Diabetes [Internet]. 2019; [cited on 2022 Dec 11]. Available from: http://www.who.int/topics/diabetes-mellitus/en
27. *Yang Q, Liu XQ*. Evaluation of reliability and validity of Chinese version of diabetes pain scale. Chin J Nurs 2010; 17(17): 8−10.
28. *Toobert DJ, Hampson SE, Glasgow RE*. The summary of diabetes self-care activities measure: results from 7 studies and a revised scale. Diabetes Care 2000; 23(7): 943−50.
29. *Li J, Jiang Y, Peng H, Liao S, Liu M, Lu Y*. A meta-analysis of a cross-sectional studies of psychological distress with Type 2 diabetes patients in China. Chin J Ment Health 2023; 37(4): 312−7.
30. *Zeng R*. Improvement of the self-management behavior scale for patients with type 2 diabetes and its reliability and validity. Chin J Mod Hops 2011; 11(3): 148−50.
31. *Silveira MSVM, Bovi TG, Pavin EJ*. Relatively young T1D adults using fixed doses of insulin have higher diabetes distress levels in a sample of patients from a Brazilian tertiary hospital. Diabetol Metab Syndr 2019; 11: 104.
32. *Jenkinson E, Knoop I, Hudson JL, Moss-Morris R, Hackett RA*. The effectiveness of cognitive behavioural therapy and third-wave cognitive behavioural interventions on diabetes-related distress: A systematic review and meta-analysis. Diabet Med 2022; 39(11): e14948.
33. *Owens-Gary MD, Zhang X, Jawanda S, Bullard KM, Allweiss P, Smith BD*. The Importance of Addressing Depression and Diabetes Distress in Adults with Type 2 Diabetes. J Gen Intern Med 2019; 34(2): 320−4.
34. *Rosland AM, Piette JD, Trivedi R, Lee A, Stoll S, Youk AO*, et al. Effectiveness of a Health Coaching Intervention for Patient-Family Dyads to Improve Outcomes Among Adults With Diabetes: A Randomized Clinical Trial. JAMA Netw Open 2022; 5(11): e2237960.
35. *Jiang Y, Liu AN, Zhang Q, Zheng HY, Sun XH, Pan ZW*. Meta-analysis of the effects of family participatory education on metabolic indexes in type 2 diabetes patients. Chin J Mod Med 2019; 29(13): 56−63.
36. *Huang CH, Cai YJ, Xu S, Guo YF, Liu YZ, Shi TY*. Research progress on family involvement in exercise intervention of cancer patients. Chin Nurs Manag 2023; 23(7): 1109−12.
37. *Wang Y, Han SF, Zhu RF, Cao Y, Feng YQ, Cheng JX*. An empirical study of dietary intervention in overweight/obese type 2 diabetes patients with family nurse dietary therapy program. Chin J Nurs Res 2023; 37(12): 2085−91. (Chinese)
38. *Wang JL, Jia HY, Feng ZH, Li L*. Research progress of Chinese medicine regulating AMPK signaling pathway for prevention and treatment of obesity type 2 diabetes mellitus. Chin J Exp Formulae 2023; 29(21): 264−73. (Chinese)
39. *Ma HH, Liang ZJ, Zhu JM, Wan M*. Research progress of dietary intervention in remission of type 2 diabetes mellitus in overweight and obesity. Chin J Nurs Res 2023; 37(6): 1041−6.
40. *Fisher L, Polonsky WH, Hessler D*. Addressing diabetes distress in clinical care: a practical guide. Diabet Med 2019; 36(7): 803−12.
41. *Cho MK, Kim MY*. Self-Management Nursing Intervention for Controlling Glucose among Diabetes: A Systematic Review and Meta-Analysis. Int J Environ Res Public Health 2021; 18(23): 12750.
42. *Nielsen PJ, Hafdahl AR, Conn VS, Lemaster JW, Brown SA*. Meta-analysis of the effect of exercise interventions on fitness outcomes among adults with type 1 and type 2 diabetes. Diabetes Res Clin Pract 2006; 74(2): 111−20.
43. *Sikalidis AK, Karaboga EP*. Healthy diet and self-care activities adherence improved life-quality and Type 2 Diabetes Mellitus management in Turkish adults. Gazzetta Med Ital 2020; 170(9): 528−37.

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Supplement

Standard care for the control group

Stage 1 – In hospital

Day 1 of admission

1. Determine patients based on inclusion and exclusion criteria.

2. Collect baseline data, including general information, the Diabetes Distress Scale, and the Self-Management Behavior Scale. Measure body weight and height, calculate BMI, and perform blood tests for glycated hemoglobin (HbA1c) levels.

3. Conduct type 2 diabetes mellitus (T2DM) self-management knowledge guidance:

Emotional guidance:

- Communicate with the patient to understand their current emotional state and provide methods for emotional relief, such as talking, listening to music, or jogging.

Exercise guidance:

- Moderate aerobic exercise – aerobic exercises (walking, swimming, cycling) can improve body sensitivity and help lower blood sugar levels.
- Patients can exercise three times a week, for about 30 minutes.
- Regular exercise timing – maintaining a consistent daily exercise schedule can help stabilize blood sugar levels and improve overall health.
- The best time for exercise is one hour after meals. Carry candy, biscuits, and identification cards while exercising.
- The appropriate exercise intensity is determined by the formula: post-exercise heart rate (beats/min) = 170 - age.

Scientific dietary guidance:

- Control the intake of staple foods like rice and wheat carbohydrates.
- Choose low-glycemic index (GI) foods – low-GI foods cause a slower increase in blood sugar levels, which helps maintain stable blood sugar control. For instance, choose whole wheat bread or brown rice instead of high-GI foods.
- Balanced diet – reasonably combine staple foods, proteins, and vegetables to ensure adequate nutrition in each meal and control overall intake to maintain an appropriate weight.
- Recommend cooking methods such as stewing, steaming, or boiling.
- Avoid high-fat foods – limit excessive intake of saturated and trans fats, and choose foods rich in healthy fats, like vegetable oils and fish.
- Control alcohol consumption – alcohol affects blood sugar regulation, so diabetic patients should limit alcohol intake and avoid drinking on an empty stomach.

Medication guidance:

- Eemphasize the importance of following the doctor's prescription and explain diabetes-related medications' effects, usage methods, and side effects.

Prevention, identification, and management of hypoglycemia:

- Diabetic patients should use medication according to the doctor's recommendations, undergo regular check-ups, and maintain good communication with their doctor to ensure proper disease management and control.

- Regularly monitor blood sugar levels and carry sugary foods (candy, snacks) when going out. Seek immediate medical attention at a nearby hospital if hypoglycemia occurs.

Implementation method: Data collection at the nurse's station, bedside education.

Time: Data collection takes 10–20 minutes, and knowledge guidance takes 30–45 minutes.

Implementers: Two specialized diabetes mellitus (DM) nurses and one graduate student conduct data collection and knowledge guidance.

From the second day of admission to the first day before discharge

Content: Improve relevant auxiliary examinations and provide corresponding guidance based on the results.

Implementation method: Bedside one-on-one.

Implementers: Two specialized DM nurses.

Discharge day

1. Invite discharged patients to join the WeChat group for "Young and Middle-aged DM Patient-Doctor Communication" for follow-up and health education after discharge.
2. Blood glucose monitoring and insulin injection guidance.

Blood glucose monitoring:

- Use a fixed blood glucose meter to monitor blood glucose levels before each meal and before bedtime daily to adjust diet and insulin dosage.
- Monitor blood glucose levels if symptoms such as dizziness or cold sweat occur to determine if hypoglycemia is happening.

Insulin injection:

- Ensure the injection site is clean and avoid injecting in the same spot consecutively.
- Preferred injection sites: the abdomen—2 inches around the navel to the iliac crest (avoiding the navel area); the buttocks—muscles in the outer upper quadrant of the buttocks; the outer front thigh; the outer arm area.
- Regularly change needles to prevent cross-infection and pause for 10 seconds after injection.
- Follow medical advice on medication and obtain the doctor's consent before altering the dosage.

Time: 20–30 minutes.

Implementation method: One-on-one bedside guidance.

Implementers: Two DM specialist nurses.

Stage 2 – Outside the hospital

Telephone follow-up

Content: Monitor the patient's blood sugar control at home, see whether any complications have occurred, and address any questions or concerns.

Frequency: Once a week, 10–20 minutes each time.

Implementers: One graduate student and two DM specialist nurses.

WeChat follow-up

Content: Through the "Middle-aged and Young DM WeChat Group," disseminate health knowledge in text, image, video, or audio form, address questions and concerns, and inform about the follow-up appointment times.

Frequency: Once a week, 10–20 minutes each time.

Implementers: One graduate student and two DM specialist nurses.

Regular outpatient visits

Content: One week after discharge and in the third month after the intervention, call the patient to inform them about a follow-up appointment at the outpatient clinic. In the third month after the intervention, collect the patient's Diabetes Distress Scale, Self-Management Behavior Scale, Patient Care Satisfaction Survey, and body mass index (BMI) data at the clinic. Additionally, perform blood sampling and monitor BMI.

Implementation method: One week after discharge, visit the hospital to collect random blood glucose samples and adjust the treatment plan based on the patient's condition. In the third month after the intervention, collect data one-on-one at the outpatient clinic, perform blood sampling, and recheck HbA1c. Adjust the treatment plan accordingly.

Duration: Data collection takes 10–20 minutes, and blood sampling takes 2–5 minutes.

Implementers: One graduate student and two DM specialist nurses.

ORIGINAL ARTICLE

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Risk factors for ischemic osteonecrosis of the femoral head after internal fixation with multiple cannulated screws for Pauwels type III femoral neck fracture

Faktori rizika od ishemijske osteonekroze glave femura posle unutrašnje fiksacije višestrukim kanuliranim zavrtnjima za frakturu vrata femura tipa III po Pauelsu

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Abstract

Background/Aim. Numerous factors lead to hip fractures that are common and often devastating in the geriatric population. The aim of the study was to determine the risk factors for ischemic osteonecrosis of the femoral head (ONFH) in patients after internal fixation with multiple cannulated screws for Pauwels type III femoral neck fracture (FNF). **Methods.** A total of 212 patients with Pauwels type III FNF, who underwent internal fixation with multiple cannulated screws from January 2019 to September 2022 and received one-year follow-up after surgery, were selected. Based on their clinical data and the incidence of ONFH, they were divided into two groups: the group with ischemic ONFH occurrence (n = 30) and the ischemic ONFH non-occurrence group (n = 182). Logistic regression analysis was conducted to analyze the risk factors for postoperative ischemic ONFH. **Results.** Age, body mass index (BMI), time from injury to surgery, partial weight bearing (PWB) time after surgery, and preoperative diabetes mellitus (DM) were significantly different between the ischemic ONFH occurrence and ischemic ONFH non-occurrence groups ($p < 0.05$), while other clinical data showed no significant differences ($p > 0.05$). The results of logistic regression analysis revealed that age (> 55 years), PWB time after surgery (≤ 3 months), BMI (≥ 24 kg/m^2), time from injury to surgery (> 2 days), and preoperative DM were independent risk factors for postoperative ischemic ONFH ($p < 0.05$). **Conclusion.** The incidence of postoperative ischemic ONFH can be prevented by effectively controlling the following factors: age, BMI, PWB after surgery, and preoperative DM.

Key words:
bone screws; femoral neck fractures; femur head; fracture fixation, internal; orthopedic procedures; osteonecrosis; risk factors.

Apstrakt

Uvod/Cilj. Mnogobrojni faktori dovode do preloma kuka, koji je u gerijatrijskoj populaciji učestao i često razoran. Cilj rada bio je da se utvrde faktori rizika od ishemijske osteonekroze glave femura (ONGF) posle unutrašnje fiksacije višestrukim kanuliranim zavrtnjima kod bolesnika sa frakturom vrata femura (FVF) tipa III po Pauelsu. **Metode.** Izdvojeno je ukupno 212 bolesnika sa FVF tipa III po Pauelsu, koji su bili podvrgnuti unutrašnjoj fiksaciji pomoću kanuliranih zavrtnja od januara 2019. do septembra 2022. godine i praćeni tokom jedne godine. Na osnovu kliničkih podataka i uočene incidencije ishemijske ONGF, bolesnici su raspoređeni u dve grupe: grupu sa pojavom ishemijske ONGF (n = 30) i grupu bez pojave ishemijske ONGF (n = 182). Za analizu faktora rizika od postoperativne ishemijske ONGF korišćena je logistička regresiona analiza. **Rezultati.** Životno doba, indeks telesne mase (ITM), vremenski interval od povrede do operacije, period parcijalnog opterećenja težinom tela (POT) posle operacije i preoperativni dijabetes melitus (DM) značajno su se razlikovali između grupe sa pojavom ishemijske ONGF i one bez pojave ishemijske ONGF ($p < 0,05$), dok nije bilo značajne razlike u ostalim kliničkim podacima ($p > 0,05$). Rezultati logističke regresione analize ukazuju na to da su životno doba (> 55 godina), period POT posle operacije (≤ 3 meseca), ITM (≥ 24 kg/m^2), vremenski interval od povrede do operacije (> 2 dana) i preoperativni DM bili nezavisni faktori rizika od nastanka posteoperativne ishemijske ONGF ($p < 0,05$). **Zaključak.** Incidencija

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postoperativne ishemijske ONGF može biti snižena efikasnom kontrolom sledećih faktora rizika: životnog doba, vremena od povrede do operacije, perioda POT posle operacije i prisustva preoperativnog DM.

Ključne reči:
zavrtnji za kost; femur, prelomi vrata; femur, glava; prelomi, fiksacija, unutrašnja; ortopedske procedure; osteonekroza; faktori rizika.

Introduction

Femoral neck (FN) fracture (FNF) is a common hip fracture found between the femoral head and the base of the FN, also known as intracapsular hip fracture, since the fracture line lies in joint [1]. Epidemiological data indicate that FNF accounts for about 3.66% of all fractures. In the young population, it is mainly attributed to high-energy forces, whereas in the elderly, it is mostly associated with osteoporosis, same-level falls, and other low-energy forces [2, 3]. Pauwels type III (PTIII) FNF refers to fractures with an angle of ≥ 50° between the distal fracture line and the horizontal line, having a large vertical shear force and many complications after surgery. The surgical treatment of FNF includes internal fixation (IF) and artificial hip replacement. Currently, IF with multiple cannulated screws (CSs) is often used to treat PTIII FNF, which can provide relatively satisfactory outcomes. However, such treatment leads to a fairly high incidence rate of long-term complications and disability rate due to the anatomical and blood supply characteristics of the femoral head and neck. As a severe complication, ischemic osteonecrosis of the femoral head (ONFH) has an incidence rate of about 8–40% after IF with multiple CSs for PTIII FNF [3]. In addition, it has been proven that ischemic ONFH is difficult to treat, resulting in varying degrees of femoral head collapse accompanied by impaired walking and joint movement, which seriously affects the quality of life [4]. Therefore, ischemic ONFH is currently a major problem in PTIII FNF treatment. Defining the risk factors for ischemic ONFH following multiple IF with multiple CSs for PTIII FNF is of great significance in preventing such a complication and decreasing its incidence.

Methods

This retrospective analysis included 212 patients with PTIII FNF. IF was performed on all patients with multiple CSs at our hospital from January 2019 to September 2022, and they were followed up for one year after surgery. This study was approved by the Jiangsu Integrated Chinese and Western Medicine Hospital on January 4, 2019.

The patients were divided into two groups based on the incidence of ischemic ONFH: an ONFH occurrence group (n = 30) and an ONFH non-occurrence group (n = 182).

The inclusion criteria were set as follows: age > 18 years, relevant criteria of FNF in the "Clinical Guidelines for Diagnosis and Treatment: Orthopedics" [5], IF with multiple CSs, unilateral FNF, and fracture time < 3 weeks. The exclusion criteria were incomplete clinical data, associated fractures, or severe diseases.

Patient data included gender, mechanism of injury, injured side, conducted or not conducted traction, duration of surgery, age, body mass index (BMI), time from injury to surgery (TIS), partial weight bearing (PWB) time after surgery, preoperative diabetes mellitus (DM), and preoperative hypertension.

Statistical analysis

A Statistical Package for the Social Sciences (SPSS) software version 26.0 was adopted for data processing and analysis. Descriptive data were analyzed, and the Chi-squared χ^2 test was used to compare the occurrence and non-occurrence groups. Logistic regression was performed to find the risk factors for postoperative ischemic ONFH in patients with PTIII FNF. The difference was considered statistically significant if $p < 0.05$.

Results

Thirty (14.15%) patients suffered from ischemic ONFH out of the 212 patients with PTIII FNF who performed IF with multiple CSs.

The comparisons of clinical data between the two groups shown indicated no statistically significant differences regarding gender, mechanism of injury, injured side, traction conducted performance, duration of surgery, and preoperative hypertension presence ($p > 0.05$). The numbers of patients older than 55 years, with BMI ≥ 24 kg/m^2, TIS > 2 days, PWB time after surgery ≤ 3 months, and preoperative DM showed a statistically significant difference between the ischemic ONFH occurrence and ischemic ONFH non-occurrence groups ($p < 0.05$) (Table 1).

Logistic regression analysis was performed using the incidence of ischemic ONFH after IF with multiple CSs for PTIII FNF as the dependent variable (ONFH occurrence group = 1, ONFH non-occurrence group = 0) and the factors with statistically significant differences (age, BMI, TIS, PWB time after surgery, and preoperative DM) as the independent variables (Table 2). It was found that age > 55 years, BMI ≥ 24 kg/m^2, TIS > 2 days, PWB after surgery ≤ 3 months, and the presence of preoperative DM were confirmed as the risk factors for postoperative ischemic ONFH (odds ratio > 1, $p < 0.05$) (Table 3).

Table 1

General data of patients with Pauwels type III femoral neck fractures who underwent surgery

Parameter	Group		χ^2	*p*
	ONFH occurrence (n = 30)	ONFH non-occurrence (n = 182)		
Gender			1.428	0.232
male	15 (50.00)	112 (61.54)		
female	15 (50.00)	70 (38.46)		
Age, years			21.310	< 0.001
> 55	20 (66.67)	45 (24.73)		
≤ 55	10 (33.33)	137 (75.27)		
Body mass index, kg/m^2			11.996	0.001
≥ 24	18 (60.00)	51 (28.02)		
< 24	12 (40.00)	131 (71.98)		
Injury cause			0.002	0.963
falls	12 (40.00)	72 (39.56)		
car accidents	18 (60.00)	110 (60.44)		
Injured side			1.322	0.250
left	14 (46.67)	65 (35.71)		
right	16 (53.33)	117 (64.29)		
Traction			2.640	0.104
yes	13 (56.67)	52 (28.57)		
no	17 (43.33)	130 (71.43)		
Duration of surgery, hrs			1.205	0.272
> 2	11 (36.67)	49 (26.92)		
≤ 2	19 (63.33)	133 (73.08)		
TIS, days			21.220	< 0.001
> 2	17 (56.67)	33 (18.13)		
≤ 2	13 (43.33)	149 (81.87)		
PWB time, months			17.641	<0.001
≤ 3	14 (46.67)	26 (14.29)		
> 3	16 (53.33)	156 (85.71)		
Preoperative diabetes mellitus			28.984	<0.001
yes	18 (60.00)	29 (15.93)		
no	12 (40.00)	153 (84.07)		
Preoperative hypertension			2.110	0.146
yes	12 (40.00)	52 (28.57)		
no	18 (60.00)	140 (71.43)		

ONFH – osteonecrosis of the femoral head; TIS – time from injury to surgery; PWB – partial weight bearing. All values are given as numbers (percentages).

Table 2

Variable assignment methods

Variable	Meaning	Assignment	
		1	0
X1	Age, years	> 55	≤ 55
X2	Body mass index, kg/m^2	≥ 24	< 24
X3	TIS, days	> 2	≤ 2
X4	PWB time after surgery, months	≤ 3	> 3
X5	Preoperative diabetes mellitus	yes	no
Y	Ischemic ONFH	yes	no

ONFH – osteonecrosis of the femoral head; For other abbreviations, see Table 1.
1 – ischemic ONFH occurrence group; 0 – ischemic ONFH non-occurrence group.

Table 3

Logistic regression analysis results of multiple risk factors for ischemic ONFH after internal fixation with multiple CSs for Pauwels type III FNF

Variable	β	Standard error	Wald	*p*	Odds ratio	95% confidence interval
Age	1.806	0.424	18.178	< 0.001	6.089	2.654–13.970
Body mass index	1.349	0.408	10.951	0.001	3.853	1.733–8.565
TIS	1.776	0.416	18.251	< 0.001	5.904	2.614–13.334
PWB	1.658	0.423	15.379	< 0.001	5.250	2.292–12.025
Preoperative DM	2.069	0.424	23.785	< 0.001	7.914	3.446–18.173

DM – diabetes mellitus; CSs – cannulated screws; FNF – femoral neck fracture.
For other abbreviations, see Table 1.

Discussion

The incidence of FNF is gradually increasing because of transportation development and the acceleration of the aging process [6]. FNF is caused by external forces on the FN. It is highly related to osteoporosis. The fracture can occur when FN is under a slight torsional force, and it is more likely to be found in middle-aged and elderly people since such populations are the most vulnerable to osteoporosis. Displacement of the FNF damages the basilar artery ring outside the joint capsule, which compromises the main blood supply to the femoral head, thus resulting in avascular ONFH [7]. Moreover, FNF leads to changes in neck-shaft angle and FN anteversion and destroys the balance between structure and function, which results in stress concentration in the joint and remodeling of the femoral head bone trabecular microstructure. Bone trabecular degeneration and collapse may occur if the remodeled structure fails to adapt to the new compressive stress and biomechanical environment, inducing further malformation and obstruction of peripheral blood vessels. Eventually, ONFH occurs after FNF surgery, reflecting on the postoperative quality of life [8]. PTIII FNF is followed by a large vertical shear force, poor stability, and relatively high risk of IF failure or fracture displacement after surgical reduction, easily causing severe damage to the femoral head blood supply and consequent fracture nonunion and ONFH [9, 10]. Studies have manifested that the IF approach of inserting multiple CSs into FN is the most stable [11, 12]. Various studies have demonstrated that despite the high clinical value, IF with multiple CSs still leads to ONFH. If not detected on time and if not treated actively, ONFH is followed by painful and dysfunctional hip joint, inducing socioeconomic problems and even requiring artificial joint replacement [13, 14]. For this reason, actively preventing the ONFH after IF with multiple CSs for PTIII FNF is a crucial measure for effectively improving the prognosis of these patients.

In this study, 30 patients had postoperative ischemic ONFH, with an incidence rate of 14.15%, which is in line with that reported in previous literature [10]. In the face of the high incidence rate of ONFH in patients undergoing surgery for PTIII FNF, age has always been a key research issue. Kim and Kim [15] pointed out that a higher incidence rate of postoperative ONFH is found in older patients with PTIII FNF. The reason may be the fact that older patients experience greater calcium loss, gradual decline in body functions, and progressive failure of organ functions, thus developing osteoporosis. Besides, Zhao et al. [16] reported that PTIII FNF patients with BMI ≥ 24 kg/m^2 had a higher incidence rate of postoperative ONFH, which is in accordance with our study. Excessive BMI usually suggests higher blood viscosity, which increases the risk of thrombosis and microcirculation supply disorder, further inducing postoperative ONFH. In addition, the incidence of postoperative ischemic ONFH is related to TIS. A higher number of patients with TIS > 2 days in the ischemic ONFH occurrence group, as found in our study, can be explained by the state that earlier surgery can cure vascular distortion, compression, and spasm after quick fracture reduction. This leads to a decrease of the pressure in the joint capsule (previously increased by the fracture) and improves or eliminates the effect of packing, which thereby reopens the temporarily closed blood vessels and restores the remaining blood supply. However, regarding the time of > 2 days from injury to surgery, the bone structure undergoes great changes, and joint stability becomes worse, leading to a higher incidence rate of postoperative ischemic ONFH. Furthermore, the incidence rate of ONFH rises significantly in patients with PWB time after surgery ≤ 3 months [17]. The results of this study revealed that the proportion of patients with PWB time after surgery ≤ 3 months was remarkably larger in the ischemic ONFH occurrence group than in the ischemic ONFH non-occurrence group. A higher incidence of ischemic ONFH when PWB time after surgery is ≤ 3 months can be explained by the fact that within three months after surgery, the patient's fractures are not fully healed, and premature PWB will cause considerable stress on the FN. As a result, IF screw loosening and cutting easily occur, making IF ineffective and thereby resulting in unstable immobilization, shortening, and *varus* deformity. Eventually, nonunion occurs, further increasing the incidence rate of ONFH [18]. Moreover, patients with preoperative DM are more susceptible to postoperative ONFH [19]. It was indicated in our study that the ischemic ONFH occurrence group had more patients with preoperative DM than the ischemic ONFH non-occurrence group. Patients with preoperative DM suffer from a multi-system dysfunction caused by impaired glucose metabolism

in the body. Abnormal glucose metabolism will directly cause vascular endothelial proliferation and thicken the capillary basement membrane, narrowing the vascular lumen, reducing the elastic function of the wall, and impairing the blood supply capacity. However, the femoral head has a greater demand for blood supply due to its specific blood supply. The remodeling of blood vessels after FNF directly impacts the blood supply to the femoral head. In patients with preoperative DM, blood vessels have undergone different degrees of damage before the injury, and the blood supply to the femoral head suffers secondary damage caused by FNF. As a result, the risk of ONFH in such patients is greatly increased.

Conclusion

In summary, age > 55 years, body mass index ≥ 24 kg/m^2, time from injury to surgery > 2 days, partial weight bearing time after surgery ≤ 3 months, and preoperative diabetes mellitus presence are considered the risk factors for ischemic osteonecrosis of the femoral head after internal fixation with multiple cannulated screws for Pauwels type III femoral neck fracture.

Conflict of interest

The authors declare no conflict of interest.

R E F E R E N C E S

1. *Liu P, Zhang Y, Sun B, Chen H, Dai J, Yan L.* Risk factors for femoral neck fracture in elderly population. Zhong Nan Da Xue Xue Bao Yi Xue Ban 2021; 46(3): 272–7.
2. *Ziesler C, Engebretsen L.* Femoral neck stress fracture. Tidsskr Nor Laegeforen 2020; 140(5). (Norwegian)
3. *Bernstein EM, Kelsey TJ, Cochran GK, Deafenbaugh BK, Kuhn KM.* Femoral Neck Stress Fractures: An Updated Review. J Am Acad Orthop Surg 2022; 30(7): 302–11.
4. *Wang Y, Ma JX, Yin T, Han Z, Cui SS, Liu ZP,* et al. Correlation Between Reduction Quality of Femoral Neck Fracture and Femoral Head Necrosis Based on Biomechanics. Orthop Surg 2019; 11(2): 318–24.
5. *Zhao D, Zhang F, Wang B, Liu B, Li L, Kim SY,* et al. Guidelines for clinical diagnosis and treatment of osteonecrosis of the femoral head in adults (2019 version). J Orthop Translat 2020; 21: 100–10.
6. *Bastard C.* Femoral neck fracture. Rev Prat 2019; 69(10): 1124–8. (French)
7. *Konarski W, Poboży T, Śliwczyński A, Kotela I, Krakowiak J, Hordowicz M,* et al. Avascular Necrosis of Femoral Head-Overview and Current State of the Art. Int J Environ Res Public Health 2022; 19(12): 7348.
8. *Steele CE, Cochran G, Renninger C, Deafenbaugh B, Kuhn KM.* Femoral Neck Stress Fractures: MRI Risk Factors for Progression. J Bone Joint Surg Am 2018; 100(17): 1496–502.
9. *Su Z, Liang L, Hao Y.* Medial femoral plate with cannulated screw for Pauwels type III femoral neck fracture: A meta-analysis. J Back Musculoskelet Rehabil 2021; 34(2): 169–77.
10. *Lim EJ, Shon HC, Cho JW, Oh JK, Kim J, Kim CH.* Dynamic Hip Screw versus Cannulated Cancellous Screw in Pauwels Type II or Type III Femoral Neck Fracture: A Systematic Review and Meta-Analysis. J Pers Med 2021; 11(10): 1017.
11. *Gao Z, Wang M, Shen B, Chu X, Ruan D.* Treatment of Pauwels type III femoral neck fracture with medial femoral neck support screw: a biomechanical and clinical study. Sci Rep 2021; 11(1): 21418.
12. *Teng Y, Zhang Y, Guo C.* Finite element analysis of femoral neck system in the treatment of Pauwels type III femoral neck fracture. Medicine (Baltimore) 2022; 101(28): e29450.
13. *Xu JL, Liang ZR, Xiong BL, Zou QZ, Lin TY, Yang P,* et al. Risk factors associated with osteonecrosis of femoral head after internal fixation of femoral neck fracture: a systematic review and meta-analysis. BMC Musculoskelet Disord 2019; 20(1): 632.
14. *Wang W, Li Y, Xiong Z, Guo Y, Li M, Mei H,* et al. Effect of the Number, Size, and Location of Cannulated Screws on the Incidence of Avascular Necrosis of the Femoral Head in Pediatric Femoral Neck Fractures: A Review of 153 Cases. J Pediatr Orthop 2022; 42(3): 149–57.
15. *Kim CH, Kim JW.* A recent update on the fixation techniques for femoral neck fractures: A narrative review. J Clin Orthop Trauma 2024; 54: 102497.
16. *Zhao C, Kan J, Xu Z, Zhao D, Lu A, Liu Y,* et al. Higher BMI and lower femoral neck strength in males with type 2 diabetes mellitus and normal bone mineral density. Am J Med Sci 2022; 364(5): 631–7.
17. *Trauma Orthopedics Group, Orthopedics Branch, Chinese Medical Association, Fixation and Limb Reconstruction Group, Orthopedics Branch, Chinese Medical Association.* Expert consensus on weight bearing after lower limb fractures. Chin J Orthop Trauma 2023; 25(2): 93–100.
18. *Yuan KX, Yang F, Fu K, Zhu DY, Jiang CY, Jin DX,* et al. Internal fixation using fully threaded cannulated compression screws for fresh femoral neck fractures in adults. J Orthop Surg Res 2022; 17(1): 108.
19. *Lai SW, Lin CL, Liao KF.* Real-world database examining the association between avascular necrosis of the femoral head and diabetes in Taiwan. Diabetes Care 2019; 42(1): 39–43.

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COVID-19 pandemic and vaccination rate in patients with psoriasis treated with biologics: a single center experience

COVID-19 pandemija i stopa vakcinacije kod obolelih od psorijaze lečenih biološkim lekovima: iskustvo jednog centra

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Abstract

Background/Aim. Psoriasis is a chronic, immune-mediated, genetically determined disease, which is manifested by the appearance of erythematous scaly plaques. Treatment includes conventional therapies and biologics. The coronavirus disease 2019 (COVID-19) raised widespread concern for patients with psoriasis treated with immunosuppressive drugs, especially biologics. Even though there was no data at the beginning of the pandemic on the efficacy and safety of vaccines against COVID-19 in patients with psoriasis treated with biologics, the National Psoriasis Foundation (United States of America) recommended vaccination in these patients. The aim of this study was to evaluate the influence of COVID-19 on clinical characteristics and quality of life of psoriatic patients treated with biologics and evaluate the effectiveness of biologic therapy during the pandemic. **Methods.** A retrospective cross-sectional study was conducted at the Clinic of Dermatology and Venereology of the University Clinical Center of Serbia from March 2020 to January 2022. Data was collected from medical documentation during the consecutive hospitalization of patients with psoriasis who received biologics. **Results.** The study included a total of 181 patients with psoriasis divided into two groups. Patients from each group were treated with different biologics (ustekinumab in 63.0% and secukinumab in 37.0% of patients). They achieved significant improvement regarding their clinical characteristics after a two-year follow-up [Psoriasis Area and Severity Index (PASI) before treatment: 14.1 (0–50.5) and after treatment: 1.2 (0–49.7), $p < 0.001$] and quality of life [Dermatology Life Quality Index (DLQI) before treatment: 15.0 (0–34) and after treatment: 0 (0–28), $p < 0.001$]. Due to unsatisfactory therapeutic response in 4 (2.2%) patients, secukinumab was changed to ustekinumab. The vaccine against COVID-19 was given to 53.0% of patients, but only 20.4% received all three doses. Infection with severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was confirmed in 64 (35.4%) patients, and 68.0% of those infected contracted the disease before the first dose of the vaccine. Therapy with biologics was delayed due to SARS-CoV-2 infection in 52 (28.7%) patients, of which 11 (21.2%) had exacerbation of psoriasis. **Conclusion.** The vaccination rate in patients with psoriasis receiving biologics was hardly 50.0%, and about a third of the vaccinated patients had a milder form of COVID-19. The therapy with biologics was successful regardless of the short-term interruption of drug administration due to the beginning of the COVID-19 pandemic and the worsening of psoriasis in some patients during that time.

Key words:
biological therapy; covid-19; psoriasis; vaccination.

Apstrakt

Uvod/Cilj. Psorijaza je hronična, genetski uslovljena bolest, posredovana imunskim mehanizmima, koja se manifestuje pojavom eritematoznih plakova sa skvamom. Lečenje podrazumeva konvencionalnu i biološku terapiju. Bolest izazvana korona virusom 2019. (*coronavirus disease 2019* – COVID-19) izazvala je zabrinutost za obolele od psorijaze koji se leče imunosupresivnim lekovima, posebno biološkom terapijom. Iako na početku pandemije nije bilo dovoljno podataka o efikasnosti i bezbednosti vakcine protiv COVID-19 kod obolelih od psorijaze na biološkoj terapiji, Nacionalna fondacija za psorijazu (Sjedinjene Američke Države) preporučila je vakcinaciju protiv COVID-19 kod ovih bolesnika. Cilj studije bio je da se ispita uticaj COVID-19 na kliničku sliku i kvalitet života

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obolelih od psorijaze koji su lečeni biološkom terapijom, kao i da se ispita efikasnost biološke terapije tokom pandemije. **Metode.** Retrospektivna studija preseka sprovedena je na Klinici za dermatologiju i venerologiju Univerzitetskog kliničkog centra Srbije, u periodu od marta 2020. do januara 2022. godine. Podaci su prikupljeni iz medicinske dokumentacije tokom konsekutivne hospitalizacije obolelih od psorijaze lečenih biološkom terapijom. **Rezultati.** Istraživanje je obuhvatilo 181 bolesnika obolelih od psorijaze podeljenih u dve grupe. Bolesnici iz obe grupe lečeni su različitim biološkim lekovima (ustekinumab kod 63,0% i sekukinumab kod 37,0% bolesnika). Kod njih je postignuto značajno poboljšanje kliničke slike, koja je praćena tokom dve godine [*Psoriasis Area and Severity Index* (PASI) pre lečenja: 14,1 (0–50,5) i posle lečenja: 1,2 (0–49,7), $p < 0,001$], i kvaliteta života [*Dermatology Life Quality Index* (DLQI) pre lečenja: 15,0 (0–34) i posle lečenja: 0 (0–28), $p < 0,001$]. Zbog nezadovoljavajućeg terapijskog odgovora kod 4 (2,2%) bolesnika, sekukinumab je zamenjen ustekinumabom. Vakcinu protiv COVID-19 primilo je 53,0% bolesnika, ali je sve tri doze primilo samo 20,4% bolesnika. Infekcija virusom *severe acute respiratory syndrome coronavirus 2* (SARS-CoV-2) potvrđena je kod 64 (35,4%) bolesnika a 68,0% od ukupnog broja zaraženih obolelo je pre prve doze vakcine. Terapija biološkim lekovima bila je odložena zbog SARS-CoV-2 infekcije kod 52 (28,7%) bolesnika, od kojih je 11 (21,2%) imalo pogoršanje psorijaze. **Zaključak.** Stopa vakcinacije obolelih od psorijaze koji su primali biološku terapiju bila je jedva 50,0%, a približno trećina vakcinisanih bolesnika imala je blaži oblik COVID-19. Biološka terapija bila je uspešna bez obzira na privremeni prekid kod pojedinih bolesnika usled COVID-19 i pogoršanja psorijaze u tom periodu. Biološka terapija je bila uspešna bez obzira na privremeni prekid lečenja koji je nastao usled početka pandemije COVID-19 i pogoršanja psorijaze kod pojedinih bolesnika u tom periodu.

Ključne reči:
biološka terapija; covid-19; psorijaza; vakcinacija.

Introduction

Psoriasis is a chronic, immune-mediated, genetically determined disease of the skin and nails, with a profound negative impact on the patient's quality of life [1, 2]. It affects 1–3% of the population, which is more than 125 million people worldwide [3]. The most common form of the disease is chronic plaque psoriasis (*Psoriasis vulgaris*), affecting 85–90% of patients [4]. Treatment of psoriasis includes topical and/or systemic therapy, such as retinoids, methotrexate, or cyclosporine, used for moderate-to-severe cases [5]. Since the early 2000s, the treatment of psoriasis was improved by biologics, which include inhibitors of tumor necrosis factor (TNFi) (infliximab, etanercept, adalimumab), interleukin (IL)-17 (secukinumab, ixekizumab, brodalumab), IL-12/23 (ustekinumab), and IL-23p19 (guselkumab, risankizumab) [6]. Currently, only two biologics are available in our country – secukinumab and ustekinumab. In terms of its scope, intensity, and dangers to the population's health, the crisis caused by the Coronavirus disease 2019 (COVID-19) pandemic has been, by far, one of the biggest challenges of the 21st century [7]. As the pandemic continued, COVID-19 vaccines were developed. Four types of vaccines have been approved, and those include whole virus vaccines (Sinopharm), mRNA vaccines (Pfizer BioNTech and Moderna), non-replicating viral vector (Oxford-AstraZeneca, Sputnik V), and protein subunit vaccines (Novavax) [8, 9]. This pandemic has raised widespread concern about the use of immunosuppressive agents in the treatment of several diseases, as well as psoriasis, especially in patients treated with biologics [10]. Owing to the differing methods across different studies, at the beginning of the pandemic, it was unknown whether patients with psoriasis had a higher risk of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection [11]. However, the Italian National Psoriasis Foundation suggested that vaccines play a pivotal role in protecting patients with psoriasis against SARS-CoV-2 infection and that these patients do not have to discontinue their prescribed antipsoriatic therapies [12]. Clinical trials showed high efficacy rates and no major safety concerns regarding the use of vaccines against COVID-19 [13]. Brazzelli et al. [14] conducted an observational monocentric prevalence study where they concluded that patients treated with biologics are not susceptible to COVID-19 compared to other psoriatic patients. The National Psoriasis Foundation (United States of America) recommended the use of vaccines against COVID-19 in patients undergoing biological treatment without the need to discontinue therapy [15]. Wu et al. [16] have reported cases of new-onset psoriasis as well as psoriasis flares as adverse events after COVID-19 vaccination.

The aim of this study was to evaluate the impact of COVID-19 on worsening of clinical manifestations of psoriasis, evaluating Psoriasis Area and Severity Index (PASI) and Body Surface Area (BSA) scores, in patients in whom the polymerase chain reaction (PCR) test has confirmed the infection. Moreover, the aim was to evaluate the vaccination rate in patients with psoriasis treated with biologics. To the best of our knowledge, this is the only study in our region that has evaluated these types of data during the COVID-19 pandemic.

Methods

A cross-sectional retrospective study was conducted at the Clinic of Dermatology and Venereology of the University Clinical Center of Serbia during the COVID-19 pandemic (March 2020 – January 2022). The study included 181 psoriatic patients treated with two biological drugs available in our country – secukinumab and ustekinumab. We collected data on the following: the type of biologics, their therapeutic effects [we evaluated PASI and BSA and Dermatology Life Quality Index (DLQI) scores before and after the treatment, but also after the confirmed infection with SARS-CoV-2], the incidence of COVID-19, vaccination rate and type of COVID-19 vaccine that patients received, possible delay of therapy, as

well as the further course of psoriasis. Data was collected from the computer database during consecutive hospitalizations of patients with psoriasis who received biologics.

The PASI and BSA scores were used to assess the disease severity. Based on PASI, psoriasis can be classified as mild (PASI < 3), moderate (PASI 3–10), and severe (PASI > 10) [17]. BSA measures the affected skin area according to the patient's palm surface. For PASI and BSA scores above 10, systemic therapy is recommended. To evaluate the impact of psoriasis on the patient's quality of life, we used a validated DLQI questionnaire. Scores above 10 require systemic therapy [18, 19].

Statistical analysis

Categorical variables were expressed as counts and percentages, while continuous variables were presented as mean ± standard deviation or range. For comparison between variables, the Chi-square test, Student's *t*-test, and Mann-Whitney rank sum test were applied. All statistical methods were considered statistically significant if the *p*-value was less or equal to 0.05.

Results

Demographic and clinical data are shown in Table 1. Initially, ustekinumab was introduced in 60.8% of patients and secukinumab in 39.2% without a significant difference in the frequency of those medications prescribed between genders. Due to unsatisfactory therapeutic response in 4 (2.2%) patients, secukinumab was changed to ustekinumab. Therefore, 63.0% of patients were treated with ustekinumab and 37.0% with secukinumab. The average PASI score before introducing biologics was 14.1 (0–50.5), and two years later, it was 1.2 (0–49.7), $p < 0.001$. There was a statistically significant improvement in the PASI score for all psoriatic patients. The PASI score for patients before starting secukinumab was 13.25 (0–42.0), and two years later, it was 0.95 (0–16.4), $p < 0.001$. For patients before starting ustekinumab, the PASI score was 14.7 (0–50.5), and two years later, it was 1.3 (0–49.7), $p < 0.001$.

The patients were also evaluated for PASI 50, PASI 75, PASI 90, and PASI 100 scores – PASI reduction of 50%, 75%, 90%, and 100% scores, respectively. In both groups (secukinumab and ustekinumab), PASI 50 was achieved in 89.8%, PASI 75 in 77.3%, PASI 90 in 54.7%, and PASI 100 in 23.2% of all patients. In patients treated with secukinumab, PASI 50 was registered in 95.5%, PASI 75 in 80.9%, PASI 90 in 58.8%, and PASI 100 was achieved in 29.4% of patients. In patients treated with ustekinumab, PASI 50 was observed in 86.4% of patients, PASI 75 in 76.4%, PASI 90 in 52.7%, and PASI 100 was achieved in 19.1% of patients. There were no statistically significant differences between PASI 50, 75, 90, and 100 between patients treated with secukinumab and patients treated with ustekinumab (Table 2).

Table 1

Demographic and clinical data of patients with psoriasis treated with biologics (n = 181)

Parameter	Values
Male/Female	101/80 (56/44)
Age (years)	47.36 ± 15.42 (13–78)
Type of psoriasis	
psoriasis vulgaris	168 (92.8)
psoriatic erythroderma	5 (2.8)
nail psoriasis	5 (2.8)
palmoplantar pustular psoriasis	3 (1.6)
PASI	14.1 (0–50.5)
BSA	20.0 (0–98.0)
DLQI	15.0 (0–34)
Ustekinumab	110 (60.8)
Ustekinumab (male/female)	63/47 (57/43)
Secukinumab	67 (37.0)
Secukinumab (male/female)	36/31 (54/46)
Secukinumab changed to ustekinumab	4 (2.2)

PASI – Psoriasis Area and Severity Index; BSA – Body Surface Area; DLQI – Dermatology Life Quality Index.
Values are expressed as numbers (percentages) or numbers (range), except for age which is shown as mean ± standard deviation (range).

Table 2

PASI 50, 75, 90, and 100 in patients treated with secukinumab and ustekinumab

PASI percentage response rate	Total	Secukinumab	Ustekinumab	*p*-value*
PASI 50	159 (89.8)	64 (95.5)	95 (86.4)	0.051
PASI 75	139 (77.3)	55 (80.9)	84 (76.4)	0.368
PASI 90	98 (54.7)	40 (58.8)	58 (52.7)	0.365
PASI 100	41 (23.2)	20 (29.4)	21 (19.1)	0.100

For abbreviations, see Table 1. Results are shown as numbers (percentages).
***For the level of significance of 0.05 according to the Chi-square test.**

The average BSA score before biologic therapy was 20.0 (0–98.0), and two years later, it was 1.0 (0–85.0), $p < 0.001$. There was a statistically significant improvement in BSA score two years after starting biologic therapy. For patients before starting secukinumab, the BSA score was 20 (0–65), and two years later, it was 1.0 (0–30.0), $p < 0.001$. For patients before starting ustekinumab, the BSA score was 18.0 (0–98.0), and two years later, it was 2.0 (0–85.0), $p < 0.001$. The DLQI score before biologic therapy was 15.0 (0–34), and two years later, it was 0 (0–28.0), $p < 0.001$. For patients before starting secukinumab, it was 15 (0–29), and two years later, it was 0 (0–20), $p < 0.001$. For ustekinumab, before therapy, the DLQI score was 14.5 (0–34), and two years later, it was 0 (0–28), $p < 0.001$.

SARS-CoV-2 infection, proven with standard PCR test, was reported in 64/181 (35.4%) psoriatic patients, with 30/64 (46.9%) females and 34/64 (53.1%) males ($p = 0.0592$). Secukinumab was applied among 22/64 (34.4%) patients and ustekinumab among 42/64 (65.6%) patients, with no statistically significant difference ($p = 0.505$). Of the 46.9% of female COVID-19 patients, 9/30 (30.0%) were on secukinumab and 21/30 (70.0%) were on ustekinumab. Of 53.1% of COVID-19 patients of male gender, 13/34 (38.2%) were on secukinumab, and 21/34 (61.8%) were on ustekinumab, with no statistically significant difference ($p = 0.550$).

Therapy with biologics was delayed due to COVID-19 in 52/181 (28.7%) patients, while in another 14/181 (7.7%) patients, the therapy was postponed due to other medical issues (urinary tract infections, other respiratory diseases). Of the 28.7% of patients who had postponed therapy due to COVID-19, exacerbation of psoriasis occurred in 11/52 (21.2%) patients (seven patients on secukinumab and four on ustekinumab). The mean PASI score before exacerbation was 5.1 (0.8–16.2), and after exacerbation, it was 10.9 (1.7–25.8). There was a significant difference in the PASI score ($p < 0.003$). Patients treated with secukinumab had a significant impairment of the PASI score ($p = 0.018$), while patients treated with ustekinumab did not have a significant increase in the PASI score ($p = 0.068$) (Figure 1). The average therapy delay was two months, but it should be emphasized that some patients had a longer delay not only due to COVID-19 but also due to other medical and non-medical reasons (examinations of other organ systems, lack of health insurance, etc.). The average therapy delay time was significantly longer among psoriatic patients who received ustekinumab than among those treated with secukinumab ($p = 0.002$). There was no association between PASI and delay time.

COVID-19 vaccination was performed in 96/181 (53.0%) of our patients; 9/181 (5.0%) received only one dose, 50/181 (27.6%) received two doses, and 37/181 (20.4%) received all three doses. However, 63/181 (34.8%) patients did not receive the vaccine, while for 22/181 (12.2%) of our patients, we have no record of their vaccination. Among the vaccinated patients, the highest percentage received the Sinopharm vaccine at 25.4%, followed by Pfizer at 17%, and Sputnik at 4.4%. We also had patients who combined vaccines with 3.3% receiving a combination of Sinopharm and Pfizer, 1.8% receiving Pfizer and Sputnik, and 1.1% receiving Pfizer and AstraZeneca (Figure 2). There were 42.0% of vaccinated males and 45.0% of females on secukinumab, while 58.0% of males and 55.0% of females were on ustekinumab ($p = 0.340$). There was no significant difference in gender distribution among vaccinated psoriatic patients according to the use of biologic therapy.

Of the total number of patients, 23.8% got infected with the SARS-Cov-2 virus before getting vaccinated (23.0% of females and 26.3% of males). Of the remaining 11.6% of infected patients (who got COVID-19 after vaccination),

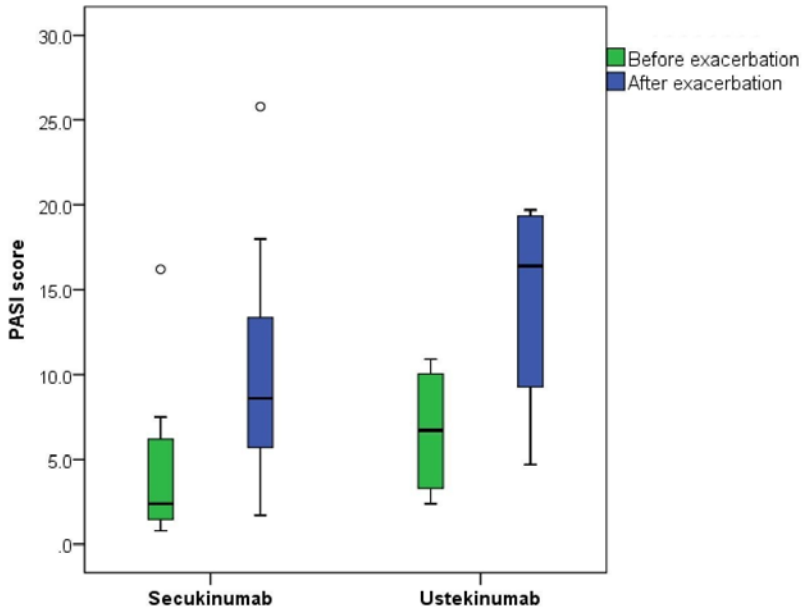

Fig. 1 – Worsening of psoriasis due to coronavirus disease 2019 measured by PASI score.
For abbreviations, see Table 1.

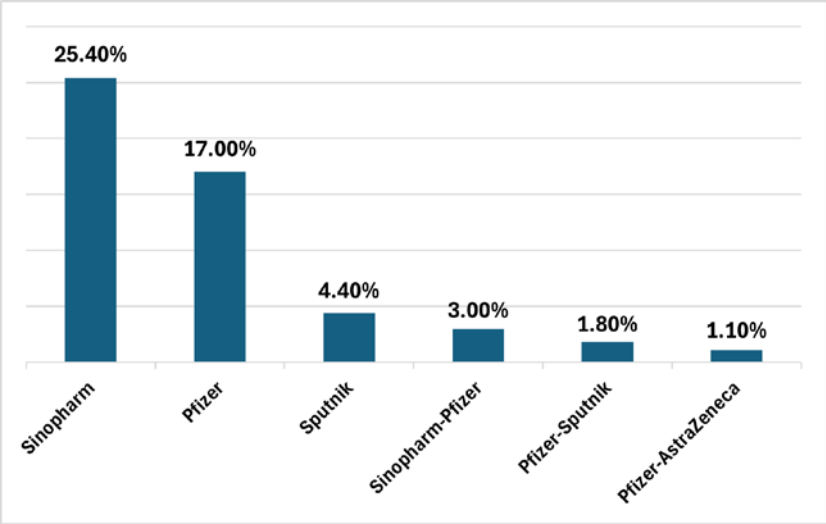

Fig. 2 ─ Vaccination rates in psoriatic patients treated with biologics.

3.1% got COVID-19 a year after two doses of vaccine, 2.0% got COVID-19 right after the second dose, one person (0.55%) got COVID-19 a month after the second dose of the vaccine, and another person (0.55%) got COVID-19 four months after the second dose of the vaccine. Those patients were, in fact, approximately one-fifth of all vaccinated patients, and all of them, except one patient (0.55%), had milder forms of the disease and did not require hospitalization. According to our data, only one male patient treated with ustekinumab died from COVID-19. He was not vaccinated; he had hypertension and a cytokine storm that occurred as part of the COVID-19.

Discussion

This research aimed to investigate the vaccination and SARS-CoV-2 infection rate in patients with psoriasis treated with biologics. We also explored the beneficial effects of treatment with biologics on psoriasis and tried to see if and how the COVID-19 pandemic affected those patients in our country.

In our study, we had a slightly larger number of male patients with psoriasis treated with biologics, which is consistent with other studies [20, 21]. This finding could be related to a slightly higher prevalence of psoriasis and a more severe form of the disease in the male population [22]. For instance, in the Swedish registry for systemic psoriatic treatment (PsoReg), 60.0% of registered patients are men. Other European registries show even bigger numbers in favor of men: Denmark 66.0%, Italy 67.0%, and Spain 63.0% [23]. The introduction of biologics for the treatment of psoriasis has not only drastically changed the course of the disease but also improved the quality of life of these patients. We can conclude this based on the findings of PASI, BSA, and DLQI scores before and after the treatment. PASI 50, PASI 75, PASI 90, and PASI 100 achieved with biologics were also quite satisfactory, as shown in several other studies around the world [24–27]. Complete vaccination was considered the most important means for overcoming the COVID-19 pandemic. Complete vaccination of a significant number of people worldwide helps create collective immunity and drastically decreases the probability of spreading the disease [28]. In a prospective single-center study, Lodde et al. [29] have shown that in people with psoriasis treated with systemic immunosuppressive therapy, as well as in those treated with biologics, an anti-SARS-CoV-2 IgG seroconversion was achieved in 96.1% of patients. Slightly more than half (53.0%) of our patients with psoriasis have been vaccinated against COVID-19, but complete vaccination was conducted in only 20.4%. The most commonly used vaccine was Sinopharm, followed by Pfizer. In a study conducted on Chinese patients with psoriasis, 68.9% received complete vaccination against COVID-19. The most commonly used vaccine among them (89.5%) was the inactivated vaccine (Sinopharm), followed by the protein subunit vaccine (Novavax) in 8.3% of patients, and 2.2% of patients received the adenovirus vector vaccine (AstraZeneca and Johnson & Johnson). The difference in vaccination coverage between Chinese and our patients probably lies in patient awareness, as well as in official recommendations. No severe adverse reactions were reported in vaccinated patients. Moreover, the use of biologics in the treatment of psoriasis was not associated with an increased risk of adverse reactions [30], which is consistent with our experience. The COVID-19 pandemic has influenced and changed the lives of all people around the globe and has certainly affected chronic patients, especially those on immunosuppressive therapy. In our study, we reported that 35.0% of patients had COVID-19 without significant differences in relation to the biological drug they received. Of course, we considered only patients in whom COVID-19 was proven by adequate testing – PCR test for SARS-CoV-2. Due to COVID-19, 29.0% of patients postponed their treatment with biologics, and the average therapy delay was two months. Significant exacerbation of psoriasis presented as an elevated PASI score and was present in one-fifth of the patients. Patients treated with secukinumab had

significant impairment of the PASI score compared with those treated with ustekinumab, which can be explained by the time interval of drug administration (secukinumab is administered every four weeks, while ustekinumab every 12 weeks). Most patients did not have a serious exacerbation of psoriasis. Furthermore, they had mild forms of COVID-19, which did not require hospitalization, and all of them continued therapy with biologics after recovery, and their PASI score improved. Our findings are similar to those from a study by Mroz et al. [20]. They reported that from 57 patients, 19 developed COVID-19, of which three patients on ustekinumab had exacerbation of psoriasis, while only one patient on secukinumab got infected and had no exacerbation of psoriasis. However, all COVID-19 patients had a mild course and did not require hospitalization. One other study conducted in a clinic in central Italy for 136 weeks, including 151 patients with moderate-to-severe plaque psoriasis treated with secukinumab, found that not a single patient reported infection with SARS-CoV-2 [31]. Unfortunately, in our research, one patient treated with ustekinumab died from COVID-19. According to our knowledge, he was not vaccinated, had hypertension, and a cytokine storm occurred as part of the COVID-19 symptoms. He obtained his biologics regularly and was healthy when the treatment was administered. Recent studies showed no adverse impacts from biologics on COVID-19 outcomes in patients with psoriasis [32]. No significant difference was found in the rates of hospitalization when compared to the general population, stratified by age or class of the biologics. Those findings are consistent with other studies that reported that patients with psoriasis on biologics were not at an increased risk of intensive care unit admission or death; they can be, however, at higher risk for testing positive for SARS-CoV-2, to be self-quarantined at home or hospitalized. The authors suggest further application of biologics during the pandemic because there is no evidence that these drugs are responsible for the development of severe complications in COVID-19 [32, 33]. However, some studies suggest that biologics should be discontinued in COVID-19 patients and that the risks and benefits should be carefully weighed in these therapies [34].

Conclusion

The vaccination rate in patients with psoriasis receiving biologics was not satisfactory. Just over half of the patients have been vaccinated against COVID-19, and only one-fifth received all three recommended doses, all without unusual side effects. One-third of all patients had milder forms of COVID-19, but it should be noted that a great majority of them got infected before receiving the vaccine. Almost one-third of the patients discontinued treatment with biologics due to COVID-19, and one-fifth had exacerbation of psoriasis. Fortunately, after re-introducing biologics, the disease subsided, and the biologics were continued according to protocol. We can also confirm that patients treated with both available biologics had very good therapeutic effects and were quite satisfied with the improvement in their quality of life.

Conflict of interest

The authors declare no conflict of interest.

R E F E R E N C E S

1. *Korman NJ*. Management of psoriasis as a systemic disease: what is the evidence? Br J Dermatol 2020; 182(4): 840–8.
2. *Milčić D, Janković S, Vesić S, Milinković M, Janković J*. Assessment of quality of life in patients with psoriasis: a study from Serbia. Int J Dermatol 2015; 54(5): 523–8.
3. *Gupta R, Debbaneh MG, Liao W*. Genetic Epidemiology of Psoriasis. Curr Dermatol Rep 2014; 3(1): 61–78.
4. *Di Meglio P, Villanova F, Nestle FO*. Psoriasis. Cold Spring Harb Perspect Med 2014; 4(8): a015354.
5. *Warren RB, Griffiths CE*. Systemic therapies for psoriasis: methotrexate, retinoids, and cyclosporine. Clin Dermatol 2008; 26(5): 438–47.
6. *Amin M, No DJ, Egeberg A, Wu JJ*. Choosing first-line biologic treatment for moderate-to-severe psoriasis: what does the evidence say? Am J Clin Dermatol 2018; 19(1): 1–13.
7. *Malinić D, Malinić M*. The Impact of COVID-19 pandemic on strategic and operational risk. Financing 2020; 12(4): 3–19.
8. *Fox A, Al-Wassiti H*. 4 things about mRNA COVID vaccines researcher still want to find out [Internet]. Gavi The Vaccine Alliance; 2021 [accessed on 2024 Aug 20]. Available from: https://www.gavi.org/vaccineswork/4-things-about-mrna-covid-vaccines-researchers-still-want-find-out?gclid=CjwKCAiAxJSPBhAoEiwAeO
9. *British Society for Immunology*. Types of vaccines for COVID-19 [Internet]. London, EN: British Society for Immunology; 2021 [accessed on 2024 Aug 20]. Available from: https://www.immunology.org/coronavirus/connect-coronavirus-public-engagement-resources/types-vaccines-for-covid-19
10. *Dhama K, Khan S, Tiwari R, Sircar S, Bhat S, Malik YS*, et al. Coronavirus Disease 2019-COVID-19. Clin Microbiol Rev 2020; 33(4): e00028–20.
11. *Damiani G, Allocco F, Young Dermatologists Italian Network, Malagoli P*. COVID-19 vaccination and patients with psoriasis under biologics: real-life evidence on safety and effectiveness from Italian vaccinated healthcare workers. Clin Exp Dermatol 2021; 46(6): 1106–8.
12. *Gelfand JM, Armstrong AW, Bell S, Anesi GL, Blauvelt A, Calabrese C*, et al. National Psoriasis Foundation COVID-19 Task Force guidance for management of psoriatic disease during the pandemic: Version 2-Advances in psoriatic disease management, COVID-19 vaccines, and COVID-19 treatments. J Am Acad Dermatol 2021; 84(5): 1254–68.
13. *Polack FP, Thomas SJ, Kitchin N, Absalon J, Gurtman A, Lockhart S*, et al. Safety and Efficacy of the BNT162b2 mRNA Covid-19 Vaccine. N Engl J Med 2020; 383(27): 2603–15.
14. *Brazzelli V, Isoletta E, Barak O, Barruscotti S, Vassallo C, Giorgini C*, et al. Does therapy with biological drugs influence COVID-19 infection? Observational monocentric prevalence study on the clinical and epidemiological data of pso-

riatic patients treated with biological drugs or with topical drugs alone. Dermatol Ther 2020; 33(6): e14516.
15. *Koons S*. Vaccinating in the Time of COVID-19 [Internet]. USA: National Psoriasis Foundation; 2020 [accessed on 2024 Aug 20]. Available from: https://www.psoriasis.org/advance/vaccinating-in-the-time-of-covid/
16. *Wu PC, Huang IH, Wang CW, Tsai CC, Chung WH, Chen CB*. New Onset and Exacerbations of Psoriasis Following COVID-19 Vaccines: A Systematic Review. Am J Clin Dermatol 2022; 23(6): 775–99.
17. *Steen Martin D*. How Severe is Your Psoriasis [Internet]. WebMD; 2021 [accessed on 2024 Aug 20]. Available from: https://www.webmd.com/skin-problems-and-treatments/psoriasis/how-severe-your-psoriasis
18. *Finlay AY*. Current Severe Psoriasis and the Rule of Tens. Br J Dermatol 2005; 152(5): 861–7.
19. *Mazzotti E, Barbaranelli C, Picardi A, Abeni D, Pasquini P*. Psychometric properties of Dermatology Life Quality Index (DLQI) in 900 Italian patients with psoriasis. Acta Derm Venereol 2005; 85(5): 409–13.
20. *Mroz M, Mućka S, Miodońska M, Ziolkowska D, Hadas E, Bożek A*. Influence of SARS-CoV-2 Virus Infection on the Course of Psoriasis during Treatment with Biological Drugs. Medicina (Kaunas) 2021; 57(9): 881.
21. *Bragazzi NL, Riccò M, Pacifico A, Malagoli P, Kridin K, Pigatto P*, et al. COVID-19 knowledge prevents biologics discontinuation: Data from an Italian multicenter survey during RED-ZONE declaration. Dermatol Ther 2020; 33(4): e13508.
22. *Hägg D, Eriksson M, Sundström A, Schmitt-Egenolf M*. The higher proportion of men with psoriasis treated with biologics may be explained by more severe disease in men. PLoS One 2013; 8(5): e63619.
23. *Ormerod AD, Augustin M, Baker C, Chosidow O, Cohen AD, Dam TN*, et al. Challenges for synthesising data in a network of registries for systemic psoriasis therapies. Dermatology 2012; 224(3): 236–43.
24. *Rønholt K, Iversen L*. Old and New Biological Therapies for Psoriasis. Int J Mol Sci 2017; 18(11): 2297.
25. *Raudonis T, Gliebute A, Grigaityte AG, Lukosiunaite Z, Karmaziene T, Grigaitiene J*. A Six-Year Analysis of Biological Therapy for Severe Psoriasis in a Lithuanian Reference Centre of Dermatovenereology. Medicina (Kaunas) 2020; 56(6): 275.
26. *Chaptini C, Quinn S, Marshman G*. Durable dermatology life quality index improvements in patients on biologics associated with psoriasis areas and severity index: a longitudinal study. Australas J Dermatol 2016; 57(3): e72–5.
27. *Mattei PL, Corey KC, Kimball AB*. Psoriasis Area Severity Index (PASI) and the Dermatology Life Quality Index (DLQI): the correlation between disease severity and psychological burden in patients treated with biological therapies. J Eur Acad Dermatol Venereol 2014; 28(3): 333–7.
28. *Rashedi R, Samieefar N, Masoumi N, Mohseni S, Rezaei N*. COVID-19 vaccines mix-and-match: The concept, the efficacy and the doubts. J Med Virol 2022; 94(4): 1294–9.
29. *Lodde GC, Krefting F, Placke JM, Schneider L, Fiedler M, Dittmer U*, et al. COVID-19 vaccination in psoriasis patients receiving systemic treatment: A prospective single-center study. Front Immunol 2023; 14: 1107438.
30. *Wang Q, Lv C, Han X, Shen M, Kuang Y*. A Web-Based Survey on Factors for Unvaccination and Adverse Reactions of SARS-CoV-2 Vaccines in Chinese Patients with Psoriasis. J Inflamm Res 2021; 14: 6265–73.
31. *Galluzzo M, D'Adamio S, Silvaggio D, Lombardo P, Bianchi L, Talamonti M*. In which patients the best efficacy of secukinumab? Update of a real-life analysis after 136 weeks of treatment with secukinumab in moderate-to-severe plaque psoriasis. Expert Opin Biol Ther 2020; 20(2): 173–82.
32. *Gisondi P, Zaza G, Del Giglio M, Rossi M, Iacono V, Girolomoni G*. Risk of hospitalization and death from COVID-19 infection in patients with chronic plaque psoriasis receiving a biologic treatment and renal transplant recipients in maintenance immunosuppressive treatment. J Am Acad Dermatol 2020; 83(1): 285–7.
33. *Amerio P, Prignano F, Giuliani F, Gualdi G*. COVID-19 and psoriasis: Should we fear for patients treated with biologics? Dermatol Ther 2020; 33(4): e13434.
34. *Conforti C, Giuffrida R, Dianzani C, Di Meo N, Zalaudek I*. Biologic therapy for psoriasis during the COVID-19 outbreak: The choice is to weigh risks and benefits. Dermatol Ther 2020; 33(4): e13490.

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LETTER TO THE EDITOR

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Re-evaluation of the psychometric properties of the Serbian version of the Primary Sjögren's Syndrome Quality of Life questionnaire

Reevaluacija psihometrijskih karakteristika srpske verzije Upitnika za procenu kvaliteta života osoba sa primarnim Sjegrenovim sindromom

To the Editor

Numerous research conducted worldwide have demonstrated that the health-related (HR) quality of life (QoL) – HRQoL – of patients with primary Sjögren's syndrome (PSS) is notably reduced compared to healthy controls due to the broad spectrum of daily problems they face [1, 2]. In the majority of these studies, HRQoL was assessed by the most frequently used instruments of a generic nature that cannot adequately capture all aspects of disease essential to patients [3–5]. Therefore, specific tools, such as the Primary Sjögren's Syndrome Quality of Life (PSS-QoL) questionnaire developed in 2018, are highly recommended [6]. The pilot study, published in 2022, revealed that the Serbian version of the PSS-QoL questionnaire showed satisfactory feasibility, reliability, and validity [7]. Hence, the aim of this article was to reassess the psychometric properties of the questionnaire in a larger and more diverse group of PSS patients regarding their socio-demographic characteristics, disease activity, and disease-related complications.

This prospective, non-interventional study was carried out at the Rheumatology Clinic of the University Clinical Center of Kragujevac, Serbia between July 2021 and September 2022. The research received approval from the Ethics Committee of the University Clinical Center of Kragujevac (No. 01/20-657). Eighty patients with the diagnosis of PSS established by two rheumatologists [according to the American College of Rheumatology (ACR) or the European League Against Rheumatism (EULAR) classification criteria], aged over 18, with sufficient mental, physical, and linguistic abilities, were enrolled in the study. Prior to the research onset, all participants provided their written informed consent. Patients under 18 with psychiatric disorders and those who refused to participate were excluded. Each participant completed five surveys, including the PSS-QoL, Euro Quality of Life-5D (EQ-5D), EULAR (E) Sjögren's Syndrome (SS) Patients Reported Index (ESSPRI), Oral Health Impact Profile-14 (OHIP-14), and Emotion Regulation Questionnaire (ERQ). Written permission was obtained from the authors of all scales applied in this study. Our pilot research conducted in 2022 provides a comprehensive explanation of the abovementioned questionnaires used [7]. Two methods of administration were tested. Formal translation, adaptation, and validation of the PSS-QoL were performed in the previously published study that included 30 participants, implementing the standard translation/back-translation protocol, adhering to the internationally accepted principles [7, 8]. Temporal stability was tested 14 days after the first completion of the questionnaire by researchers interviewing the participants.

Eighty subjects were enrolled in the study, with a mean age (± standard deviation) of 63.81 ± 10.8 years. The majority of the patients were women (96.2%). The average disease duration was 9.15 ± 7.1 years. The extraglandular manifestations of the PSS were detected in 87.5% of participants (musculoskeletal complications were the most prevalent). The patients were treated with antimalarial therapy, either alone (77.5%) or in combination with corticosteroids (22.5%).

The results that were obtained are given in the following text. The mean values of total and subscale scores of the PSS-QoL at baseline (both modes of administration) and follow-up are illustrated in Table 1. No statistically significant difference was observed between these results ($p > 0.05$). The feasibility of the PSS-QoL scale was excellent, as demonstrated by the high response rate (100%) and absence of missing data. The mean time for completing the questionnaire was 2.57 min (from 1.44 to 4.22 min) when the researchers were questioning the participants and 2.54 min (from 1.33 to 4.26 min) when the subjects did it themselves, indicating minimal patient burden.

Cronbach's alpha coefficient values were 0.931 (when the researcher completed the questionnaire) and 0.921 (when the participants did it on their own), so the reliability of the questionnaire was considered excellent. Seeing that the Spearman-Brown coefficient was 0.923 (when researchers were questioning the patients) and 0.890 (when subjects completed the questionnaire themselves), the reliability of the Serbian version of the PSS-QoL was confirmed. The intraclass correlation coefficient, a measure of temporal

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Table 1

Baseline and follow-up PSS-QoL scores

PSS-QoL	Baseline		Follow-up
	rated by researchers	rated by patients	
Score	41.98 ± 16.50	42.24 ± 16.31	42.04 ± 15.51
Physical	14.01 ± 8.77	13.74 ± 8.49	14.90 ± 8.37
Discomfort	4.46 ± 3.55	4.19 ± 3.29	4.85 ± 3.28
Dryness	9.55 ± 5.53	9.55 ± 5.53	10.05 ± 5.42
Psychosocial	27.97 ± 8.83	28.50 ± 8.85	27.14 ± 8.19

PSS-QoL – Primary Sjögren's Syndrome Quality of Life.
Results are shown as mean ± standard deviation.

Table 2

Multitrait-multimethod matrix

	PSS-QoL (R)	PSS-QoL (P)	EQ-5D (R)	EQ-5D (P)	ESSPRI (R)	ESSPRI (P)	OHIP-14 (R)	OHIP-14 (P)	ERQ (R)	ERQ (P)
PSS-QoL (R)	1	/	/	/	/	/	/	/	/	/
PSS-QoL (P)	0.999**	1	/	/	/	/	/	/	/	/
EQ-5D (R)	-0.766**	-0.771**	1	/	/	/	/	/	/	/
EQ-5D (P)	-0.763**	-0.766**	0.914**	1	/	/	/	/	/	/
ESSPRI (R)	0.848**	0.849**	-0.869**	-0.798**	1	/	/	/	/	/
ESSPRI (P)	0.846**	0.847**	0.917**	-0.821**	0.917**	1	/	/	/	/
OHIP-14 (R)	0.834**	0.836**	-0.767**	-0.746**	0.789**	0.808**	1	/	/	/
OHIP-14 (P)	0.809**	0.812**	-0.741**	-0.723**	0.750**	0.722**	0.967**	1	/	/
ERQ (R)	0.064	0.069	-0.028	-0.031	-0.068	-0.059	0.032	0.047	1	/
ERQ (P)	0.057	0.061	-0.019	-0.012	-0.044	-0.037	0.026	0.041	0.903**	1

R – rated by researchers; P – rated by patients; PSS-QoL – Primary Sjögren's Syndrome Quality of Life; EQ-5D – Euro Quality of life-5D; ESSPRI – European League Against Rheumatism (EULAR) Sjögren's Syndrome Patients Reported Index; OHIP-14 – Oral Health Impact Profile-14; ERQ – Emotion Regulation Questionnaire. * $p < 0.05$; ** $p < 0.001$.

stability, was 0.983 [95% confidence interval (CI): 0.973–0.989], suggesting high PSS-QoL reliability. Considering that all participants completed the questionnaire again after a 14-day interval, an attrition rate of 0% was observed.

The principal axis factoring method was employed for factor analysis. Using Promax rotation, we extracted two factors based on eigenvalue criteria, explaining 59.58% of the total variance. The first factor had an eigenvalue of 6.82 (accounting for 48.72% of variance), while the second factor had an eigenvalue of 1.52 (representing 10.86% of variance). Furthermore, a scree plot supported the decision to extract two factors, as demonstrated by a noticeable "elbow" on the graph. Most questionnaire items were loaded onto the first factor, except for items 12, 13, and 17.

The PSS-QoL outcomes were correlated with the results of EQ-5D, ESSPRI, and OHIP-14 questionnaires for estimating the scale's convergent validity. Spearman's rank correlation test illustrated a strong and significant correlation between observed scales (Table 2). Additionally, the divergent validity of the PSS-QoL instrument was assessed by comparing its total scores with those of the ERQ. The results suggested a weak and non-significant correlation, further indicating the PSS-QoL's ability to distinguish between different constructs (Table 2).

Overall, our findings indicated that the PSS-QoL is a valid scale that can be completed in under five minutes, as evidenced by results derived from both modes of administration. Additionally, PSS-QoL exhibited excellent reliability and significant convergent validity. These outcomes are similar to those attained by our pilot study and the original instrument's authors [6, 7].

In conclusion, the Serbian version of the PSS-QoL scale is a reliable and valid instrument for assessing HRQoL in PSS patients. This scale can be applied in both research and clinical settings as a valuable indicator of patients' QoL, now recognized as one of the crucial treatment outcomes.

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REFERENCES

1. *Dias LH, Miyamoto ST, Giovelli RA, de Magalhães CIM, Valim V.* Pain and fatigue are predictors of quality of life in primary Sjögren's syndrome. Adv Rheumatol 2021; 61(1): 28.
2. *Miyamoto ST, Valim V, Fisher BA.* Health-related quality of life and costs in Sjögren's syndrome. Rheumatology (Oxford) 2021; 60(6): 2588–601.
3. *Azuma N, Katada Y, Yoshikawa T, Yokoyama Y, Nishioka A, Sekiguchi M,* et al. Evaluation of changes in oral health-related quality of life over time in patients with Sjögren's syndrome. Mod Rheumatol 2021; 31(3): 669–77.
4. *Rojas-Alcayaga G, Herrera A, Espinoza I, Rios-Erazo M, Aguilar J, Leiva L,* et al. Illness Experience and Quality of Life in Sjögren

Syndrome Patients. Int J Environ Res Public Health 2022; 19(17): 10969.

5. *Zhang Q, Wang X, Chen H, Shen B.* Sjögren's syndrome is associated with negatively variable impacts on domains of health-related quality of life: evidence from Short Form 36 questionnaire and a meta-analysis. Patient Prefer Adherence 2017; 11: 905–11.
6. *Lackner A, Stradner MH, Hermann J, Unger J, Stamm T, Graninger WB*, et al. Assessing health-related quality of life in primary Sjögren's syndrome-the PSS-QoL. Semin Arthritis Rheum 2018; 48(1): 105–10.
7. *Vujović S, Desnica J, Mijailović S, Milovanović D.* Translation, transcultural adaptation, and validation of the Serbian version of the PSS-QoL questionnaire – a pilot research. Vojnosanit Pregl 2023; 80(6): 493–9.
8. *Wild D, Grove A, Martin M, Eremenco S, McElroy S, Verjee-Lorenz A,* et al. Principles of good practice for the translation and cultural adaptation process for Patient-Reported Outcomes (PRO) measures: report of the ISPOR task force for translation and cultural adaptation. Value Health 2005; 8(2): 94–104.

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I N M E M O R I A M

Prof. dr sc. med. Jefta Kozarski
Pukovnik i redovni profesor u penziji
(1958–2024)

U Kikindi je 23. novembra ove godine preminuo dr Jefta Kozarski, redovni profesor plastične i rekonstruktivne hirurgije na Medicinskom fakultetu Vojnomedicinske akademije (VMA) Univerziteta odbrane u Beogradu i pukovnik u penziji. Vest o njegovoj smrti duboko je potresla brojne kolege, prijatelje, njegove učenike i bivše pacijente.

Profesor Kozarski rođen je 20. oktobra 1958. godine u Skoplju. Osnovnu školu i gimnaziju završio je u Kikindi, a uporedo sa opštim obrazovanjem, profesor Kozarski je završio i osnovnu i srednju muzičku školu. Medicinski fakultet Univerziteta u Beogradu završio je 1982. godine. Posle obavljenog lekarskog staža, započeo je specijalizaciju iz plastične, rekonstruktivne i estetske hirurgije na VMA, gde je 1988. godine položio specijalistički ispit sa odličnom ocenom. Odmah po završetku specijalizacije postavljen je na mesto odeljenskog lekara na Klinici za plastičnu hirurgiju i opekotine VMA. Tokom ratnih dešavanja (1991–1995) prof. dr Jefta Kozarski bio je deo tima koji se naročito istakao u zbrinjavanju velikog broja ranjenika koji su svakodnevno lečeni na VMA i među prvima koji su defekte ratnih rana rekonstruisali mikrohirurškom tehnikom slobodnim režnjevima. Stečeno iskustvo prof. Kozarski je pretočio u doktorsku disertaciju pod nazivom „Biološke karakteristike transplantiranih mikrohirurških režnjeva“, koju je odbranio na VMA 1998. godine.

Za načelnika Kabineta za plastičnu hirurgiju i opekotine postavljen je 1997. godine. Načelnik Odseka za aseptične opekotine postao je 2001. godine, a 2005. godine načelnik Odeljenja za opekotine Klinike za plastičnu hirurgiju i opekotine VMA. Na mesto načelnika Odeljenja za plastičnu hirurgiju, odnosno Prvog odeljenja Klinike postavljen je 2007. godine.

Uporedo sa stručnom karijerom, prof. Kozarski gradio je i više nego uspešnu nastavno-naučnu karijeru, pa je u zvanje asistenta na predmetu Plastična hirurgija izabran 1992. godine. U zvanje docenta izabran je 1999. godine, dok je vanredni profesor postao 2004. godine. Od 2008. godine bio je redovni profesor na Medicinskom fakultetu VMA Univerziteta odbrane. Bio je redovni član komisija za polaganje studentskih i specijalističkih ispita na Medicinskom fakultetu VMA, kao i član komisija za polaganje specijalističkih ispita na Medicinskom fakultetu Univerziteta u Nišu i Medicinskom fakultetu Univerziteta u Novom Sadu. U svojoj bogatoj nastavnoj karijeri prof. Kozarski bio je i šef Katedre za hirurgiju I na Medicinskom fakultetu VMA. Bio je mentor desetinama studenata i specijalizanata plastične, rekonstruktivne i estetske hirurgije.

Prof. dr Jefta Kozarski bio je član nacionalnog udruženja za plastičnu hirurgiju – SRBPRAS, vanredni član Akademije medicinskih nauka Srpskog lekarskog društva, kao i međunarodnih stručnih udruženja BA(S)PRAS, ISAPS, EURAPS i ICOPLAST. Autor je velikog broja radova koji su objavljeni u naučnim časopisima, a svoje rezultate prikazivao je na velikom broju kako domaćih tako i međunarodnih kongresa, kao predavač po pozivu. Takođe, napisao je i dve knjige – monografiju „Biološka vrednost transplantiranih slobodnih režnjeva“ i udžbenik „Povrede tela dejstvom temperature“. Bio je i recenzent u više domaćih i međunarodnih naučnih časopisa.

Prof. Kozarski je voleo medicinu i hirurgiju. Voleo je ljude, muziku, filozofiju, kvizove. Svoje ljubavi spajao je i pretakao jedne u druge, dok se trudio da pomogne ljudima. Pevao je (sa) pacijentima, trudeći se da im olakša njihove muke i bio tu za njih u svakom trenutku. O njegovom

hirurškom umeću govoriće i kolege i mnogobrojni zahvalni pacijenti. Bio je uvek uz kolege, posebno one mlađe, kojima su njegovi saveti i podrška izuzetno značili.

Iza sebe je ostavio suprugu, dvoje dece i unuka Stefana.

U srcu svih nas koji smo ga poznavali ostaće praznina, koju će ispuniti nada da je on sada pronašao svoju nebesku ravnicu, baš onakvu kakva je bila u Kikindi, koju je toliko voleo i pominjao. Verujem da i u toj ravnici uspešno rešava kviz pitanja iz Slagalice, dok sluša „Mndra mja“ i „Stari laloški vals“.

Oficir, nastavnik, hirurg, lekar, ali pre svega čovek bio je naš profesor.

Neka mu je večna slava i hvala!

Pukovnik vanredni prof. dr Goran Šijan

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Examples of references:

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DiMaio VJ. Forensic Pathology. 2nd ed. Boca Raton: CRC Press; 2001.

Blinder MA. Anemia and Transfusion Therapy. In: Ahya NS, Flood K, Paranjothi S, editors. The Washington Manual of Medical Therapeutics, 30th edition. Boston: Lippincot, Williams and Wilkins; 2001. p. 413-28.

Christensen S, Oppacher F. An analysis of Koza's computational effort statistic for genetic programming. In: *Foster JA, Lutton E, Miller J, Ryan C, Tettamanzi AG*, editors. Genetic programming. EuroGP 2002: Proceedings of the 5th European Conference on Genetic Programming; 2002 Apr 3-5; Kinsdale, Ireland. Berlin: Springer; 2002. p. 182-91.

Abood S. Quality improvement initiative in nursing homes: the ANA acts in an advisory role. Am J Nurs [serial on the Internet]. 2002 Jun [cited 2002 Aug 12]; 102(6): [about 3 p.]. Available from: http://www.nursingworld.org/AJN/2002/june/Wawatch.htm

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Primeri referenci:

Đurović BM. Endothelial trauma in the surgery of cataract. Vojnosanit Pregl 2004; 61(5): 491–7. (Serbian)

Balint B. From the haemotherapy to the haemomodulation. Beograd: Zavod za udžbenike i nastavna sredstva; 2001. (Serbian)

Mladenović T, Kandolf L, Mijušković ŽP. Lasers in dermatology. In: *Karadaglić Đ*, editor. Dermatology. Beograd: Vojnoizdavački zavod & Verzal Press; 2000. p. 1437–49. (Serbian)

Christensen S, Oppacher F. An analysis of Koza's computational effort statistic for genetic programming. In: *Foster JA, Lutton E, Miller J, Ryan C, Tettamanzi AG*, editors. Genetic programming. EuroGP 2002: Proceedings of the 5th European Conference on Genetic Programming; 2002 Apr 3-5; Kinsdale, Ireland. Berlin: Springer; 2002. p. 182-91.

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Sve tabele pripremaju se sa proredom 1,5 na posebnom listu. Obeležavaju se arapskim brojevima, redosledom pojavljivanja, u levom uglu (**Tabela 1**), a svakoj se daje kratak naslov. Objašnjenja se daju u fus-noti, ne u zaglavlju. Svaka tabela mora da se pomene u tekstu. Ako se koriste tuđi podaci, obavezno ih navesti kao i svaki drugi podatak iz literature.

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