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Selçuk Tıp Dergisi, Türkiye Atıf Dizini, Index Copernicus, International Citation Index, Türk Atıf Dizini (ULAKBİM) ve EBSCO tarafından indekslenmektedir.

Cilt: 38 Sayı: 4 ARALIK 2022

Araştırma Makalesi / Research Article

Olgu Sunumu / Case Report

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Selçuk Tıp Dergisi Editörlüğü
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Selçuk Tıp Dergisi yılda dört sayı (Mart, Haziran, Eylül, Aralık) yayınlanan "peer review" yöntemi ile çalışan hakemli bir dergidir. Selçuk Tıp Dergisi Türkiye Atıf Dizini, Index Copernicus, International Citation Index, Türk Atıf Dizini (ULAKBİM) ve EBSCO arafından indekslenmektedir.

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DERGİ HAKKINDA

İlk olarak 1984 yılında yayın hayatına başlayan Selçuk Tıp Dergisi (Selcuk Med J) (ISSN: 1017-6616, e-ISSN: 2149-8059), Necmettin Erbakan Üniversitesi, Meram Tıp Fakültesi'nin bağımsız, çift kör, hakemli bilimsel yayın organıdır. Dergimiz Mart, Haziran, Eylül ve Aralık aylarında üç ayda bir yayımlanmaktadır.

Derginin sayılarına tam erişim aşağıdaki adresten temin edilebilir.

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Selçuk Tıp Dergisi (Selcuk Med J) dünya genelinden yayınları kabul etmektedir. Yayınlar en az iki hakem tarafından orijinal bilgi, fikir ve sunum açısından değerlendirilmektedir.

Selçuk Tıp Dergisi (Selcuk Med J), Türkçe ve/veya İngilizce makaleleri yayınlar, dergi içeriklerine ücretsiz erişim sağlar (Open Access) ve yazarların makalelerin son kabul edilmiş halini OAI uyumlu kurumsal / konu bazlı bir havuzda kendi arşivleyebilmelerine izin verir.

Selçuk Tıp Dergisi (Selcuk Med J) tıp doktorları, araştırmacılar ve bilim adamlarından oluşan bir kitleye yönelik çok disiplinli bir dergidir.

Etik Kurallara Uygunluk

Derginin yayın ve yayın süreçleri Uluslararası Medikal Dergisi Editörleri Komitesi (International Committee of Medical Journal Editors) (ICMJE), Dünya Tıbbi Editörler Birliği (World Association of Medical Editors) (WAME), Bilim Editörleri Konseyi (Council of Science Editors) (CSE), Yayın Etiği Komitesi (Committee on Publication Ethics) (COPE)), Avrupa Bilim Editörleri Derneği (European Association of Science Editors) (EASE) ve Ulusal Bilgi Standartları Örgütü (National Information Standards Organization) (NISO) yönergelerine uygun olarak şekillendirilmiştir.

Dergimiz 'Şeffaflık ve Akademik Yayıncılık En İyi Uygulamalar İlkelerine' (Principles of Transparency and Best Practice in Scholarly Publishing) (doaj.org/bestpractice) uygundur.

Makalelerin daha önce hiçbir yerde yayınlanmamış ve yayın için başka bir dergiye gönderilmemiş olması gerekir. Selçuk Tıp Dergisi'nde intihal programı (iThendicate) kullanılmaktadır. Akademik atıf sınırını aşan benzerlik taşıyan makaleler ve yayın kurallarına uygun olarak hazırlanmamış makaleler değerlendirmeye alınmayacaktır. Tüm çalışmalarda etik kurul onayı gerekmektedir ve bu onamın belgelendirilmesi yazıların yayınlanmasında esas teşkil edecektir.

Tüm çalışmalarda yazarların çalışmaya katkı düzeyi ve onayı bildirilmelidir. Çalışmada veri toplanması, deney aşaması, yazım ve dil düzenlemesi dahil olmak üzere herhangi bir aşamasında finansal çıkar çatışması olmadığı bildirilmelidir. Çalışmada varsa ticari sponsorluk bildirilmelidir.

Yazarlardan Selçuk Tıp Dergisi'nde yayımlanacak makaleleri için herhangi bir ödeme istenmez.

Bütün makaleler editor ve yayın kurulu tarafından üç ay içerisinde sonuçlandırılacaktır. Fakat elde olmayan gecikmelerden dolayı bu süre uzayabilir.

Dergi Amaç ve Kapsamı

Selçuk Tıp Dergisi amacı, genel tıp alanında tanı ve tedavideki güncel gelişmeler, cerrahi yenilikler ve bilim dünyasına katkıda bulunacak deneysel çalışmaların ulusal ve uluslararası literatürde paylaşımının sağlanmasıdır.

Selçuk Tıp Dergisi, sağlık bilimlerindeki tüm etik yönergelere uygun olarak hazırlanmış klinik ve deneysel araştırma makaleleri, olgu bildirileri, derleme makaleleri, teknik notlar ve editöre mektupları yayınlamaktadır.

Bu Sayıda Görev Alan Hakemler

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YAZARA AÇIKLAMA

Selçuk Tıp Dergisi (Selcuk Med J) Necmettin Erbakan Üniversitesi, Meram Tıp Fakültesi Dekan'lığının yayın organıdır. Dergimize yazı hazırlanırken aşağıdaki açıklamaları lütfen bütünüyle okuyunuz.

Selçuk Tıp Dergisi (Selcuk Med J) tıp bilimine ve akademik çalışmalara katkısı olan, klinik ve deneysel çalışmaları, editoryal yazıları, kısa raporları, klinik olgu bildirimlerini, teknik ve eğitici derlemelerini, tıp konusundaki son gelişmeler ile orijinal görüntü raporlarını, görüntülü hastalık tanımlama sorularını ve editöre mektupları yayınlar. Ayrıca daha önce yayınlanmış makale ve deneysel çalışmalarla ilgili okuyucu soru ve katkıları kısaca yayınlanır. Yayına kabul edilme, editöryal komite ile en az iki hakem kararı ile alınır. Bir hakem, hakemlik talebini kabul etmeye karar vermeden önce, hakem değerlendirme süreci ve gözden geçirmenin nasıl yapılacağı hakkında daha fazla bilgi edinmek isteyebilir.

Hakemler, Selçuk Tıp Dergisi'nin gereklerine, önceden tanımlanmış kriterlere ve sunulan araştırmanın kalitesine, eksiksizliğine ve doğruluğuna dayanarak makale gönderimini değerlendirir. Hakemler makale hakkında geri bildirimde bulunur, iyileştirmeler önerir ve makalede yapılan değişiklikleri kabul edip etmeme, talep etme veya reddetme konusunda editöre tavsiyede bulunur. Nihai karar her zaman baş editöre aittir, ancak hakemler sonucu belirlemede önemli bir rol oynamaktadır. Bir hakemin makaleyle çıkar çatışması varsa, editöre bildirmesi gerekir. Hakemler, hakem gözden geçirme sistemine katılarak bilimsel sürecin katı standartlarını sağlamalıdır. Ayrıca, geçersiz araştırmaları tespit ederek ve derginin kalitesini korumaya yardımcı olarak derginin bütünlüğünü korumalıdırlar. Hakemler, intihal, araştırma sahtekarlığı ve diğer sorunları tespit ederek etik konuların ihlal edilmesini önlemeye gönüllü olmalıdır.

Yayına kabul edilen yazıların her türlü yayın hakkı dergiye aittir. Bu hak özel düzenlenmiş yayın hakkı devir formu ile bütün yazarların imzası ile tespit edilir. Dergi 3 ayda bir, yılda 4 kez yayınlanır. Derginin yayın dili Türkçe ve/veya İngilizcedir. Gönderilen yazılar daha önce herhangi bir dergide yayınlanmamış olmalıdır (Bilimsel kongrelerde sunulan sözlü bildiri ve posterler bildirmek kaydı ile hariçtir). Dergide yayımlanan yazıların her türlü sorumluluğu (etik, bilimsel, yasal vb.) yazarlara aittir. Yazım kurallarına uygun olarak hazırlanmamış olan yazıların incelenmeye alınıp alınmaması Yayın Kurulu'nun insiyatifindedir.

Makalelerin daha önce hiçbir yerde yayınlanmamış ve yayın için başka bir dergiye gönderilmemiş olması gerekir. Selçuk Tıp Dergisi'nde intihal programı (iThendicate) kullanılmaktadır. Akademik atıf sınırını aşan benzerlik taşıyan makaleler ve yayın kurallarına uygun olarak hazırlanmamış makaleler değerlendirmeye alınmayacaktır. Tüm çalışmalarda etik kurul onayı gerekmektedir ve bu onamın belgelendirilmesi yazıların yayınlanmasında esas teşkil edecektir.

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Derginin editöryal ve yayın süreçleri International Committee of Medical Journal Editors (ICMJE), World Association of Medical Editors (WAME), Council of Science Editors (CSE), Committee on Publication Ethics (COPE), European Association of Science Editors (EASE) ve National Information Standards Organization (NISO) organizasyonlarının kılavuzlarına uygun olarak biçimlendirilmiştir. Selçuk Tıp Dergisi'nin editöryal ve yayın süreçleri, Principles of Transparency and Best Practice in Scholarly Publishing (doaj.org/bestpractice) ilkelerine uygun olarak yürütülmektedir. Yayın Kurulu, dergimize gönderilen çalışmalar hakkındaki intihal, atıf manipülasyonu ve veri sahteciliği iddia ve şüpheleri karşısında COPE kurallarına uygun olarak hareket edecektir.

Derginin Yayın Kurulu, itiraz ve şikayet vakalarını, COPE rehberleri kapsamında işleme almaktadır. Yazarlar, itiraz ve şikayetleri için doğrudan baş editör veya yayın kurulu ile temasa geçebilirler. İhtiyaç duyulduğunda Yayın Kurulu'nun kendi içinde çözemediği konular için tarafsız bir temsilci atanmaktadır. İtiraz ve şikayetler için karar verme süreçlerinde nihai kararı Baş Editör verecektir. Yayıncı ve editör gerektiğinde düzeltmeler, açıklamalar, geri çekilmeler ve özürler yayınlamaya her zaman hazırdır.

Selçuk Tıp Dergisi (Selcuk Med J) ile ilgili tüm yazışmalar, makale gönderme, makalenin takibi, danışman raporları, düzeltmelerin yapılıp yüklenmesi, kabul yazısı gönderimi ve diğer tüm makale ile ilgili formların yüklenmesi https://www.selcukmedj.org sayfasından yapılacaktır. Bu site üzerinden yüklenecek makaleler için kurallar aşağıda belirtilmiştir.

YAZIM KURALLARI

Yayına gönderilen yazılar Microsoft Word programında yazılmalıdır. Yazı, şekil ve grafilerin tamamı elektronik ortamda https://www.selcukmedj.org word ve pdf formatında gönderilmelidir.

Tüm yazılar:

1. Başlık sayfası,
2. Türkçe özet,
3. İngilizce özet,

4. Makale kısmı,
5. Kaynaklar,
6. Tablolar,
7. Şekiller ve resimler,
8. Alt yazılar şeklinde dizilmelidir.

Araştırma inceleme yazılarının makale kısmı (özet, referanslar, tablo, şekil ve alt yazılar hariç) toplam 4000 kelimeyi, özet kısmı 400 kelimeyi, referanslar 60'ı, tablo ve şekil sayısı 10'u geçmemelidir. Özet amaç, gereç ve yöntemler, bulgular ve sonuç bölümlerini içermelidir.

Olgu bildirileri şu bölümlerden oluşmalıdır: Başlık, İngilizce başlık, Türkçe ve İngilizce özet, giriş, olgunun/olguların sunumu, tartışma ve kaynaklar. Olgu sunumları toplam 8 sayfayı geçmemeli ve 3 resimden fazla olmamalıdır. Özet 200 kelimeyi geçmemeli ve tek bir paragraf şeklinde olmalıdır.

Derlemeler İngilizce ve Türkçe özet içermeli ve özet kelime sayısı 300'ü aşmamalıdır. Tablo sayısı ve şekiller (veya resimler) toplam 6 adedi aşmamalıdır. REferanslar 80'i geçmemelidir. Özet tek bir paragraf şeklinde olmalıdır. Editöre mektup, kısa raporlar, görüntü raporları, teknik ve tıp alanındaki gelişmelere ait yazılar ve orijinal konulara ait görüntü sunumları 2 sayfayı geçmemelidir. Kısa bir (100 kelime) İngilizce ve Türkçe özet içermelidir.

YAZILARIN HAZIRLANMASI

Yazının başlığı hem İngilizce hem de Türkçe olarak yazılmalıdır. Yazıda çalışmaya katkısı olan yazarların ad ve soyadları açık olarak yazılmalı. Yazıların altına çalışmanın yapıldığı kurumun açık adresi yazılmalıdır. Çalışma daha önce herhangi bir kongrede sunulmuş ise kongre adı, zamanı (gün-ay-yıl olarak) belirtilmelidir. Başlık sayfasının en altına iletişim kurulacak yazarın adı, soyadı, açık adresi, posta kodu, telefon ve faks numaraları ile e-posta adresi yazılmalıdır.

Özetler

Ayrı bir sayfa olarak verilmelidir. İngilizce özetin başında İngilizce başlık bulunmalıdır. Araştırma inceleme yazılarında 400, olgu sunumlarında 200 kelimeyi geçmemelidir. Araştırma makalelerinde özet amaç, gereç ve yöntemler, bulgular ve sonuç bölümlerini içermelidir. Araştırma ve inceleme yazılarında özetlerden sonra Türkçe ve İngilizce anahtar kelimeler verilmelidir. Anahtar kelime sayısı 5'i geçmemelidir. Anahtar Kelimelerin İngilizcesi Index Medicus'daki Medical Subjects Headings'e uygun olmalı, Türkçe Anahtar kelimeler ise Türkiye Bilim Terimleri'nden (http://www.bilimterimleri.com) seçilmelidir. Özetlerde kısaltma olmamalıdır.

Makale

Yazı Giriş, Gereçler ve Yöntem, Bulgular ve Tartışma bölümlerinden oluşur.

Giriş: Konuyu ve çalışmanın amacını açıklayacak bilgilere yer verilir.

Gereçler ve Yöntem: Çalışmanın gerçekleştirildiği yer, zaman ve çalışmanın planlanması ile kullanılan elemanlar ve yöntemler bildirilmelidir. Verilerin derlenmesi, hasta ve bireylerin özellikleri, deneysel çalışmanın özellikleri ve istatistiksel metotlar detaylı olarak açıklanmalıdır. Çalışma klinik bir çalışma ise başlık 'Hastalar ve Yöntem' şeklinde olmalıdır.

Bulgular: Elde edilen veriler istatistiksel sonuçları ile beraber verilmelidir.

Tartışma: Çalışmanın sonuçları literatür verileri ile karşılaştırılarak değerlendirilmelidir.

Tüm yazımlar Türkçe yazım kurallarına uymalı, noktalama işaretlerine uygun olmalıdır. Kısaltmalardan mümkün olduğunca kaçınılmalı, eğer kısaltma kullanılacaksa ilk geçtiği yerde ( ) içerisinde açıklanmalıdır. Kaynaklar, şekil tablo ve resimler yazı içerisinde geçiş sırasına göre numaralandırılmalıdır. Metin içerisindeki tüm ölçüm birimleri uluslararası standartlara uygun biçimde verilmelidir.

Kaynaklar

Kaynaklar iki satır aralıklı olarak ayrı bir sayfaya yazılmalıdır. Kaynak numaraları cümle sonuna nokta konmadan ( ) içinde verilmeli, nokta daha sonra konulmalıdır. Kaynak yazar isimleri cümle içinde kullanılıyorsa ismin geçtiği ilk yerden sonra ( )
içinde verilmelidir. Birden fazla kaynak numarası veriliyorsa arasına ",", ikiden daha
fazla ardışık kaynak numarası veriliyor ise rakamları arasına ",-" konmalıdır [ör.(1,2), (1-3)gibi]. Kaynak olarak dergi kullanılıyorsa: yıl, cilt, başlangıç ve bitiş sayfaları verilir. Kaynak olarak kitap kullanılıyorsa: sadece yıl, başlangıç ve bitiş sayfaları verilir. Kaynaklarda yazarların soyadları ile adlarının baş harfleri yazılmalıdır. Dergi isimleri Index Medicus'a göre kısaltılmalıdır. Kaynak yazılma şekli aşağıdaki örnekler gibi olmalıdır. Yazar sayısının üçten fazla olması durumunda ise ilk üç yazarın ismi yazılmalı, sonrasında "et al." eklenmelidir.

Dergiler için

1) Kocakuşak A, Yücel AF, Arıkan S. Karına nafiz delici-kesici alet yaralanmalarında rutin abdominal eksplorasyon yönteminin retrospektif analizi. Van Tıp Dergisi 2006;13(3):90-6.

2) Vikse BE, Aasard K, Bostad L, et al. Clinicalprognostic factors in biopsy-proven benign nephrosclerosis. Nephrol Dial Transplant 2003;18:517-23.

Kitaplar için

1) Danovitch GM. Handbook of Kidney Transplantation. Boston: Little, Brown and Company (Inc.), 1996: 323-8.

Kitaptan Bölüm İçin

1) Soysal Z, Albek E, Eke M. Fetüs hakları. Soysal Z, Çakalır C, ed. Adli Tıp, Cilt III, İstanbul Üniversitesi Cerrahpaşa Tıp Fakültesi Yayınları, İstanbul, 1999:1635-50.

2) Davison AM, Cameron JS, Grünfeld JP, et al. Oxford Textbook of Clinical Nephrology. In: Williams G, ed. Mesengiocapillary glomerulonephritis. New York: Oxford University Press, 1998: 591- 613.

Tablolar

Tablolar ayrı sayfaya iki satır aralıklı yazılmalı, her tablonun üzerinde numara ve açıklayıcı ismi olmalıdır. Tabloda kısaltmalar varsa tablonun altında alfabetik sıraya göre açılımları yazılmalıdır. Örnekler: PS: pulmoner stenoz, VSD: ventriküler septal
defekt. Tablolar yazı içindeki bilgilerin tekrarı olmamalıdır.

Şekil ve Resimler

Şekil ve resimler mutlaka isimlendirilmeli ve numaralandırılmalıdır. Resimler minimum 300 dots per inch (dpi) çözünürlüğünde ve net olmalıdır. Resimler makaleden ayrı bir şekilde makale gönderimi esnasında elektronik olarak JPEG formatında gönderilmelidir.

Makale içerisinde geçen resimler kabul edilmeyecektir. Renkli resimlerin basımı ancak yazarın basım ücretini kabul etmesi ve bu ücreti ödemesi halinde mümkün olacaktır. Aksi takdirde resim siyah-beyaz olarak basılır. Şekil ve resim altlarında kısaltmalar kullanılmış ise, kısaltmaların açılımı alfabetik sıraya göre alt yazının altında belirtilmelidir. Mikroskobik resimlerde büyütme oranı ve tekniği açıklanmalıdır. Yayın kurulu, yazının özünü değiştirmeden gerekli gördüğü
değişiklikleri yapabilir.

HAKEM RAPORU SONRASINDA DEĞERLENDİRME

Yazarlar hakem raporunda belirtilen düzeltme istenen konuları maddelendirerek bir cevap olarak kendilerine ayrılan cevap bölümüne yazmalıdırlar ve ek bir dosya şeklinde yüklenmelidir https://www.selcukmedj.org. Ayrıca makale içerisinde de gerekli değişiklikleri yapmalı ve bunları makale içerisinde belirterek (boyayarak) online olarak tekrar göndermelidirler.

SON KONTROL

1. Yayın hakkı devir ve yazarlarla ilgili bildirilmesi gereken konular formu gereğince
doldurulup imzalanmış,
2. Özet makalede 400, olgu sunumunda 200 kelimeyi aşmamış,
3. Başlık Türkçe ve İngilizce olarak yazılmış,
4. Kaynaklar kurallara uygun olarak yazılmış,
5. Tablo, resim ve şekillerde bütün kısaltmalar açıklanmış olmalıdır.

ONLİNE YÜKLEME BASAMAKLARI

https://www.selcukmedj.org sayfasında
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3. Kısa başlık *
4. Türkçe ve İngilizce özet*
5. Türkçe ve İngilizce anahtar kelimeler *
6. Yüklenmesi gerekli bölümler (word makale dosyası, makale dosyasının pdf
formatı, Başlık sayfası, copyright formu, ek dosyalar (resim, şekil ve tablolar) şeklinde
5 basamakta tamamlanmalıdır.

INSTRUCTIONS FOR AUTHORS

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2) Vikse BE, Aasard K, Bostad L, et al. Clinicalprognostic factors in biopsyproven benign nephrosclerosis. Nephrol Dial Transplant 2003;18:517-23.

Book references:

1) Danovitch GM. Handbook of kidney transplantation. Boston: Little, Brown and Company (Inc.), 1996: 323-8. Chapter in book references:

1) Soysal Z, Albek E, Eke M. Fetüs hakları. Soysal Z, Çakalır C, ed. Adli Tıp, Cilt III, İstanbul Üniversitesi, Cerrahpaşa Tıp Fakültesi Yayınları, İstanbul, 1999: 1635-50.

2) Davison AM, Cameron CS, Grünfeld CF, et al. Oxford textbook of clinical nephrogology. In: Williams G, ed. Mesengiocapillary glomerulonephritis. New York: Oxford University Press, 1998: 591- 613.

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NEÜ YAYINEVİ

Evaluation of Executive Functions, Obesity and Self-Esteem in Adolescents with Attention Deficit Hyperactivity Disorder

Dikkat Eksikliği Hiperaktivite Bozukluğu Tanılı Ergenlerde Yönetici İşlevler, Obezite ve Benlik Saygısının Değerlendirilmesi

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Öz

Amaç: Yönetici işlevlerde bozukluklar dikkat eksikliği hiperaktivite bozukluğu (DEHB) kliniğinde oldukça sık görülmektedir. Güncel çalışmalarda obezite ve DEHB'nin patofizyolojisindeki ortak mekanizmaların dürtü kontrolü ve yönetici işlevlerle ilişkili olabileceği öne sürülmektedir. Bu çalışma, aşırı kiloluluk / obezitesi olan ve olmayan DEHB tanılı çocuk ve ergenlerin yönetici işlevleri ve benlik saygısı düzeylerinin karşılaştırılması amaçlanmıştır. Ayrıca, çocuk ve ergenlerde aşırı kiloluluk / obezite, yönetici işlevler, DEHB ve benlik saygısı arasındaki karmaşık ilişkinin incelenmesi amaçlanmıştır.
Hastalar ve Yöntem: Mart 2021- Nisan 2022 arasında Gazi Üniversitesi Çocuk ve Ergen Psikiyatrisi polikliniklerine başvuran DEHB tanılı herhangi bir ilaç kullanmayan 71 ergenin dahil edildiği örneklemin yaş ortalaması 16.12±1.71 (yaş aralığı= 12-18 yıl) yıl olup, %71.83'ü erkeklerden oluşmaktadır. Katılımcılara Yönetici İşlevlere Yönelik Davranış Değerlendirme Envanteri Ölçeği (anne baba formu), Rosenberg Benlik Saygısı Ölçeği ve Conners Ana baba Derecelendirme Ölçeği- Yenilenmiş Kısa Formu verilmiştir.
Bulgular: DEHB tanılı ergenlerde hiperaktivite belirtilerinin ve çalışma belleği fonksiyonunun benlik saygısının en önemli yordayıcıları olduğu saptanmıştır. Ayrıca aşırı kiloluluk / obezite durumuna göre DEHB'li ergenler karşılaştırıldığında çalışma belleği ve planlama/örgütleme fonksiyonları açısından gruplar arası farklılık bulunmuştur. Ancak gruplar arasında benlik saygısı ve DEHB şiddeti açısından bir farklılık saptanmamıştır.
Sonuç: Çalışmamızın sonuçları değerlendirildiğinde DEHB'li ergenlerin benlik saygısında çalışma belleği ve hiperaktivite belirti şiddetinin önemli rol oynayabileceğini göstermektedir. Aşırı kiloluluk/obezite durumuna göre DEHB tanılı ergenlerde benlik saygısı ve DEHB kliniğini açısından farklılık saptanmazken çalışma belleği ve planlama/örgütleme becerilerinde farklılık saptanması, DEHB'de yönetici işlevlerin aşırı kiloluk/obezite için kritik bir rol oynadığını düşündürmektedir.

Anahtar Kelimeler: Yürütücü fonksiyon, obezite, özsaygı, dikkat eksikliği hiperaktivite bozukluğu

Abstract

Aim: Executive function deficits are very common in attention deficit hyperactivity disorders (ADHD). Recent studies suggest that common mechanisms in the pathophysiology of obesity and ADHD may be related to impulse control and executive functions. This study aimed to compare the executive functions and self-esteem levels of children and adolescents with ADHD with and without overweight/obesity. The study also aimed to examine the relationships between overweight/obesity, executive functions, ADHD and self-esteem in children and adolescents.
Patients and Method: Participants included 71 children/adolescents (mean age = 16.12±1.71; age range= 12-18 years; 71.83% males) with ADHD who applied to Gazi University Child and Adolescent Psychiatry outpatient clinics between March 2021 and April 2022 and did not use any medication. Participants completed the Executive Functions Behavior Evaluation Inventory Scale (parent form), Conners Parent Rating Scale-Revised Short Form and Rosenberg Self-Esteem Scale.
Results: Results showed that hyperactivity symptoms and working memory function were significant predictors of self-esteem in adolescents with ADHD. In addition, when adolescents with ADHD were compared according to their overweight/obesity level, significant differences were found between the groups in working memory and planning/organization functions. However, no difference was found between the groups in terms of self-esteem and ADHD severity.
Conclusion: The findings suggest that working memory and hyperactivity symptom severity may play an important role in understanding the self-esteem of adolescents with ADHD. While no difference was found in terms of self-esteem and ADHD clinic in adolescents diagnosed with ADHD according to overweight/obesity status, differences in working memory and planning/organization skills suggest that executive functions play a critical role in overweight/obesity in ADHD.

Key words: Executive function, obesity, self-esteem, attention deficit hyperactivity disorder

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INTRODUCTION

Attention deficit hyperactivity disorder (ADHD) is a neurodevelopmental disorder that begins in childhood and is characterized by symptoms of inattention, hyperactivity, and impulsivity that are inappropriate for the person's age (1). In recent epidemiological studies, the prevalence of ADHD has been reported to be between 5.9% and 7.1% in the world and 12.7% in Turkey (2). A growing body of evidence shows a significant association between ADHD and overweight/obesity (3). Recent studies suggest that common mechanisms in the pathophysiology of obesity and ADHD include changes in abnormal reward center responses, impulse control, and executive functions (4). Executive functions, which are defined as high-level cognitive functions, are skills such as inhibition, abstraction, working memory, emotional regulation, the use of previously acquired skills for environmental conditions, towards the appropriate target, verbal fluency, organization and planning (5). Impairments in executive functions can lead to inadequate attention and planning, difficulties in forming and implementing solution strategies, difficulty in benefiting from feedback, and impaired flexible thinking (6). It has been suggested that learning problems related to executive functions, difficulties in regulating emotional responses, and low academic achievement may lead to negative environmental comments, leading to frustration, failure, and a decrease in self-efficacy and self-esteem in children with ADHD (7, 8). Self-esteem is related to the evaluation of the person about himself, expressing whether he likes it or not, and it shows how important and valuable he sees himself (9-11). This concept is a value judgment expressed by the attitudes developed by the individual for himself. Self-esteem is one of the leading components of mental health and personality development. (12). Low self-esteem can hinder social and cognitive development in children and contribute to the emergence of various mental and physical diseases in adulthood. There is strong evidence that childhood obesity has negative effects on self-esteem and quality of life. (13). Similarly, there are studies showing that children with ADHD have lower self-esteem than controls. (8). In a recent study, children with ADHD are prone to obesity; therefore, it has been suggested that low self-esteem may create more serious problems in obese children with ADHD (14). The findings suggest that there may be a complex relationship between overweight/obesity, self-esteem and executive functions in children and adolescents with ADHD. As far as in our knowledge, there is no study examining this relationship simultaneously. To fill this gap in the literature, this research aimed to examine the complex relationship between overweight/obesity, executive functions, ADHD and self-esteem in children and adolescents with ADHD.

PATIENTS AND METHODS

The sample of this study consisted of 71 adolescents aged 12-18 years who met the diagnostic criteria for attention deficit hyperactivity disorder according to the 5th Edition of the Diagnostic and Statistical Manual of Mental Diseases (DSM-5), who applied to the outpatient clinics of Gazi University Hospital between March 2021 and April 2022. The exclusion criteria of the study were;

1. Diagnosed with additional psychiatric disorders other than ADHD in the DSM-5-based clinical evaluation.
2. Using any psychopharmacological agent (methylphenidate, atomoxetine, risperidone, aripiprazole etc.) in the last three months.
3. While the medical history was questioned in the clinical interview with both the adolescent and the parent, the adolescents who were learned to have a history of any chronic physical disease (eg asthma, epilepsy, hypothyroidism, phenylketonuria, diabetes mellitus, hypertension etc.) that were or should be followed up by pediatrics were not included in the study.

Measures

Sociodemographic Data Form: For this study, sociodemographic form was prepared to obtain from interviews with children, adolescents, and their parents. The form included participants' age, class, family status and age at first diagnosis, clinical evaluation data such as body mass index scores and percentile values calculated according to the reference values of weight, height and body mass index in Turkish children determined by Neyzi et al alongside ADHD predominantly presentation (15).

Conners Parent Rating Scale-Revised Short (CPRS-SF): CPRS-SF was revised from its long form by Conners in 1997 (16). Kaner et al. translated the scale into Turkish and conducted a validity and reliability study in 2013 (17). CPRS-SF consists of 27 items and is answered on a 4-point Likert scale ranging from 0 (never) to 3 (always). Items were collected in three subscales (Oppositional, Hyperactivity and Cognitive problems) and an auxiliary scale (ADHD Index). The CPRS-SF is frequently used in the screening/

determination of ADHD severity and symptoms and in the evaluation of treatment effectiveness in the treatment follow-up period (17).

Rosenberg Self-Esteem Scale (RSES): RSES, developed by Rosenberg to evaluate people's judgments about themselves, consists of 10 items (9). RSES, which is a 4-point Likert-type scale ranging from 1 (very trure) to 4 (very false). High scores on the self-report scale indicate high self-esteem. The validity and reliability study of the scale in our country was performed by Çuhadaroğlu (18).

Behavior Rating Inventory of Executive Function (BRIEF): It was developed by Gioia et al (2002). BRIEF is used to measure executive functions, complex problem-solving skills and adaptive behaviors in daily life in individuals between the ages of 5 and 18 (19). The 86 questions in the BRIEF include 8 subscales (Inhibition, Shift, Emotional Control, Initiate, Working Memory, Plan/ Organize, Organization of Materials and Monitor), 2 comprehensive indexes (Behavioral Regulation Index, Metacognition Index) and the Global Executive Composite, which evaluates both indexes together. (20). High scores on the scale indicate high levels of executive dysfunction. The validity and reliability study for its use in patients with ADHD in our country was conducted by Bakar et al. (2011) (21).

Procedure

After the purpose and method of the study were verbally explained, written and verbal informed consent forms were obtained from the adolescents and their parents who agreed to participate in the study. Overweight and obesity were determined by considering the body mass index values of the World Health Organization (under 5th percentile underweight; 5-84th percentile normal; 85-94th percentile overweight; 95th percentile and above obese) (22). Adolescents with ADHD were divided into two groups according to their body mass index as above the 85th percentile and below the 85th percentile. To evaluate the executive functions and ADHD severity of adolescents, their parents were given the Executive Functions Behavior Evaluation Inventory Scale (parent form), and Conners Parent Rating Scale-Revised Short form, respectively. A self-reported Rosenberg Self-Esteem scale was given to assess adolescents' self-esteem. Ethics committee approval of the study was received from the Ethics Committee of Ağrı İbrahim Çeçen University (Date: 2021, Number: 51).

Statistical Analysis

After the raw data of this study were entered into Microsoft Excel, Statistical Package for Social Sciences (SPSS) version 24.0 was used for statistical analysis of the data. Descriptive analysis was performed to present the demographic characteristics of the variables. Categorical variables such as gender, body mass index, and family structure were presented as numbers and percentages. Continuous variables such as self-esteem, ADHD, and executive function scores were expressed as mean and standard deviation. Skewness and kurtosis statistics are reported to explain the distribution of the variables (Table 1). Findings from the preliminary analysis showed that skewness scores ranged from -1.17 to .46, and kurtosis values ranged from -1.27

Table 1. Descriptive statistics for variables

Variable	Min	Max	Mean	S.D.	Skewness	Kurtosis
Body mass index (Percentile)	5.30	99.20	65.78	28.61	-0.77	-0.56
Self-esteem	17	39	28.86	6.73	-0.34	-0.99
CPRS-SF Oppositional	2	18	9.07	3.90	0.46	-0.11
CPRS-SF Hyperactivity	0	18	8.79	5.00	0.13	-1.27
CPRS-SF Cognitive problems	3	18	13.90	3.96	-0.25	-0.56
CPRS-SF ADHD index	9	35	23.52	6.24	-0.24	-0.40
BRIEF Inhibition	12	30	21.68	4.19	-0.22	-0.39
BRIEF Shift	10	22	16.87	2.92	-0.43	-0.55
BRIEF Emotional control	10	29	20.70	4.54	-0.41	-0.10
BRIEF Initiate	10	24	17.56	3.04	-0.40	0.41
BRIEF Working Memory	13	29	22.17	3.46	-0.69	0.29
BRIEF Plan/Organize	13	34	27.37	4.12	-1.17	1.68
BRIEF Organization of Materials	6	18	13.00	2.47	-0.70	0.90
BRIEF Monitor	12	24	18.07	2.72	-0.45	-0.40
BRIEF Behavioral Regulation Index	41	80	59.24	9.06	-0.10	-0.85
BRIEF Metacognition Index	64	124	98.17	12.26	-0.89	0.96
BRIEF Global Executive Composite	111	204	157.41	19.25	-0.43	0.13

Table 2. Clinical and Sociodemographic data (N=71)

Variable	Groups	N	Yüzde
Gender	Boys	51	71.83
	Girls	20	28.17
Body mass index (Percentile)	Obese	9	12,68
	Over-weight	22	30,98
	Normal	40	56.34
Family	Married	55	77.46
	Divorced	16	22.54
ADHD Presentations	Predominantly Inattentive	27	38.02
	Predominantly Hyperactive/impulsive	2	2.82
	Combined	42	59.16

to 1.68. These scores showed that all variables had a relatively normal distribution (skewness and skewed values <|2|). Visual examination of the variable distribution (eg, histogram) , as well as the Kolmogorov-Smirnov and Shapiro-Wilk tests, also provided further evidence of the normality of the data. An independent sample t-test was used to compare scores for the main variables based on overweight/ obesity. Pearson correlation analysis was applied to determine the relationships between the variables. Multiple regression analysis was performed to determine the predictors of self-esteem. The statistical significance value is based on p <0.05.

RESULTS

The mean age of the sample was 16.12±1.71 years (age range= 12-18 years), while there was71.83% (n=51) boys. Their height ranges from 133 cm to 182 cm (average height = 161.03, SD = 10.73), and their weight ranges from 37kg to 109 kg (average weight = 57.23 kg, SD = 15.43). According to the percentiles calculated according to the body mass index, 12.68% of the participants were considered obese (n=9) and 30.98% (n=22) were considered overweight.

Table 3. Independent sample t-test results

Variable	Over-weight and Obese ADHD Group n=31	Normal Weight ADHD Group n=40	p
Self-Esteem (Mean ± S.D)	27.48±7.52	29.93±5.93	0.13
According to CPRS-SF ADHD scores Oppositional (Mean ± S.D)	9.26±4.16	8.93±3.74	0.72
Hyperactivity (Mean ± S.D)	9.16±5.33	8.93±3.74	0.58
Cognitive problems (Mean ± S.D)	12.81±3.93	11.20±3.88	0.09
ADHD index (Mean ± S.D)	24.77±5.74	22.55±6.51	0.14
According to BRIEF, executive function scores Inhibition (Mean ± S.D)	22.00±4.72	21.43±3.78	0.57
Shift (Mean ± S.D)	17.42±2.88	16.45±2.92	0.17
Emotional control (Mean ± S.D)	21.35±4.62	20.20±4.47	0.29
Initiate (Mean ± S.D)	18.16±2.49	17.10±3.36	0.15
Working Memory (Mean ± S.D)	23.13±2.33	21.43±3.99	0.04
Plan/Organize (Mean ± S.D)	28.45±3.22	26.53±4.56	0.05
Organization of Materials (Mean ± S.D)	13.55±2.29	12.58±2.54	0.10
Monitor (Mean ± S.D)	18.55±2.50	17.70±2.85	0.19
Behavioral Regulation Index (Mean ± S.D)	60.74±8.91	58.08±9.11	0.22
Metacognition Index (Mean ± S.D)	101.84±9.65	95.33±13.39	0.03
Global Executive Composite (Mean ± S.D)	162.58±15.89	153.40±20.81	0.05

Table 4. Correlation between variables

Variable	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.
1. Body mass index (Percentile)	—													
2. Self-Esteem	-0.20	—												
According to CPRS-SF ADHD scores														
3. Oppositional	0.03	-0.04	—											
4. Hyperactivity	0.00	0.14	0.63**	—										
5. Cognitive problems	0.23*	-0.14	0.61**	0.39**	—									
6. ADHD index	0.21	-0.18	0.59**	0.44**	0.80**	—								
According to BRIEF, executive function scores														
7. Inhibition	-0.01	-0.28*	0.25*	0.14	0.36**	0.29*	—							
8. Shift	0.10	-0.22	0.20	0.26*	0.33**	0.39**	0.35**	—						
9. Emotional control	0.04	-0.13	0.14	0.05	0.25*	0.11	0.38**	0.46**	—					
10. Initiate	0.17	-0.13	0.32**	0.48**	0.44**	0.47**	0.47**	0.40**	0.33**	—				
11. Working Memory	0.22	-0.34**	0.27*	0.40**	0.44**	0.49**	0.39**	0.48**	0.17	0.61**	—			
12. Plan/Organize	0.10	-0.12	0.33**	0.37**	0.49**	0.45**	0.44**	0.69**	0.50**	0.56**	0.50**	—		
13. Organization of Materials	0.14	-0.16	0.23	0.30*	0.36**	0.39**	0.16	0.41**	0.09	0.51**	0.52**	0.41**	—	
14. Monitor	0.06	-0.23	0.35**	0.45**	0.27*	0.31**	0.42**	0.44**	0.20	0.43**	0.53**	0.54**	0.28*	—

Not. *. $p < .05$; **. $p < .05$

Detailed sociodemographic and clinical information is presented in Table 2.

To compare the CPRS-SF subscale scores, self-esteem, and executive functions assessed by BRIEF of adolescents with ADHD who are at or above the 85th percentile (overweight and obese adolescents) and below the 85th percentile according to body mass index, independent sample test-test was applied. (Table 3). It has been determined that overweight/obese adolescents have higher working memory, planning/organization, metacognition index and global executive scores of BRIEF than normal-weight adolescents.

Pearson correlation analysis was performed to examine the relationships between self-esteem, percentile, executive functions and ADHD symptoms in adolescents with ADHD and is shown in Table 4. Accordingly, it was found that percentile values in

Table 5. Predictors of self-esteem levels

Variable	B	β	t	p
Model 1		F (3, 70) = 1.52, R = .25, R2 = .06, p > 0.05		
Age	0.67	0.17	1.44	0.15
Gender	0.51	0.03	0.29	0.78
Obesity	2.43	0.18	1.51	0.13
Model 2		F (15, 70) = 2.28, R = .62, R2 = .38, ΔR2 = .32, p < 0.05		
Age	0.47	0.30	0.84	0.22
Gender	1.72	0.12	1.00	0.32
Obesity	0.93	0.07	0.60	0.55
Oppositional	-0.30	-0.17	-1.01	0.32
Hyperactivity	0.73	0.54	3.23	0.01
Cognitive problems	0.20	0.12	0.58	0.56
ADHD index	-0.22	-0.20	-0.98	0.33
Inhibition	-0.22	-0.14	-0.98	0.33
Shift	-0.10	-0.04	-0.27	0.79
Emotional control	-0.11	-0.08	-0.54	0.59
Initiate	-0.06	-0.03	-0.17	0.87
Working Memory	-0.60	-0.31	-1.85	0.07
Plan/Organize	0.25	0.15	0.82	0.42
Organization of Materials	-0.23	-0.08	-0.61	0.55
Monitor	-0.52	-0.21	-1.39	0.17

Not. B = non-standardized coefficients; β = Standard coefficients; Gender (1 = boy, 2 = girl); Obezite (1= existing, 2 = absence)

adolescents with ADHD were positively correlated with the cognitive problems/inattention subscale scores of CPRS-SF. In addition, a negative correlation was found between self-esteem, working memory and inhibition subscales of BRIEF. Positive correlations were found between CPRS-SF's oppositional subscale scores and BRIEF's inhibition, initiate, working memory, plan/organize, and monitor subscales and between CPRS-SF's hyperactivity subscale scores and BRIEF subscales other than inhibition and emotional control. In ,addition positive correlations were found between CPRS-SF's inattention subscale scores and all subscales of BRIEF, and between the ADHD index and all subscales except emotional control.

Multiple regression analysis was performed to examine the roles of executive functions and clinical features of ADHD in predicting self-esteem in adolescents with ADHD. In the regression model, age, gender and obesity variables were controlled in Model 1. The results showed that executive functions (Model 2) explained 32% of the variance in self-esteem. In the regression model, hyperactivity (β = .73, $p<0.01$) and working memory (β = -.60, $p<0.05$) were found to be significant predictors of self-esteem in adolescents with ADHD F (15, 70) = 2.28, R = .62, $R2$ = .38, $\Delta R2$ = .32, $p < 0.05$. The results are shown in Table 5.

DISCUSSION

In this study, the relationship between overweight/obesity, executive functions, self-esteem, and ADHD symptoms and severity were investigated in adolescents with ADHD. Hyperactivity symptoms and working memory function were found to be the most important predictors of self-esteem in adolescents with ADHD. In addition, when adolescents with ADHD were compared according to their overweight/obesity status, although there was a difference between the groups in terms of working memory and planning/organization functions, no difference was found in terms of self-esteem, ADHD symptoms, severity and other executive functions.

In this study, it was found that hyperactivity symptoms were a positive predictor of self-esteem in children and adolescents with ADHD. Although there are conflicting findings in studies investigating self-esteem in children with ADHD, many studies have shown that children with ADHD have lower self-esteem than controls (23-25). However, some studies did not find a significant relationship between ADHD and self-esteem. (26). In parallel with our results, in a recent study in which self-esteem was examined according to ADHD presentation; It was determined that the self-esteem of adolescents with predominantly hyperactive/impulsive ADHD was higher than that of adolescents with both combined and predominantly inattentive ADHD. In the same study, it was stated that the level of self-esteem was related to cognitive functions, and when cognitive functions were evaluated according to predominantly presentations, those with low cognitive functions also had low self-esteem. (27). In our study, due to the low sample size, self-esteem in adolescents with ADHD could not be examined according to predominantly presentations. While internalizing symptoms and learning disorders are more common in adolescents with predominantly inattentive presentation ADHD, it has been reported that adolescents with combined or predominantly hyperactive/impulsive presentation are more frequently accompanied by externalizing problems such as conduct disorder, oppositional defiant disorder, and aggression (28). The fact that self-esteem is more closely associated with internalizing disorders may help explain the relationship between hyperactivity symptoms and self-esteem in adolescents with ADHD, although not directly.

A negative correlation was found between self-esteem and inhibition and working memory scores in children and adolescents with ADHD. In addition, working memory functions were found to be an important predictor of self-esteem in children and adolescents with ADHD. According to these results, low self-esteem is associated with impaired working memory and inhibition in adolescents with ADHD. There are conflicting results in the literature regarding the relationship between working memory and self-esteem. In a recent study evaluating the relationship between executive functions and self-esteem in children and adolescents with ADHD, a positive relationship was found between working memory functions and self-esteem (27). Contrary to these results, Alloway et al. (2009), with the of 308 children aged 5-11 years, it was shown that there is no strong relationship between self-esteem and working memory functions (29). Conflicting findings in the literature may have resulted from differences in sample selection or the methods used to assess working memory and self-esteem. Our results support the results of studies showing a relationship between self-esteem and executive functions in children with ADHD. Besides, it draws attention to the clinical importance of inhibition

and working memory functions in interventions to be developed to strengthen the self-esteem of these children.

While a difference was found in inattention symptoms between adolescents with normal weight and overweight/obese adolescents, no difference was found in terms of self-esteem and ADHD severity. In a few studies, one of which was carried out in our country; When predominantly presentations of ADHD were compared, it was found that there was a higher risk of obesity for the predominantly inattentive presentation compared to the other dominant presentations. (30, 31). It was also pointed out that patients with ADHD may be relatively inattentive to the internal symptoms of hunger and satiety (32). It has been suggested that this situation can be explained by the ADHD-related attention deficit and consequent deficiencies in executive functions, causing difficulties in adhering to a regular eating pattern, and thus the emergence of abnormal eating behaviors.

In a study conducted with the participation of 580 university students with ADHD, the effects of ADHD severity and overweight/obesity on self-esteem, anxiety, depression and stress were examined. In parallel with our results, no significant relationship was found between the presence of overweight/obesity and self-esteem in youth with ADHD (33). Many studies have shown that adolescents with ADHD have lower self-esteem than controls, and that obesity has negative effects on self-esteem. (7, 24, 34). The lack of difference in self-esteem levels between groups with and without obesity in this study can be explained by the low sample size. To our knowledge, this is the first study to examine obesity and self-esteem together in children and adolescents with ADHD. Multicenter longitudinal studies with larger participation are needed to confirm the results of this study.

Many studies from preschool age to adolescence show a negative relationship between overweight/obesity and executive functions, especially inhibition (35-37). According to the results of a comprehensive meta-analysis examining 72 studies evaluating executive functions in overweight/obese individuals, it was reported that while common executive dysfunctions were observed in obese individuals, the most prominent impairments among executive functions in overweight individuals were in inhibition and working memory. (38). In our study, when adolescents with ADHD were compared according to their overweight/obesity status, the working memory, planning organization, metacognition index and global executive scores of the overweight/obese group were higher than the other group. However, no difference was found between the groups in terms of ADHD severity and self-esteem. Similar to our findings, Graziano et al. (2012) in a study conducted with the participation of children and adolescents with ADHD between the ages of 4-18; It has been reported that children with ADHD with poor executive functions have higher BMI scores and are more likely to be classified as overweight/obese compared to children with ADHD with better executive functions (39). However, in a recent study conducted in our country, when children and adolescents aged 6-13 with ADHD were divided into groups according to the 85th percentile, no difference was found between the groups in terms of ADHD severity and executive functions (40). While the results of this study on ADHD severity are similar to our results, the results obtained regarding executive functions are contradictory. This may be due to the differences between the evaluation methods of executive functions.

One of the strengths of this study is that it is the first time that the overweight/obesity, executive functions, self-esteem, and ADHD severity have been examined simultanously in adolescents with ADHD. In addition, considering that variables such as overweight/obesity, executive functions, and self-esteem may also be affected by other psychiatric disorders, the inclusion of only ADHD adolescents can be considered as another strength of the study. The main limitation of this study that should be accepted is the absence of a control group and the examination of overweight and obese adolescents in the same group. In addition, the fact that the sample was collected from a single center and is relatively small makes it difficult to generalize the findings. Finally, the fact that the study is in a cross-sectional model causes no cause-effect relationship between the variables. Our results should be supported by multicenter, broader participation and longitudinal studies.

CONCLUSIONS

When the results of our study are evaluated, it shows that working memory and hyperactivity symptom severity may play an important role in the self-esteem of adolescents with ADHD. While no difference was found in terms of self-esteem and ADHD clinic in adolescents diagnosed with ADHD according to overweight/obesity status, differences in working memory and planning/organization skills

suggest that executive functions play a critical role in overweight/obesity in ADHD. These results, which show that executive functions may have an impact on the psychosocial status of adolescents with ADHD, emphasize the importance of evaluating working memory functions in adolescents with ADHD, both psychosocially and clinically.

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NEÜ YAYINEVİ

Is Glenohumeral Joint Proprioception Affected by Hand Preference?

Glenohumeral Eklem Propriosepsiyonu El Tercihinden Etkilenir mi?

Serdar Arslan[1], Gokmen Yapali[1]

Öz

Amaç: Bu çalışmanın amacı tercih edilen ekstremitenin glenohumeral eklem hareket hissi ile tercih edilmeyen ekstremitenin hareket hissi arasında fark olup olmadığını incelemektir.
Gereçler ve Yöntem: Çalışmaya omuz ekleminde herhangi bir problemi olmayan, sağlıklı, sedanter 20 kişi (10 erkek, 10 kadın) dahil edildi. Katılımcıların yaş ortalaması 23,60±3,64 yıl idi. Veriler 6 Temmuz 2020 ile 3 Ağustos 2020 tarihleri arasında toplandı. Katılımcıların tercih edilen ve tercih edilmeyen taraf glenohumeral eklem hareket hissi ölçüldü. Hareket hissi ölçümü izokinetik dinamometre kullanılarak, 0,1°/s açısal hızda pasif hareket hissi eşik değeri ölçülerek belirlendi. Hareket hissi ölçümleri, 0°'den (0°-IR) ve 30°'den (30°-IR) internal rotasyon yönüne ve 0°'den (0°-ER) ve 30°'den (30°-ER) eksternal rotasyon yönüne olmak üzere 4 yöne doğru yapıldı.
Bulgular: Katılımcıların tercih edilen taraf için internal rotasyon hareket hissi eşik değerleri 0°-IR, 30°-IR açı ve yönleri için sırasıyla 1,27±0,47°, 1,30±0,45°, eksternal rotasyon hareket hissi eşik değerleri 0°-ER, 30°-ER açı ve yönleri için sırasıyla 1,25±0,39°, 1,41±0,32° ve tercih edilmeyen taraf için internal rotasyon hareket hissi eşik değerleri 0°-IR, 30°-IR açı ve yönleri için sırasıyla 1,33±0,59°, 1,37±0,49°, eksternal rotasyon hareket hissi eşik değerleri 0°-ER, 30°- ER açı ve yönleri için sırasıyla 1,39±0,49°, 1,18±0,42° idi. Her iki tarafın pasif hareket hissi eşik değerleri arasında farklar istatiksel olarak anlamlı değildi (p<0,05).
Sonuç: Çalışma sonuçlarına göre tercih edilen ve tercih edilmeyen ekstremitelerin omuz propriosepsiyonu farklı değildir. Omuzu ilgilendiren yaralanmaların rehabilitasyonunda propriosepsiyona ilişkin hedef; yaralanmış ekstremitenin tercih edilen veya tercih edilmeyen olmasına bakılmaksızın sağlam taraf omuz propriosepsiyonuna göre belirlenebilir.

Anahtar Kelimeler: Kinestezi, glenohumeral eklem, tercih edilen el.

Abstract

Aim: The purpose of this study is to examine whether there is a difference between the sense of movement of the glenohumeral joint of the dominant extremity and the sense of movement of the non-dominant extremity.
Materials and Methods: In the study, 20 healthy and sedentary, volunteer participants (10 males, 10 females) who did not have shoulder problems were included. Mean age of the participants were 23.60±3.64 years. Data collection were performed between July 6, 2020 and August 3, 2020. The sense of movement of the dominant and non-dominant extremities of the glenohumeral joint was measured for the participants. The measurement of sense of movement was done by using an isokinetic dynamometer by measuring the passive sense of movement threshold value at an angular speed of 0.1°/s. The sense of movement measurement was performed towards four directions: from 0° (0°-IR) and from 30° (30°-IR) to internal rotation direction, and from 0° (0°-ER) and from 30° (30°-ER) to external rotation direction.
Results: The sense of movement of the participants for internal rotation at angles and directions of 0°-IR, 30°-IR were respectively 1.27±0.47, 1.30±0.45, and for external rotation at angles and directions of 0°-ER, 30°-ER were respectively 1.25±0.39, 1.41±0.32 for the dominant side extremity, for internal rotation at angles and directions of 0°-IR, 30°-IR were respectively 1.33±0,59°, 1.37±0,49°, and for external rotation at angles and directions of 0°-ER, 30°-ER were respectively 1.39±0,49°, 1.18±0,42° for the non-dominant side extremity. For neither of the two extremities, the differences between the sense of movement were statistically significant (p<0.05).
Conclusion: According to the results of the study, shoulder proprioception of dominant and non-dominant extremities is not different. The goal of proprioception in the rehabilitation of shoulder-related injuries; regardless of whether the injured extremity is dominant or not, may be determined according to the shoulder proprioception of the sturdy extremity.

Key words: Kinesthesia, glenohumeral joint, reference hand.

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INTRODUCTION

Neuromuscular control refers to the control of the nervous system over muscle activation. This control is provided by the integration of sensory input (proprioceptive, vestibular, cutaneous, visual, etc.) in the central nervous system. Multisensory input and its integration are vital for the muscular system to function in order (1). Proprioception is afferent information that is collected by mechanoreceptors in muscles, joints and skin and transported to the central nervous system to integrate with other sensory information. In other words, proprioceptive information is sensory input that forms the basis for neuromuscular responses (2). Measuring the activity of mechanoreceptors provides information about proprioception. Detecting the sense of motion in the joint, actively or passively, detecting the angle at which the joint is being actively or passively removed and positioning the joint at that angle again are the most commonly used methods for evaluating the functions of mechanoreceptors (3). Furthermore, the questioning of changes in joint speed and muscle strength provides information about joint proprioception (4, 5). It is claimed that proprioception is affected by certain factors such as age (6), gender (7), physical activity level (8), type of physical activity (9), musculoskeletal injuries (10) and extremity preference (11).

Some studies report that the preference of the extremity affects the proprioception of both the upper extremity and lower extremity joints (12,13). Some research for the upper extremity indicates that the proprioceptive acuity of the upper extremity joints, which is non-dominant, is better. These studies suggest that the non-dominant extremity stabilizes the body, object, etc. during the functions of the dominant upper extremity, which provides this extremity with an advantage in terms of proprioceptive feedback (14,15). On the other hand, there is research suggesting that the dominant upper extremity is better than the extremity, which is non-dominant in terms of dynamic proprioceptive acuity (16,17). As it turns out, the information on this subject is not yet clear. Therefore, the effect of hand preference on upper extremity joints should be investigated more and more thoroughly. In line with this information, the study aims to examine whether there is a relationship between hand preference and sense of motion in the glenohumeral joint (GHJ).

MATERIALS AND METHODS

The study was carried out with the participation of 20 (10 women and 10 men) students from Necmettin Erbakan University Faculty of Health Sciences. Inclusion criteria of the study; It was to be between the ages of 18-25 and to be healthy. Criteria for exclusion from the study were having suffered from shoulder injuries, having undergone shoulder surgery, having general joint laxity and participating in sports (basketball, volleyball) that regularly involve overhead activities, spinal disease related to cervical or thoracic vertebrae, having a disease that concerns the peripheral and/or central nervous system, and using psychoactive or vasoactive drugs.

The research was carried out according to the Helsinki Declaration and the ethics committee approval was taken according to the decision no. 2020/2629 of the Meeting no. 110 dated June 19, 2020. Participants' upper extremity preferences were determined by the Edinburgh Hand Preference Survey (18), whether there was a general joint laxity by the Beighton Scoring (19) and whether there was a musculoskeletal injury involving the upper extremity by the Quick Disability of the Arm, Shoulder and Hand Questionnaire (Q-DASH) (20).

Edinburgh Hand Preference Questionnaire

Edinburgh Hand Preference Questionnaire was used to determine the upper extremity preferences of the participants. It is a questionnaire applied to determine hand preferences, questioning the hand or hands used in performing 10 different hand activities during daily activities, and depending on this, it is used to decide whether the person can use his left hand, right hand or both hands (21). In the Turkish reliability study of the questionnaire, it was stated that the questionnaire had excellent reliability for the Turkish population (18).

The Beighton Score

The presence of general joint laxity of the participants was determined by using the Beighton Score. In this scoring, the lowest score is 0 and the highest score is 9. A total score of 4 and above indicates general joint laxity (19).

The Disability of the Arm, Shoulder and Hand Questionnaire (Q-DASH)

The Q-DASH was used to determine whether the participants had a musculoskeletal injury involving their upper extremities. This questionnaire has been shown to be a valid and reliable questionnaire that measures physical function and symptoms in patients with upper extremity problems, answered by the patient himself, Turkish validity and reliability studies were conducted. It includes 11 topics extracted from

the DASH survey. At least 10 of the 11 items must be answered in order for the Q-DASH score to be calculated. Each title contains 5 answer options, the score of the scale is calculated from the title scores (0, no disability, 100, most severe disability) (20).

Proprioception measurement was performed with an isokinetic dynamometer. Before measurements, the system was calibrated according to the manufacturer's instructions and recommendations (22). For the measurement of proprioception, glenohumeral joint (GHJ) internal (IR) and external (ER) rotation direction sense of motion test was used (23). Initially, each participants was given a comprehensive explanation of the methodology of the study and instructions on the way of communication with the researcher during the tests (22). Participants warmed up for 5 minutes with active range of motion exercises before the tests (24). After the warm-up period, the participant lay on his back on the isokinetic dynamometer device. To reduce sensorial input, the extremity to be measured was inserted into a pneumatic splint and placed on the dynamometer with the elbow at 90º flexion and the shoulder at 90º abduction. Visual and auditory input was eliminated using eye patches and headphones (22,25). First, the measurement of the dominant extremity, then the measurements of the non-dominant extremity were made.

Within the scope of motion sensation measurement, the isokinetic dynamometer (Cybex Humac Norm CSMI, New YORK, USA) passively moved the extremity in the direction of IR or ER at a speed of 0.1°/s. The participant was asked to express whether he felt the motion to the individual who tested the motion as soon as he first felt it. The time between the moment the test started and the moment the participant felt the motion was recorded in seconds. Measurements were made in the following positions and directions; 0º to IR direction, 30º IR position to IR direction, 0º to ER direction and 30º ER position to ER direction. Each measurement was repeated three times, and the average of the three measurements was recorded as a sense of motion test result.

Statistical Analysis

The data was uploaded to the computer environment and analyzed with "SPSS (Statistical Package for Social Sciences) for Windows 22.0 (SPSS Inc, Chicago, IL)". Participants' GHJ sense of motion test results were grouped as dominant and non-dominant extremity. Descriptive statistics were presented as median (interval between quarters), frequency distribution and percentage. The suitability of variables to normal distribution was examined using visual (histogram and probability graphs) and analytical methods (Shapiro Wilk Test). Mann-Whitney U Test was used as a statistical method for statistical signification between two independent groups for variables that did not conform to normal distribution. The level of statistical significance was considered $p<0.05$.

RESULTS

The average age, height and weight of the participants were respectively; 23.60±3.64 years, 1.70±0.11 meters and 71.60±14.05 kilograms. Demographics are presented in Table 1. The proportion of those who preferred their right hand was 80% (n=16), while the proportion who preferred their left hand was 20% (n=4).

Results of sense of motion in 0°-IR, 30°-IR, 0°- ER, 30°-ER angles and directions of participants; were 1.27±0.47, 1.30±0.45, 1.25±0.39, 1.41±0.32 degrees for the dominant extremity and 1.33±0.59, 1.37±0.49, 1.39±0.49, 1.18±0.42 degrees for the non-dominant extremity respectively ($p<0.5$, Tablo 2).

Table 1. Demographic characteristics of the participants

	Age (year)	Height (cm)	Weight (Kg)	BMI (Kg/cm²)
Min-Max	20.00-32.00	156-186	52.00-99.00	17.99-33.08
Mean±SD	23,60±3,64	1.70±0.11	71.60±14.05	24.49±3.34

Table 2. Preferred and non-preferred extremities' GHJ sense of movement

	GHJ of preferred extremity (n=20) Mean±SD	GHJ of non-preferred extremity (n=20) Mean±SD	Z*	p
0°-IR	1.27±0.47°	1.33±0.59°	-0.606	0.579
30°-IR	1.30±0.45°	1.37±0.49°	-0.266	0.796
0°-ER	1.25±0.39°	1.39±0.49°	-0.457	0.684
30°-ER	1.41±0.32°	1.18±0.42°	-0.683	0.529

GHJ: Glenohumeral joint, IR: Internal Rotation, ER: Eksternal Rotation,* Mann Whitney U Test

DISCUSSION

Results of the study planned to examine whether the dominant and non-dominant GHJ sense of motion test results are different in young adult individuals; GHJ joint IR and ER sense of motion showed that the test results were not different.

Visual and proprioceptive feedback is critical for targeted motion (11,26). The central nervous system is familiar with the proprioceptive acuity of both upper extremities during motion. Nevertheless, it controls motion using proprioceptive information of the extremity, which it finds more reliable than proprioceptive knowledge of the two upper extremities it has (27). Deciding which proprioceptive information from the dominant upper extremity or the dominant upper extremity is important, which is depend on which of the learned motion patterns the person uses (28). Nevertheless, it is unclear and confusing at which of the dominant or non-dominant upper extremities the acuity of mechanoreceptors providing proprioceptive input to the upper centers is better. While some studies report that the proprioceptive acuity in the mechanoreceptors of the non-dominant upper extremity is better, others report better proprioceptive feedback from the mechanoreceptors of the dominant upper extremity. Methodological differences such as the measured joint, measured direction of motion and the measured variable can be cited as the cause of this contradiction (14-17).

In the measurement of shoulder proprioception, similar to other joints, it can be used in combination with tests such as sense of position, sense of motion, force reproduction and motion speed reproduction. The most reliable tests to measure the sensitivity of the mechanoreceptors are sense of passive position and sense of motion tests performed isokinetic dynamometer to IR and ER direction in 90 degree abduction (23). Nevertheless, it is known that the sense of motion test is more reliable in testing proprioception due to the fact that it represents afferent proprioceptive sensory processing processes better and it demonstrates the contributions of passive structures to the process better (29, 30). Many of the studies that provided information about dominant and non-dominant shoulder proprioception tried to conclude by examining the results of sense of position tests, but there was no consensus (14-17). Kumar CG S et al. (17) reported that in healthy young individuals, sense of motion acuity of shoulder joint rotation direction was better in the dominant extremity compared to the non-dominant one. Echalier C et al. (16) demonstrated that the results of sense of position tests for the flexion and abduction direction of the dominant extremities for healthy participants between the ages of 16 and 54 were better than that of the non-dominant extremity. Han J et al. (14) tested the proprioceptive acuity of the dominant and non-dominant lower and upper extremities of 12 participants with an average age of 21±4 with active motion extent discrimination apparatus. The researchers reported that the participants' non-dominant extremity results were more successful in shoulder-related tests than dominant extremity results. Schmidt L et al. (15) reported that the non-dominant shoulder active sense of position tests of participants between the ages of 20 and 70 were better than the dominant extremity. As far as we know, there is no research comparing the dominant and non-dominant sense of shoulder motion in healthy individuals. The only study that gave an idea was the study of Allegrucci M and colleagues in which they examined 20 participants with an age average of 18.8±1.3 engaged in upper extremity sports that involve the dominant use of a single extremity. The results of the study revealed that the sense of passive shoulder joint motion in the non-dominant extremity is better than the sense of passive shoulder joint motion in the dominant extremity. It has been suggested that kinesthetic deficits that may have developed in the dominant extremity, which is more commonly used in throwing activity, affect these results (31). The most important and previously unexposed result of this study is that the dominant and non-dominant and undesirable extremity GHJ sense of passive motion test results in healthy young individuals are not different from each other. Depending on this result, it can be inferred that mechanoreceptors that receive the passive joint sense of motion of dominant and non-dominant extremities do not differ functionally. Contrary to the results of previous studies (14-17), which claimed that dominant or non-dominant extremity shoulder proprioception was better based on sense of position tests, it can be concluded that the dominant and non-dominant shoulder proprioception is not different according to the results of this study (14-17). Although methodological differences appear to be the most important reason for this contradiction, the results of the study can also be interpreted as a review of the conclusions that proprioception of the dominant or non-dominant extremity reached by sense of position tests is better.

It has been reported that functional deficiencies caused by glenohumeral joint pathologies are not

affected by hand preference (32-34). Razmjou H et al. (32) reported that preferred side involvement was not associated with higher disability in individuals with glenohumeral joint osteoarthritis. Kelly MA et al. (33) reported that the functional outcomes of preferred and non-preferred-side rotator cuff repair were similar. Lim CR et al. (34) reported that there is no relationship between traumatic shoulder dislocation and hand preference. Current study results suggest that GHE proprioception in healthy shoulders is not different. However, investigating hand preference and proprioception in subgroups with pathological involvement may contribute to a better understanding of the subject. In addition, hand preference, propriceptive acuity and how this is reflected in functional results in individuals with shoulder pathology may be another intriguing issue.

There are some limitations to this study. The fact that the participants are from a certain age group will prevent the generalization of the study results for children and the elderly. Future studies can research the effect of childhood and old age processes on dominant and non-dominant upper extremity GHJ proprioception. Another limitation is that GHJ proprioception is evaluated only by sense of motion test. However, proprioceptive feedback consists of the sum of afferent information from many mechanoreceptors. In future studies, the conclusions on the subject can be strengthened by expanding the work by adding tests such as force reproduction and reproduction of the speed of motion. In addition, the results of the study will be insufficient to reveal how GHJ proprioception is affected after surgery and injuries related to GHJ. New research on this topic will help make the subject more understandable.

CONCLUSION

According to the results of the study, shoulder proprioception of dominant and non-dominant extremities is not different. The goal of proprioception in the rehabilitation of shoulder-related injuries; regardless of whether the injured extremity is dominant or not, the sturdy side can be determined according to shoulder proprioception.

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NEÜ YAYINEVİ

Use of the First Dorsal Metacarpal Artery Flap in Finger Defects – Case Series

Parmak Defektlerinde Birinci Dorsal Metakarpal Arter Flebinin Kullanımı – Vaka Serisi

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Öz

Amaç: Bu çalışmanın amacı, parmaklardaki yumuşak doku defektlerinde FDMA flep kullanımının çok yönlülüğünü değerlendirmektir.
Hastalar ve Yöntemler: Mayıs 2018-Mayıs 2021 tarihleri arasında üst ekstremitede yumuşak doku defekti sebebiyle rekonstrüksiyon yapılan hastalar dosya üzerinden tarandı. Bu hastalardan parmakta defekti olan ve birinci dorsal metakarpal arter flebi ile rekonstrükte edilen hastalar çalışmaya dahil edildi.
Bulgular: 12 hasta çalışmaya dahil edildi. Defektin etiyolojisi tüm hastalarda travma idi. Flep adaptasyonu için 5 hastada tünel açma tekniği kullanıldı. Hiçbir hastada total flep veya greft kaybı yaşanmadı. Komplikasyonlar açısından yaş, cinsiyet, komorbidite, defekt lokalizasyonu, defekt boyutu ve operasyon süresi incelendi. İstatistiksel olarak anlamlı bir fark saptanmadı. Sigara içenler ve içmeyenler incelendi, istatistiksel olarak anlamlı fark bulunmadı. Flep adaptasyonu için tünel kullanımı komplikasyon açısından istatistiksel olarak anlamlı bir fark yaratmadı.
Sonuç: Birinci dorsal metakarpal arter flebi 1. ve 3. parmaklardaki defektlerde oldukça güvenilir bir seçenektir. Tünel tekniği kullanılıyorsa tünel genişliğinin yeterli olduğundan emin olunmalıdır.

Anahtar Kelimeler: Dorsal metakarpal arter, flep, parmak defekti

Abstract

Aim: The aim of this study is to evaluate the versatility of the use of FDMA flaps in soft tissue defects in the fingers.
Patients and methods: Patients who underwent reconstruction due to soft tissue defect in the upper extremity between May 2018 and May 2021 were scanned over the file. Among these patients, patients who had a finger defect and were reconstructed with the first dorsal metacarpal artery flap were included in the study.
Results: 12 patients were included in the study. The etiology of the defect was trauma in all patients. Tunneling technique was used in 5 patients for flap adaptation. No patient experienced total flap or graft loss. Age, gender, comorbidity, defect localization, defect size and operation time were examined in terms of complications. No statistically significant difference was detected. Smokers and non-smokers were examined, no statistically significant difference was found. The use of tunnel for flap adaptation did not make a statistically significant difference in terms of complications.
Conclusion: First dorsal metacarpal artery flap is a very reliable option for defects in the 1st and 3rd fingers. If the tunnel technique is used, it should be ensured that the tunnel width is sufficient.

Key words: Dorsal metacarpal artery, flap, finger defect

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INTRODUCTION

As global rates of mortality decrease, rates of non-fatal injury have increased (1). Hand trauma is one of the most common non-fatal injuries and most of them require specific treatment. They constitute between 6.6% and 28.6% of all injuries (2-4). The main group affected by these traumas are young men who are actively working and most of the traumas occur in the workplace (5). They can be also occur at home, traffic accidents and sports etc (6).

Even minor injuries, if not treated well, can lead to consequences such as decreased quality of life, loss of productivity, chronic pain and limitation of movement (1). Therefore, appropriate repair of defects around the fingers and hand is important. Soft tissue defects in the hands and fingers are no different and can lead to poor outcomes if not treated appropriately.

Hand surgeons should know the reconstructive options for hand injuries with soft tissue defects. Today, there are many reconstruction options for soft tissue defects in the hands and fingers, ranging from local pedicled flaps, distant pedicle flaps and microsurgical flaps such as free flaps, finger transfers and toe-to-thumb (7-10).

The aim of this study is to evaluate the versatility of the use of FDMA flap in soft tissue defects in the fingers.

PATIENTS AND METHODS

The study was planned as a retrospectively. The approval of the ethics committee and also informed consent forms was obtained before surgery from the patients or their legal representatives if necessary (İzmir Katip Çelebi University, Non-Interventional Clinical Studies Instituonel Review Board, Date: 24.11.2022, IRB: 0483). Between May 2018 and May 2021, patients who applied to our clinic with soft tissue defect in the upper extremity and underwent reconstruction were scanned over the file. Demographic data (age and gender), details of the injuries (etiology, affected anatomical area, and defect size), and preferred flaps were examined retrospectively using the hospital's patient data system and archives. Among these patients, the patients whose defect was in the fingers and reconstructed with the first dorsal metacarpal artery flap were included in the study. Patients who did not have a defect in the finger or were repaired with another method, and did not comply with the postoperative recommendations (such as not quitting smoking, wound care recommendations) were excluded from the study. All operations were performed by the senior author. Operations were performed under regional and local anesthesia. The patients were followed up regularly in the postoperative period. During the follow-ups, the patients were photographed, physical examination was performed in terms of flap viability, wound dehiscence, soft tissue infection, and the datas were recorded.

Surgical technique

Debridements were continued until capillary bleeding was observed at the base of the defects and wound lips. When the wound sites were suitable for closure, the dorsal metacarpal artery was marked with hand doppler at the base of the 1st metacarpal bone on the dorsum of the hand. (Figure 1) Skin flap which is not smaller than the defect size was designed on the dorsal surface of the proximal phalanx of the index finger. Drawings were made from the base of the flap to the 1st dorsal metacarpal artery, a skin incision was made in accordance with the drawings, the skin flaps were elevated as a graft. Then, the flap was elevated over the paratenon with a subcutaneous pedicle of at least 1.5 cm width and the flap was obtained as a island flap. (Figure 2) The flap was adapted over the defect under the minimum tension as possible and sutured. Full-thickness skin grafts obtained from the inguinal region or inner arm were used to closure of the donor area. Graft donor areas were primarily repaired. Postoperative wound care, immobilization and elevation were applied for 2 weeks for the grafted defect on the index finger. In order to evaluate the results, the operation areas of the patients were observed clinically during their stay in the hospital,

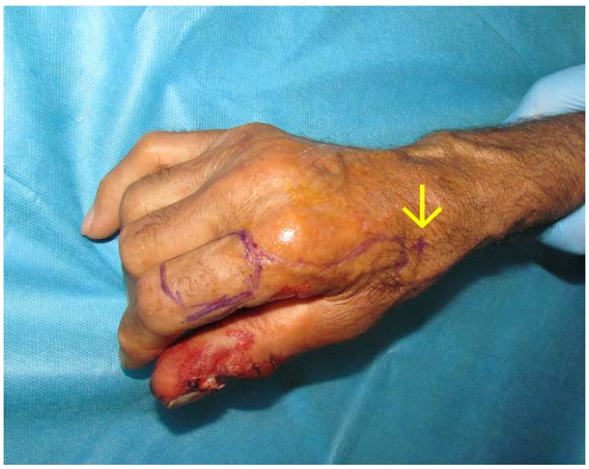

Figure 1. The location of the 1st dorsal metacarpal artery is determined by hand doppler.

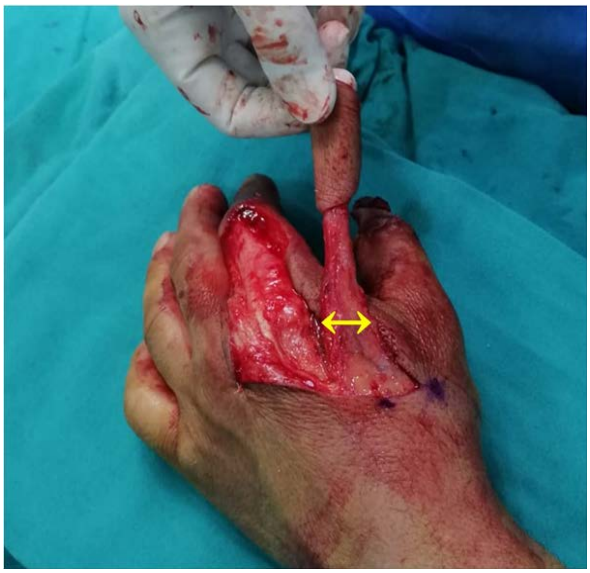

Figure 2. Care should be taken to ensure that the width of the pedicle on the flap is not less than 1.5 cm.

patients were called for controls in the postoperative period, physical examinations were performed, and the datas were recorded.

Statistical analysis

IBM SPSS Statistics Version 20 (IBM, USA) was used for statistical analysis. The Shapiro Wilk test was used for normality analysis. Chi square test was used for binomial values, independent samples T-test and Kruskal Wallis test were used for other values.

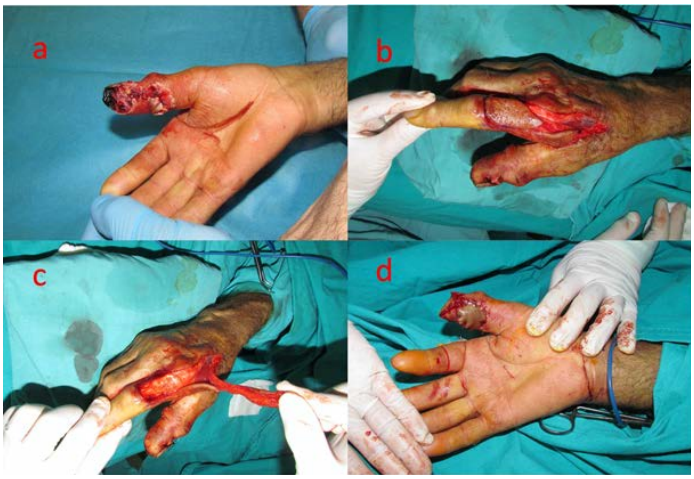

Figure 3. Preoperative and intraoperative views of a patient with a first finger injury.
a: Preoperative view of the defect.
b, c: Views of the prepared and raised flap.
d: View of the flap adapted to the defect.

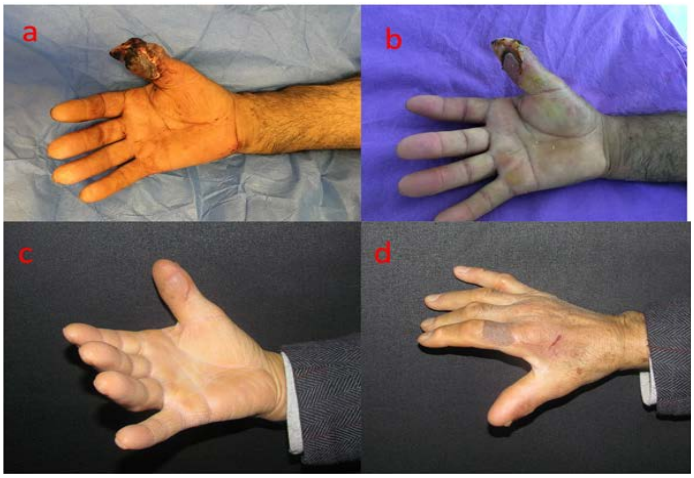

Figure 4. Postoperative views of the patient in figure 1.
a: Postoperative first week view of the patient. There is venous insufficiency in the flap.
b: Postoperative third week view of the patient. Deepitelization occurred in the flap.
c, d: Postoperative 18th month view of the patient.

Statistical significance was set as $p < 0.05$.

RESULTS

A total of 57 patients were followed up due to upper extremity defects. Twelve of these patients were included in the study. Ten of the patients were male and 2 were female. The mean age of the patients was 47.4 years. The mean defect size was 4.3 cm2. 8 of the patients was smoker. 3 patients had diabetes mellitus (DM). The etiology of the defect was trauma in all patients. The defect was located in the 1st finger in 9 patients and in the 3rd finger in 3 patients. Tunneling technique was used in 5 patients for flap adaptation. For donor site repairs, skin grafts were taken from the inner arm in 8 patients and from the inguinal region in 4 patients. The mean operation time was 38.3 minutes. The mean hospital stay of the

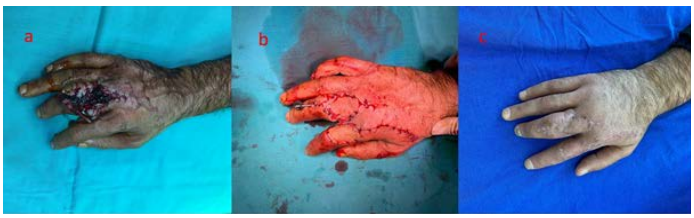

Figure 5. Preoperative, intraoperative and postoperative views of a patient with a third finger injury.
a: Preoperative view of the defect.
b: View of the flap adapted to the defect.
c: Postoperative sixth month view of the patient.

patients was 4.8 days. The mean follow-up period of the patients was 15.4 months. In the postoperative period, local soft tissue infection was detected in 3 patients, and deepitelization was detected on the flap due to venous insufficiency in 4 patients. Extremity elevation was applied to patients who developed venous insufficiency, intravenous steroid therapy was given to reduce edema in the tunnel. All patients were healed with antibiotherapy and appropriate wound care. No patient experienced total flap or graft loss. (Figure 3, 4 and 5) (Table 1)

Age, gender, comorbidity, defect localization, defect size and duration of operation were examined in terms of complications. No statistically significant difference was found. When smokers and non-smokers were examined, no statistically significant difference was found in terms of complications. The use of tunneling for flap adaptation did not make a statistically significant difference in terms of complications. There was a statistically significant difference between the length of hospital stay and complications. (Table 2)

DISCUSSION

First dorsal metacarpal artery (FDMA) flap is a local flap which is supplied by dorsal carpal arch. Traditional use of this flap is soft tissue defects in the fingers and webs. This flap can be also elevated with dorsal sensory branch of the radial nerve as a sensory flap (7).

Earley and Milner (11) reported that the FDMA was absent in only 1.1% of hands in their study. In addition, as shown in anatomical studies, the perforator at the flap site is constant and originates from the branches of the deep palmar arch, even in the absence of the dorsal metacarpal artery (12). In 2010, Bailey et al. (13) succeeded in using the dorsal metacarpal artery

Table 1. Demographic data of the patients.

Patient	Age/Gender	Etiology	Defect size(cm)	Defect location(finger)	Comorbidity	Smoking	Operating time	Tunelization	Greft donor area	Complication	Hospitalization time (day)	Follow up (month)
1	72/M	Trauma	2*2	First	None	+	35	+	Arm	Venous insufficiency	7	18
2	37/M	Trauma	3*2	First	None	+	33	-	Inguinal	None	2	14
3	52/M	Trauma	2*1	First	DM	+	35	-	Arm	Infection	8	15
4	43/F	Trauma	2*2	First	None	-	42	-	Arm	None	2	13
5	38/M	Trauma	3*2	First	None	-	47	-	Inguinal	Infection	7	18
6	50/M	Trauma	3*2	Third	None	+	40	+	Inguinal	None	1	6
7	45/M	Trauma	2*2	First	DM	-	53	+	Arm	Venous insufficiency	6	12
8	51/M	Trauma	2*1	First	None	+	30	-	Arm	Infection	7	15
9	46/F	Trauma	3*2	Third	None	+	34	-	Inguinal	None	2	19
10	23/M	Trauma	2*2	Third	DM	-	32	-	Arm	None	2	17
11	63/M	Trauma	2*2	First	None	+	36	+	Arm	Venous insufficiency	8	22
12	49/M	Trauma	2*2	First	None	+	43	+	Arm	Venous insufficiency	6	16

Table 2. Statistical analysis results in patients.

	Age	Gender	Defect size	Location	Comorbidity	Smoking	Tunelization	Surgery time	Hospitalization
Complication	0.153	0.178	0.065	0.07	0.76	0.719	0.222	0.342	0.012

flap from the previously grafted defective area in their study. In 2011, Isaraj (14) reported that perforator constancy was maintained even in scarred dorsum of the hand. In our study, the presence of dorsal metacarpal artery was confirmed by preoperative hand doppler scanning in all cases. Absence of the first dorsal metacarpal artery was not detected in any patient.

The first dorsal metacarpal artery (FDMA) flap stands out in many ways; it is constant, it is near to trauma zone, simple to raise, single-staged (most of the other treatment options require at least two stages of treatment), early mobilization (decrease risk of contracture and reduced physical therapy time), reduction in hospital stay (increased quality of life, decreased loss of productivity) and minimal donor site morbidity (donor site can be easily closed with a skin graft). As a result of all these, considering the like-to-like principle in reconstruction, this flap is a perfect option for the small to medium size defects (15).

One of the major disadvantage of this flap is that it does not have sufficient pedicle length for the fingers other than the 1st and 3rd fingers. Another down side of the flap is that the venous insufficiency and necrosis (15). Partial necrosis was reported in 2 of 42 flaps in the study of Zhang et al. (16), and in 2 of 10 flaps in the study of Couceiro and Sanmartín (17). El-Khatib (18) reported that venous congestion developed in all flaps in their series of 5 cases, while Couceiro and Sanmartín (17) reported that venous congestion developed in 2 patients in their series of 10 cases. Zhang et al. (16) reported that some degree of venous congestion developed in the flaps in their series. We did not encounter total necrosis in our case series, venous insufficiency developed in 4 of our 12 patients and deepitelization of the flap occurred in these patients.

In this study, 12 patients who had soft tissue defects in the 1st and 3rd fingers of the upper extremity and were repaired with the 1st dorsal metacarpal artery flap were retrospectively analyzed. The defect was in the 1st finger in 9 patients and in the 3rd finger in 3 patients. 3 patients had diabetes mellitus, 8 patients were smokers. Tunneling technique was used in 5 patients for flap adaptation. In all patients, flap donor sites were closed with a full-thickness skin graft. In the postoperative period, soft tissue infection developed in 3 patients and venous insufficiency in 4 patients. All patients recovered with antibiotic therapy and wound care. No major complications such as flap or graft loss were found in any of the patients. The use of tunneling for flap adaptation did not make a statistically significant difference in terms of complications. However, in our clinical observations, we found that deepithelialization was more common after venous insufficiency in cases where tunnels were used to adapt the flap. Because in this patient group, the flap pedicle gets stuck in the tunnel due to edema after the operation and this causes venous insufficiency. The lack of significant results may be due to the small number of patients. We think that larger case series will yield different statistical results. There was a statistically significant difference between the duration of hospitalization and complications, that is, the hospitalization of patients who developed complications took longer, as expected.

The disadvantage of this flap is that it does not have sufficient pedicle length for the fingers other than the 1st and 3rd fingers. In addition, the small number of patients and the use of the flap only in finger defects can be considered as limitations of the study.

CONCLUSION

In conclusion, 1st dorsal metacarpal artery flap is a very reliable option for defects in the 1st and 3rd fingers. If the tunnel technique is using for adaptation, it should be ensured that the tunnel width is sufficient to avoid venous insufficiency and deepitelization in the postoperative period.

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NEÜ YAYINEVİ

The role of the Inflammatory Biomarkers in the Early Prediction of the Severity of Paediatric Acute Pancreatitis

Pediatrik Akut Pankreatit Şiddetinin Erken Tahmininde İnflamatuar Biyobelirteçlerin Rolü

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Öz

Amaç: Yıllardır erişkin literatürünün gölgesinde kalmış olan pediatrik akut pankreatit çalışmaları, pediatrik şiddet sınıflamasının kabul edilmesinden sonra ivme kazanmıştır. Artık hangi hastalarda ciddi hastalık gelişeceğini erken aşamada hızla öngörebilecek inflamatuar biyobelirteçlerin belirlenmesine ihtiyaç vardır. Bu çalışmanın amacı, sistemik immün-enflamasyon indeksi (SII) ve nötrofil-lenfosit oranı (NLO) gibi inflamatuar biyobelirteçlerin erken prediktör olarak etkinliğini değerlendirmek ve eşik değerler belirlemekti.
Hastalar ve Yöntem: 2019-2022 yılları arasında akut pankreatit tanısı alan 53 çocuğun klinik özellikleri, laboratuvar test sonuçları ve görüntüleme bulguları retrospektif olarak değerlendirildi. Hastalar şiddetine göre "hafif" ve "orta şiddetli-şiddetli" olarak iki gruba ayrıldı. Gruplar inflamatuar belirteçler açısından karşılaştırıldı. Hastalık şiddetini öngören faktörler ROC eğrisi analizi ile incelendi. Anlamlı eşik değerler için duyarlılık, özgüllük, pozitif prediktif değer (PPD) ve negatif prediktif değer (NPD) hesaplandı.
Bulgular: NLO ve SII değerleri "orta şiddetli-şiddetli" grupta "hafif" gruba göre istatistiksel olarak anlamlı derecede yüksekti (tümü için p<0,001). NLO≥3.33 (AUC:0.894, %95 güven aralığı: 0.81-0.979, PPD %89.7, NPD %83.3%) ve SII indeksi≥1225.57(AUC: 0.912, %95 güven aralığı:0.831-0.992, PPD %90.0, NPD %87.0) eşik değerlerinin hastalık şiddetini yüksek duyarlılık ve özgüllükle tahmin edebildiği belirlendi.
Sonuç: NLO ve SII pediatrik akut pankreatitte kötü klinik sonucu erken tahmin edebilir. Mevcut çalışma pediatrik akut pankreatitte bu biyobelirteçlerin prognostik öneminini değerlendiren ilk çalışmadır.

Anahtar Kelimeler: Pediatrik ciddi akut pankreatit, sistemik immün-inflamasyon indeksi, nötrofil-lenfosit oranı

Abstract

Aim: Studies of paediatric acute pancreatitis have remained in the shadow of adult literature for many years, and have only increased following the recent acceptance of the severity classification. There is now a need to determine inflammatory biomarkers which will be able to rapidly predict in the early stage which patients will develop severe disease. This study's purpose was to research the efficacy as early predictors and determine cutoff values for inflammatory biomarkers including the systemic immune-inflammation index (SII), and the neutrophil-lymphocyte ratio (NLR).
Patients and Methods: A retrospective evaluation was made of the clinical characteristics, laboratory test results, and imaging findings of 53 children diagnosed with acute pancreatitis between 2019-2022. The study population were separated into groups as 'mild' and 'moderately severe-severe' according to severity. The groups were compared in respect of inflammatory markers. Factors predicting disease severity were evaluated with ROC curve analysis. For the significant cutoff values, positive predictive value (PPV), negative predictive value (NPV), sensitivity and specificity were calculated.
Results: The NLR, and SII values were found to be statistically significantly higher in the "moderately severe-severe" group than in th "mild" group (p<0.001 for all). The cutoff values of NLR≥3.33 (AUC:0.894, 95% CI:0.81-0.979, PPV 89.7%, NPV 83.3%), and SII≥1225.57 (AUC:0.912, 95% CI:0.831-0.992, PPV 90.0%, NPV 87.0%) were determined to be able to predict disease severity with high sensitivity and specificity.
Conclusion: The NLR, and SII are inflammatory biomarkers that can make an early prediction of a poor outcome in paediatric acute pancreatitis. To the best of our knowledge, this is the first study to have evaluated the prognostic importance of these biomarkers, in paediatric acute pancreatitis.

Key words: Paediatric severe acute pancreatitis, systemic immune-inflammation index, neutrophil-lymphocyte ratio

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INTRODUCTION

In recent times, the incidence of pediatric acute pancreatitis (AP) has increased and approached the incidence level reported in the adult age group (1). In the majority of the pediatric age group, the clinical outcome is good. However, together with the increasing incidence, pediatricians are now encountering increasingly more severe disease accompanied by local and/or systemic complications and organ failure (2). Early estimation of the serious disease picture may enable early detection of patients who need referral to the pediatric gastroenterology clinic or intensive care unit. Thus, patients can reach the appropriate treatment conditions in the appropriate center at an early time and the clinical outcome of the patients can be improved (3). Although there are many classifications and scoring systems in adult literature, the search is ongoing for reliable biomarkers that could be easily applied and provide rapid results, which would be able to make an early prediction of cases that would develop severe AP (4,5).

For many years, pediatric studies on the prediction of acute pancreatitis severity have been inadequate. Because, until the North American Society for Pediatric Gastroenterology, Hepatology and Nutrition Pancreas Committee announced the acute pancreatitis severity classification in 2017, there was no universally accepted severity classification for children with AP (2). After the acceptance of this classification, pediatric acute pancreatitis studies gained momentum. In recent years, studies on biomarkers that can predict children who will develop severe AP at an early stage have been continuing. In some of these studies, the predictors evaluated have not been inflammatory biomarkers (6,7). However, AP is a sterile inflammatory disease and a poor prognosis is associated with an uncontrolled systemic inflammatory response (8). Although the predictors recommended in several pediatric studies that have evaluated inflammatory biomarkers support the role of uncontrolled systemic inflammation in severe disease, they do not have the properties of an optimal predictor (9,10). Because, the ideal predictive marker should be low-cost, readily available, easy to apply, and provide results in a short time (11).

The neutrophil-lymphocyte ratio (NLR) and the monocyte-lymphoctye ratio (MLR) are inflammatory indexes that integrate two cell types. They have been defined as inflammatory prognostic biomarkers in some malignant and inflammatory diseases (including acute pancreatitis) in adults (4,5).

The systemic immune-inflammation index (SII), which integrates the neutrophil, thrombocyte, and lymphocyte counts, is a new predictor that reflects the balance between immune and inflammatory statuses. It was first described in 2014 in patients with hepatcellular carcinoma (12). In 2021, it was used for the first time in adult AP patients and was reported to be a better predictor than NLR in the early prediction of the development of severe disease (13).

The NLR, MLR, and SII are ideal biomarkers which can be computed from the laboratory parameters obtained at the time of presentation, are easily accessible, and low cost. No previous study has evaluated the efficacy of these inflammatory biomarkers as early predictors of the development of severe AP in children.

The goal of study was to investigate the utility of inflammatory biomarkers such as NLR, MLR, and SII as early predictors of a severe disease status in pediatric AP, and to determine the cutoff values that can be used for this purpose.

PATIENTS AND METHODS

The study included patients aged <18 years who were treated as inpatients for a diagnosis of AP in Meram Faculty of Medicine Hospital at Necmettin Erbakan University between 2019 and 2022. Ethical approval was obtained from the Ethics Committee of Necmettin Erbakan University Meram Faculty of Medicine (code number:2022/3875).

The patients included in the study were those defined as AP with at least two of the criteria of abdominal pain consistent with AP, imaging findings, and plasma levels of pancreatic enzymes more than three-fold higher than the normal upper limit, as recommended by the International Study Group Of Pediatric Pancreatitis: In Search For A Cure consortium (14).

AP severity was determined according to the 2017 NASPGHAN criteria. To apply this classification, the criteria required are the presence and continuation period of organ dysfunction, and the presence of local and systemic complications (Figure 1) (2). In accordance with the NASPGHAN recommendation, the recommendations of the International Pediatric Sepsis Consensus were used to define organ dysfunction (15).

Similar to the procedures applied in previous adult and pediatric literature, the patients were separated into two groups of “mild AP” (MAP) and “moderately severe-severe AP” (MS-SAP) to differentiate mild

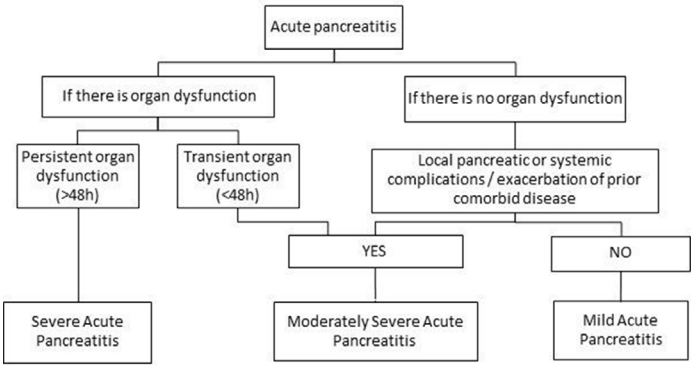

Figure 1. Paediatric acute pancreatitis severity classification of the North American Society for Paediatric Gastroenterology, Hepatology and Nutrition Pancreas Committee

cases from non-mild cases. Patients with moderately severe AP and those with severe AP were combined in one group. For patients with acute recurrent pancreatitis, only the findings of the first pancreatitis attack were included in the analyses. Patients were not included in the study if they had an inflammatory disease that could affect the full blood count parameters (malignancy, FMF, Henoch-Schönlein purpura, etc), or if the data from the first day of the disease were incomplete (Figure 2).

Data were retrieved from the patient records of the demographic characteristics, vital signs, etiology, comorbidities, laboratory test results on first presentation (white-blood cells, neutrophil, lymphocyte and monocyte absolute counts, C-reactive protein, liver enzymes, serum creatinine, glucose, calcium, amylase and lipase), and imaging findings.

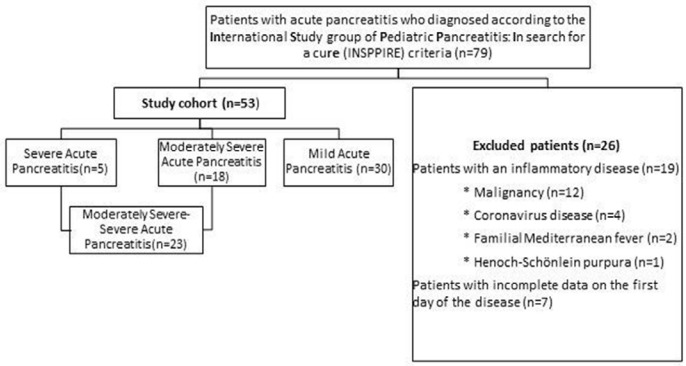

Figure 2. Flow chart of study and inclusion- exclusion criterias

The neutrophil and lymphocyte counts were used to calculate the NLR, and the absolute monocyte and lymphocyte counts to calculate the MLR. The SII was computed with the formula –"neutrophil count x platelet count/lymphocyte count".

Statistical analysis

Data obtained in the study were analyzed statistically using SPSS vn. 20.0 software (IBM Corpn., Armonk, NY, USA). Numerical data were presented as median (Q1-Q3) values, and categorical variables as number (n) and percentage (%). Conformity of the variables to normal distribution was assessed with the Shapiro Wilk test. In the comparisons of categorical variables and frequencies, the Chi-square test was applied. In the comparisons of two groups, the Mann Whitney U-test was applied. To determine relationships between variables, Spearman correlation analysis was used. The decision -making characteristics of the laboratory parameters in the prediction of disease severity were examined with Receiver Operating Characteristics (ROC) curve analysis. The sensitivity, specificity, positive predictive value, and negative predictive value were calculated for cutoff values. Type I error level was interpreted as significant at <5%. 95% confidence intervals were given for Area Under Curve (AUC).

RESULTS

Evaluation was made of 53 patients who met the study criteria, comprising 25 (47.2%) females and 28 (52.8%) males with a median age of 156 months (range, 6-204 months). Comorbidities were present in 10 (18.9%) patients; meningomyelocele in 1 (1.9%), congenital metabolic disease in 2 (3.8%), type 1 diabetes mellitus in 2 (3.8%), nephrolithiasis in 1 (1.9%), cerebral palsy in 1 (1.9%), chronic renal failure in 1 (1.9%), hyperlipidemia in 1 (1.9%), and thalassemia intermedia in 1 (1.9%). The MAP group included 30 (56.6%) patients and the MS-SAP group, 23 (43.4%) patients, comprising 18 (34%) with MSAP and 5 (9.4%) with SAP. No significant difference was detected between the groups in respect of anthropometric measurements and etiology (Table 1).

When the laboratory test results were compared according to disease severity, the white blood cell count (WBC), NLR, monocyte count, MLR, absolute neutrophil count, amylase value and SII value at the time of presentation were determined to be significantly lower in the MAP group than in the MS-SAP group (p=0.02 for amylase, p<0.001 for each of the others). The absolute lymphocyte count was determined to

Table 1. Distribution of the Anthropometric Measurements and Etiology Findings of the Patients According to the Severity Groups

		MAP (n=30)	MS-SAP (n=23)	*p value
Anthropometry	W-H z score (<5 years)	0.46 (-0.73-1.61)	-0.24 (-1.70-1.25)	0.896
Median (Q1-Q3)	BMI z score (>5 years)	0.55 (-0.91-1.16)	0.73 (-1.25-1.81)	0.230
Etiology	Idiopathic	14 (46.7%)	10 (43.5%)	0.374
N (%)	Biliary Pancreatitis	8 (26.7%)	3 (13.0%)	
	Toxic	2 (6.7%)	0 (0.0%)	
	Metabolic	2 (6.7%)	3 (13.0%)	
	Systemic Disease	3 (10.0%)	2 (8.7%)	
	Infection	1 (3.3%)	3 (13%)	
	Congenital Anatomic Malformation	0 (0.0%)	1 (4.3%)	
	Genetic	0 (0.0%)	1 (4.3%)	

*p<0.05 was accepted as statistically significant. Data are stated frequency (percentage) and median (Q1-Q3) values.
W-H: Weight for height, BMI: Body mass index, MAP: Mild acute pancreatitis, MS-SAP: Moderately severe-Severe acute pancreatitis

be significantly higher in the MAP group than in the MS-SAP group (p=0.028). The distribution of the laboratory findings according to disease severity is shown in Table 2.

Correlations were examined between the laboratory findings and the MAP, MS-SAP groups, and there was seen to be a statistically positive correlation between disease severity and WBC count, absolute neutrophil count, NLR, MLR, and SII value (r=0.556 p<0.001, r=0.662 p<0.001, r=0.677 p<0.001, r=0.661 p<0.001, r=0.707 p<0.001, respectively). A statistically negative correlation was determined between disease severity and absolute lymphocyte count (r=-0.315, p=0.022).

The diagnostic rates of the laboratory parameters according to the severity groups were calculated. WBC count, neutrophil count, NLR, MLR and SII (p<0.001 for all) were found to be at extremely good levels of diagnostic rates ort he prediction of the development of severe AP with high sensitivity and specificity. The AUCs of the parameters were interpreted using the criteria reported by Fischer et al (A test with an AUC >0.9, 0.7 to 0.9, and 0.5 to 0.7 indicate high, moderate, and low accuracy, respectively) (16). The AUC values in predicting poor clinical outcomes were 0.912 (95% confidence interval [CI]:0.831-0.992) for SII, 0.894 (95% CI:0.81-0.979) for NLR, 0.885 (95% CI:0.799-

Table 2. Distribution of the Laboratory Findings of the Patients According to the Severity Groups

	MAP (n=30)	MS-SAP (n=23)	*p value
White-blood cells (/mm^3)	7650 (6265-9502)	12400 (11320-16130)	<0.001*
Absolute neutrophil count (/mm^3)	4435 (3045-5942)	10590 (7700-13100)	<0.001*
Absolute lymphocyte count (/mm^3)	2235 (1682-2725)	1700 (1000-2430)	0.028*
Neutrophil/lymphocyte ratio	2.04 (1.38-2.94)	5 (3.46-15.4)	<0.001*
Monocyte count (/mm^3)	490 (300-742)	800 (660-1000)	<0.001*
Monocyte/ lymphocyte ratio	0.20 (0.16-0.32)	0.43 (0.32-0.89)	<0.001*
Eosinophil count (/mm^3)	85 (27.5-140)	30 (0-70)	0.133
Eosinophil/lymphocyte ratio	0.03 (0.01-0.05)	0.01 (0-0.03)	0.963
Basophil count (/mm^3)	20 (3.25-40)	10 (1-30)	0.268
Sedimentation rate(mg/s)	13 (5-22)	14 (8-28)	0.916
Creatinine (mg/dL)	0.62 (0.46-0.75)	0.52 (0.43-0.67)	0.586
Albumin (g/L)	4.6 (4.1-4.82)	4.45 (4.07-4.8)	0.387
Aspartate aminotransferase (U/L)	26 (19-50)	29 (23-38)	0.621
Alanine aminotransferase (U/L)	18 (10-57)	14 (10-66)	0.836
Gamma glutamil transferase (U/L)	16 (10-107)	15 (11.75-44.5)	0.985
Amylase (U/L)	305 (184-487)	640 (237-1340)	0.02*
Lipase (U/L)	475 (258-817)	926(261-2673)	0.069
C-reactive protein (mg/L)	2.5 (1-8.2)	12 (1-46)	0.056
Systemic immune-inflammation index	516.32(380.62-796.30)	1894.73(1301.53-3284.89)	<0.001*

*p<0.05 accepted as statistically significant, Data are stated as median (Q1-Q3) values. Comparisons between groups were made using the Mann-Whitney U-test.
MAP; mild acute pancreatitis, SAP; severe acute pancreatitis

Table 3. ROC curve analysis results according to disease severity

Risk factor	AUC* (95% CI**)	Cut off	p value***	Sensitivity	Specificity	PPV	NPV
WBC (/mm^3)	0.824(0,698-0.949)	10350	<0.001	82.6%	82.1%	85.7%	76.0%
NLR	0.894(0.81-0.979)	3.33	<0.001	87.0%	85.7%	89.7%	83.3%
MLR	0.885(0.799-0.97)	0.3292	<0.001	73.9%	75.0%	79.3%	70.8%
ANS (/mm^3)	0.886(0.779-0.992)	7095	<0.001	87.0%	89.3%	90.0%	87.0%
SII	0.912(0.831-0.992)	1225.57	<0.001	87.0%	89.0%	90.0%	87.0%

*Area under curve **Confidence interval *** p<0.05 accepted as statistically significant
WBC:White blood cells, NLR:Neutrophil lymphocyte ratio, MLR:Monocyte lymphocyte ratio, ANS:Absolute neutrophil count, SII: Systemic immune-inflammation index

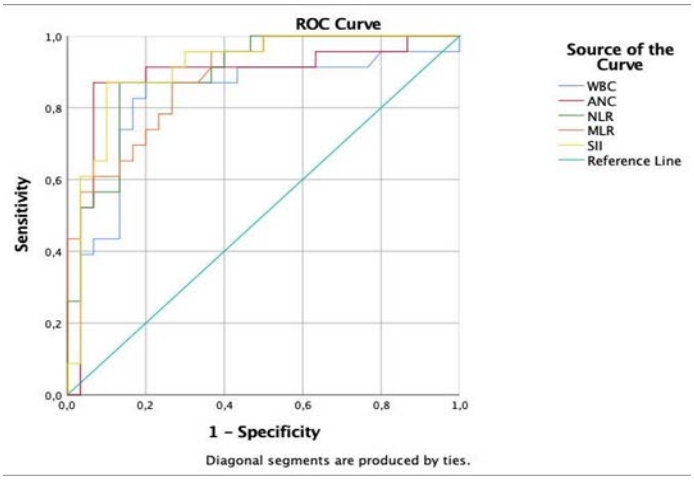

Figure 3. Graph showing the curve obtained with ROC analysis of WBC, absolute neutrophil count, NLR, MLR, and SII values

0.97) for MLR, 0.824 (95% CI:0,698-0.949) for WBC, and 0.886 (95% CI:0.779-0.992) for ANS. The AUC values of parameters indicated high accuracy for SII, and moderate accuracy for others. The results of the ROC curve analysis from which the cutoff values of the laboratory parameters according to disease severity, the area under the curve (AUC) values, and the positive and negative predictive values were calculated, are shown in Table 3 and Fig 3.

DISCUSSION

The results of this study demonstrated that NLR, and SII were significant markers in the development of severe disease in children with AP, and could predict the development of SAP. The diagnostic rates of all the indexes had high sensitivity and specificity in the prediction of SAP, with the SII value having the highest sensitivity and specificity, and the cutoff value for each index was determined. These findings can be explained by the principle of excessive systemic inflammation related to the uncontrolled adaptive and innate immune response in the pathogenesis of AP.

The results showed that at the time of diagnosis, the WBC count, absolute neutrophil count, monocyte count, and amylase value were significantly higher and the absolute lymphocyte count was significantly lower in the MS-SAP group. The increase in neutrophil and monocyte counts and decrease in lymphocyte count have been associated with SAP pathogenesis in previous studies (17).

In the current study, there was determined to be a significant positive correlation between WBC count, absolute neutrophil count and disease severity, and a significant negative correlation between absolute lymphocyte count and disease severity. At a cutoff value of 10350/mm^3 for WBC count and 7095/mm^3 for absolute neutrophil count, the development of MS-SAP could be predicted with high sensitivity and specificity. Although serum WBC count is a component of some adult AP scoring systems, the results reported related to AP prognosis are variable. Silva -Vaz et al. Reported that a cutoff value of ≥14880 mm^3 for WBC count on presentation was a good prognostic tool (3). In a multicentre study by Farkas et al., it was confirmed that WBC count was not sufficient ort he prediction of disease severity (18). Absolute WBC count is the combination of the cells in circulation such as neutrophils and lymphocytes. In a patient with no evident lymphopenia, the WBC count may be high associated with an increase in neutrophil count, and if lymphopenia is very evident, there may be no increase in WBC count (10). Moreover, the distribution of peripheral blood cells and the norms of WBC count can vary according to age in childhood (19). In this study, no statistically significant difference was found between the age distribution in the MAP and MS-SAP groups. Ratios such as NLR and MLR with a composition of two cell types are more stable than WBC and subtype counts and do not have the mentioned disadvantages (5). The cutoff value of 3.33 for NLR determined in this study was found to have extremely

high sensitivity and specificity in the prediction of MS-SAP development. For adults with AP, NLR was first reported by Azab et al. To be valuable in predicting severe disease with a cutoff value of 4.7 (5). These findings were later supported by Jeon et al. With a cutoff value of 4.76 (8). In another study of adults with pancreatitis evaluating the efficacy of ratios obtained from peripheral blood cells, Akoglu et al. Reported that NLR was the best predictor with a cutoff vslue of 5.1. MLR was seen to be statistically significantly different in patients with MS-SAP compared to MAP, but it was not recommended for use as it did not have high sensitivity and specificity (20). In the current study, although the sensitivity and specificity of the cutoff value of 0.32 for MLR was lower than the other biomarkers in the prediction of MS-SAP, it was still extremely high. Although the value of ratios such as NLR and MLR has been shown in inflammatory diseases in the pediatric ort he e, there is no study of children with acute pancreatitis, with which the current study results could be compared. In this study it was determined that both NLR and MLR could be used as independent biomarkers with sufficient sensitivity and specificity in the prediction of the development of MS-SAP. However, the diagnostic rates of NLR were higher than those of MLR.

SII had the highest reliability , in the current study. With a cutoff value of ≥1225.57, SII had extremely good diagnostic rates of the prediction of MS-SAP development. Since SII is the synthesis of 3 cell types involved in the pathogenesis of acute pancreatitis, it is more reliable than rates integrating fewer cell types. The SII was first reported by Zhang et al. İn 2021 to be a poor prognostic marker in adults with acute pancreatitis (21). Subsequently, Liu et al. Reported that the SII could be an early predictor of severe disease with a cutoff value of ≥2207.53 (13). There are few studies of adult patients related to whether or not SII can predict SAP development in acute pancreatitis, and no report could be found of pediatric AP. In several very recent pediatric studies, it has been emphasised that the SII reflects the immune-inflammatory balance. Winker et al. Reported that a decrease in SII in pediatric cancer patients was seen with the anti-inflammatory effect of exercise (22). Guneylioglu et al. Reported that a cutoff value of ≥ 2609 for SII was useful in differentiating children with empyema from those with parapneumonic effusions (23). In another study of infants (1-4 months) with fever of unknown focus, a cutoff value of ≥438.44 could predict the risk of severe bacterial infection (24).

There is no other study showing that the SII can be used to make an early prediction of the development of severe disease in children with AP. The determination of severity within the first 48 hours will be able to improve clinical results by accelerating referral to an appropriate centre, the provision of intensive care support and the appropriate treatment approach (3). However, there are several recent studies related to laboratory parameters which can predict severe disease early in children with AP. Vitale et al. Showed that an increase in BUN could predict SAP development in the early stage (6). Farell et al. Also reported that BUN and albumin values were an independent marker for SAP development (7). However, when the immune-inflammatory pathwways in the pathogenesis of SAP are taken into consideration, it is necessary to evaluate whether inflammatory markers are reliable predictors or not. In a prospective study by Vitale et al. Evaluating biomarkers which could make early predictions of the risk of SAP development in children with AP, it was shown that the matrix metalloproteinase-9 (MMP-9) and tissue inhibitors of metalloproteinase-1 (TIMP-1) levels were significantly higher in SAP patients than in the MAP group (9). In another study, Farell et al. Examined whether interleukin 6 (IL-6) and monocyte chemotactic protein-1 (MCP-1) could differentiate MAP and SAP pediatric patients, and reported that they could predict progression to SAP (10). With these studies, Vitale and Farell emphasised the inflammatory pathways in the early prediction of SAP risk. Ideally, a predictive marker should be low cost, easily accessible and applicable and provide results in the early period (11). Biomarkers such as MMP-9, TIMP-1, and MCP-1 may not be available everywhere and may not provide rapid results. NLR and SII are significant independent variables which can predict MS-SAP development in the early period and can be calculated from the laboratory data obtained on first presentation. Thus, they are ideal biomarkers which can be accessed easily and can provide results in the early period. They can reliably predict the risk of MS-SAP development in children in the early period and can be helpful in improving clinical outcomes. Therefore, NLR (with a cutoff value of ≥3.3) and SII (with a cutoff value of ≥1225.57) can be used with high sensitivity and specificity to predict severe disease that may develop in any patient at the time of presentation.

An important limitation of this study was that there are conflicting reports of the evaluation of WBC and

ANC and cutoff values in the pediatric age group. For example, cutoff values of 10350/mm^3 for WBC or 7095/mm^3 for absolute neutrophil count may not be valid for all age groups. Although high sensitivity and specificity was shown for these parameters in this cohort with no significant difference in the age distributions of the MAP and MS-SAP groups, they cannot be generalised to all age groups. Therefore, because of these above-mentioned disadvantages, WBC and neutrophil count cannot be recommended as parameters ort he prediction of disease severity in childhood. However, these disadvantages are not relevant to the NLR, MLR, and SII, which integrate more than one cell type, and the evaluation of which was the main aim of this study. Other limitations of the study could be said to be the retrospective design, that the biomarkers were only evaluated at the time of presentation, and that the ort he of the disease course were not evaluated. However, the main aim of the study was to detect inflammatory biomarkers that could predict disease severity at the time of presentation. Although the study content served this purpose, there is a need for further prospective controlled studies with larger samples to evaluate patients with high NLR and SII values on presentation. Nevertheless, as the first study on this subject in a pediatric patient group, it can be considered that this study will be of guidance for future prospective studies.

CONCLUSION

NLR and SII are reliable biomarkers ort he early estimation of the development of severe disease in children with acute pancreatitis. As this is the first study conducted on children, there is a need for further large-scale, randomised, controlled studies.

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NEÜ YAYINEVİ

Could Ozone Threapy be a Novel Strategy for Combating with Multi-Drug Resistant Bacteria?

Ozon Tedavisi Çoklu İlaca Dirençli Bakterilerle Mücadelede Yeni Bir Strateji Olabilir mi?

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Öz

Amaç: Çoklu ilaca dirençli (MDR) bakteriler ile oluşan enfeksiyonları kontrol etmek, klinisyenler için büyük bir zorluk haline geldi. Bu nedenle, ana yaklaşım alternatif tedavilerin de gözden geçirilmesidir. Bu çalışmada, gaz formundaki medikal ozonun MDR patojenler üzerindeki antibakteriyel etkisini zamana bağlı olarak değerlendirilmesi hedeflenmiştir.
Gereçler ve Yöntem: Çalışma 21 Mart-15 Nisan 2021 tarihleri arasında Necmettin Erbakan Üniversitesi Meram Tıp Fakültesi Hastanesi Tıbbi Mikrobiyoloji laboratuvarı'nda gerçekleştirildi. On MDR bakteri, yedi farklı bakteriyel konsantrasyonunda (102-108 bakteri/petri) petri kaplarında in vitro olarak değerlendirildi. 10, 20 ve 40 dakikalık 40 ug/ml dozda ozon maruziyeti ve kontrol grubunun MDR bakterileri üzerinde etkinliğini araştırıldı. Plakların 24 saatlik inkübasyonundan sonra, ozonun her maruz kalma süresi için ortalama koloni sayıları belirlendi ve Log10'a dönüştürüldü.
Bulgular: Ortalama bakteriyel azalma (log10) dikkate alındığında, ozonun bakterisidal etkisi 10 dakikaklık maruziyette sadece metisiline dirençli Staphylococcus aureus, VIM-1 üreten Klebsiella pneumoniae ve Karbapenem dirençli Acinetobacter baumannii üzerinde saptandı. Optimum etki, test edilen izolatlarda genellikle 20 dakika içinde gözlendi. Bununla birlikte, MDR-Pseudomonas aeruginosa, ozon maruziyetinden nispeten daha az etkilendi. Ancak 40 dakikalık maruziyet sonunda tüm bakteriler %99'luk seviyenin üzerinde inaktive edildi.
Sonuçlar: Ozon gazı, MDR patojenleri üzerinde etkili bakterisidal aktivite gösterdi ve etkisinin, maruz kalma süresine ve bakteri tipine bağlı olduğu tespit edildi. Pandemi dünyasında mikrobiyal enfeksiyonların kontrolü için yeni yaklaşımlara duyulan ihtiyaç dikkate alındığında, ozon tedavisinin optimizasyonu yüksek öncelikli olarak gerçekleştirilmelidir ve ozonun MDR-bakterilerin inaktivasyonu üzerindeki etkisinin derinlemesine anlaşılmasını desteklemek için daha fazla in vivo çalışmaya ihtiyaç vardır.

Anahtar Kelimeler: Ozon terapi, çoklu ilaca dirençli bakteri, tamamlayıcı tıp

Abstract

Aim: This study evaluated the antibacterial effect of gaseous ozone on multi-drug resistant (MDR) pathogens with regard to time dependency. Controlling infections with MDR bacteria became a big challenge for health care professionals. Therefore, the major corcern should be altered to alternative therapies.
Materials and Methods: The study was performed in Necmettin Erbakan University Meram Faculty of Medicine Hospital, Department of Medical Microbiology Laboratory between 21 March to 15 April 2021. Ten MDR bacteria were evaluated in vitro using in the Petri dishes at seven bacterial concentrations (10^2-10^8 bacteria/dish). The ozone showed its efficacy on these MDR bacteria under the following conditions applied: 40 µg/ml at 10, 20 and 40 minutes and control (no gas was used). After 24 hours incubation of plates, the average colony counts for each exposure time of ozone were figured out and transformed to Log10.
Results: Taking acccount the mean bacterial reduction (log10), the bactericidal effect of ozone was determined on only methicillin-resistant *Staphylococcus aureus*, VIM-1 producing *Klebsiella pneumoniae* and Carbapenem resistant-*Acinetobacter baumannii* in 10 minutes exposure. The optimum effect was generally observed within 20 minutes on tested isolates. However, the MDR-*Pseudomonas aeruginosa* showed a relatively lower response to ozone. All bacteria was inactivated over the level at 99% at the end of the 40 min exposure to ozone.
Conclusions: The gaseous ozone showed satisfactory bactericidal activity on MDR pathogens and its effect depens on exposure time and type of bacteria. Taken into account, the need for new approaches for the control of microbial infections in the pandemic world, the optimization of ozone therapy should be undertaken high priority and more in vivo studies are needed to support in depth understanding of the ozone effect on the inactivation of MDR bacteria.

Key words: Ozone therapy, multi-drug resistant bacteria, complementary medicine.

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INTRODUCTION

Antimicrobial Resistance (AMR) is one of the most critical public health threats of the 21st century as well as it continues being a significant burden for world economy (1,2). However, it is now likely to be hidden by coronavirus disease 2019 (COVID-19) pandemic for some time (1,3). Since the beginning of the antibiotic era, that were considered as novel drugs and saved millions of lives (1,2). Nevertheless, the prolonged and inappropriate use of antimicrobials cause a selective pressure on microorganisms and driving of bacterial resistance (4). The rapid emerge and dissemination of antibiotic resistant pathogens lead to failure in clinical outcomes and also associate with high morbidity and mortality, particulary, in hospital acquired infections (2,5). It was estimated that by 2050, AMR-related deaths would access 10 million (6).

As long as the current pandemic, there are probable threats that could pressure on antimicrobial stewardship policies and cause AMR (5). In the COVID-19 pandemic, it is of vital importance to recover lives of coronavirus disease 2019 patients although this signify appeal to common overmedicate of extensive-spectrum initial antibiotics for threapy or protection of complications such as secondary bacterial infections (5,7). Nevertheless, the extensive use of antibiotics (80%-100%) and antifungals (7.5%-15%) in severe ill COVID-19 patients accepted to intensive care units have been reported by several studies (7,8). It is worried that present mistakes and excesses could increase the advance of the eventual global public health problem by resistance of a great diversity of pathogens to a widespectrum of antimicrobials (5).

Similiar to COVID-19, AMR has been reportedly defined as a significant treat to global public health that “knows no bordes”. Therefore, it is likely to become the current crisis facing all countries across the world (3). In fact, with regard to many specialist, included those from from the World Health Organization, people are now in the verge of post antibiotic era. For this reason, the global neglected issue of AMR requires urgent action and attention (5,9). There is hence a increasing need for both the discovery of new classes of antibiotics, the development of alternative and natural products with pharmacoogical properties to defeat antibiotic resistant bacteria (2,5). Furthermore, once a new drug introduced to the clinic, antibiotic resistance can emerge rapidly by way of intense selective pressure soon after introduction (2,4). In additon, the long term treatment which chemical antimicrobials may have side effect such as nephrotoxicity and neurotoxicity (2,4).

One of the alternative treatment option is ozone threapy. Ozone (O_3) is an unstable triatomic from of oxygen which rapidly convert into water releasing a reactive form of oxygen (11). Ozone has been used for a long time for its antioxidant features, antimicrobial activities as well as its benefical effects on rapid tissue and wound healing (10). In many previous study have also shown that ozone has antibacterial, antiparasitic, fungacidal activities (10-13). For this purpose, in ozone threapy, oxigen-ozone (O_2-O_3) gas mixture called as “medical ozone” (5% ozone in 95% oxygen) has been increasingly utilizied for severe or cronic soft tissue and skin infections as a complementary treatment (14). So that, gaseous ozone (O_3) may be a favorable option in the threapy of infections induced by multi-drug resistant microorganisms and that a compounded treatment has the possible to extend the life of convenient antibiotics as well as to decrease the side effects of chemical antimicrobials depending on intensive usage.

It was aimed to investigate the effectiveness of gaseous ozone on various MDR clinical and reference bacterial strains according to diffirent time interval that need to be optimisation and also highlight the use of local ozone application as the therapeutical alternative to cure infections with MDR pathogens in the present study

MATERIALS AND METHODS

The study was performed in Necmettin Erbakan University Meram Faculty of Medicine, Department of Medical Microbiology Laboratory between 21 March to 15 April 2021. The nature of the study was a prospective, experimental research.

Ethics approval and consent to participate

This study was approved by the Ethics Committee of the Institution (decision number: 2021/3161).

Bacteria cultures and growth conditions

Antibacterial effect is evaluated on three MDR clinical isolates *(Acinetobacter baumannii, Pseudomonas aeruginosa* and *Klebsiella pneumoniae)* and seven reference bacterial strains. Of the six Gram negative strains (OXA-48 producing *Klebisella pneumoniae* (NCTC 13442), VIM-1 producing *K. pneumoniae* (NCTC 13440), KPC producing *K. pneumoniae* (CCUG 56233), NDM-1 producing *K. pneumoniae* (NCTC 13443), IMP producing *E. coli* (NCTC 13476), mcr-1 producing *E. coli* (NCTC 13846) and one

Gram positive strain (MRSA, ATCC BAA-1720) were provided from the bacteriology culture collection of the our Laboratory of Microbiology. The identification of the clinical isolates and antibiotic susceptibility testing were applied via conventional technicals and automated system (Vitek 2, bioMerieux, Marcy lâ€™Etoile, France).

Plating method; All isolates were plated onto blood agar (bioMerieux, Marcy lâ€™Etoile, France) and incubated in aerobic conditions at 37 °C for 24 hours, then suspended in distilled water. Bacteria suspensions were adjusted to 0.5 McFarland standard turbidity corresponding to approximately 1-2 × 10^8 colony forming units (CFUml^{-1}) via a densitometer (DENSICHEK® PLUS, bioM erieux, Marcy lâ€™Etoile, France). Prepared decrimal serial dilutions are transferred to sterile U tubes with a final concentration equaling to 1×10^2 CFU ml^{-1} The sample taken from each of the tubes was inoculated onto blood agar and then the inoculum was spread on each plate surface.

Ozone application

A commercially available ozone (O_3) generating system device (Hyper-Medozon Comfort; Herrmann Apparatebau GmbH, Kleinwallstadt, Germany) was used by the manufacturer's protocol to obtain medical ozone. The ozone dose and the gas flow were checked simultanously recommended by the Standards Committee of the International Ozone Association (IOA). In order to secure reproducibility of the findings, this study was performed in an environment with checked temperature at 25 °C and each experiment was performed in triplicate. The agar-blood in Petri dishes, which is optimum material under laboratory conditions, was used as the culture medium in this study. The plates were divided into two main groups-control (CG: not gas applied) and treated (TG). The TG plates with opened covers were inserted into the sterilized ozone-resistant plastic bags on flat cardboard suface. After fixation and sealing of the pastic bag with a special strap, the air is completely removed from the bag and later the bag is fiiled with the ozone gas mixture at 40 µg/ml concentrations and three exposure times (10, 20, 40 min). The flow of O3 was kept fixed at 1L/min in all tests. After ozone application, the TG plates were removed with the CG plates for incubation in aerobic conditions at 37 °C for 24 hours. Then the bacterial colony count in each plate was evaluated and the rate of remaning colonies was statistically analyzed. Colony-forming units on blood agar were counted. Also the colony number of the petri dishes which was not ozone gas applied was used to calculate killing rate. Log_{10} bacterial reduction factor (RF) and kill percentage (% kill) was calculated by using an equation presented in ASTM E2315[15]. RF= Log_{10} (control) − Log_{10} (treated) (where control is the number of colonies recovered from the unexposed and treated is the number of colonies recovered from the exposed to O_3) Killing rate (%) = (CFU of the control − CFU of the test) /CFU of the control)

Statistical analysis

The frequencies, ratios, mean and standard deviations of the bacteria in the groups in terms of different variables are presented with descriptive statistics. Whether the distributions of the research variables encounter the normality assumption was examined by using both skewness and kurtosis values and histograms. The evaluation of results showed that the research variables provide the normality assumption. The Kruskal-Wallis H test and the Mann-Whitney U test were used for intergroup comparisons. The changes in continuous variables measured in different time periods were tested with the Friedman F Test and Wilcoxon. The significance level for all analysis results was determined as $p < 0.05$. In this study, data analysis was performed using SPPS 25 (IBM Corp. Released 2017. IBM SPSS Statistics for Windows, Version 25.0. Armonk, NY: IBM Corp.) program.

RESULTS

All tested groups showed the reduction of bacterial colonies and there was significant different between ozone treated and control groups (p=0.000). It was determined that there was significant different between ozone treated groups (10 min p=0.000; 20 min p=0.000; 40 min p=0.000, exposure time) by Kruskal Wallis test. Especially at the concentration of 10^8 mL^{-1}, the greatest decrease was detected in the first 10 min. (Table 1). However, the same rate of decrease was not observed in continued exposure times at the concentration of 10^8 CFU mL^{-1} The lowest difference between the control and 10 min was at the concentration of 10^5 cfu/mL. Besides, the difference was greatest in the group with the 10^5 CFU mL^{-1} concentration between 10 min. and 20 min. ozone exposure. In addition, it was found that the bacterial reduction at the 10^8 and 10^7 CFU mL^{-1} concentrations were significantly higher than all other concentrations between 20 min- 40 min ozone exposure (Table 1).

Similarly, the bacterial log_{10} reduction was observed greater at high initial concentrations than

Table 1. Evaluation of bacterial reduction (log_{10}) according to exposure times and bacterial concentrations

CONC	n	C-10 min		10 min. - 20 min.		20 min.- 40 min.	
		Mean	SD	Mean	SD	Mean	SD
10^2	30	2.72	0.75	0.20	0.76	0.00	0.00
10^3	30	3.30	1.12	0.30	0.92	0.76	0.14
10^4	30	3.33	1.51	1.02	1.48	0.92	0.17
10^5	30	2.17	1.42	2.25	1.61	0.85	0.16
10^6	30	2.74	1.24	1.73	1.67	1.58	0.29
10^7	30	3.07	0.83	1.19	1.66	1.77	0.32
10^8	30	3.26	0.11	0.74	0.90	2.09	0.38
Total	210	2.94	1.15	1.06	1.49	1.53	0.11
		p < 0.001		p < 0.001		p = 0.01	

C: Control, CONC: Concentration, n: number, SD: Standard deviation

lower concentrations at the end of 40 min. For instance, the avareage reduction was detected at 10^2 initial concentration approximately 2.75 log CFU mL^{-1} in within 40 min for MRSA isolates whereas it was detected at 108 concentration nearly 3.18 log CFU mL-1 in first 10min. and ~ 8.18 log CFU mL^{-1} within 40 min. However, the killing rates of MRSA were similar both high and low concentrations at the level of nearly over> 99.9% in all exposure times. Furthermore, the killing rate was detected higher within 10 min. at greater bacterial concentrations than lower concentrations in MDR-*P. aeruginosa*. The average killing rate at 10^8 CFU mL-1 was 99.9% whereas it was determined as 33.3% for this bacteria. The mean log_{10} reduction of all bacteria with times of exposure was illustrated in Figure 1a. Even though the aveage bacterial reduction was over 5 log units in which meant revealed the decent bactericidal activity, the whole of bacteria could not inactivate in high inoculum concentrations (Figure 1b.)

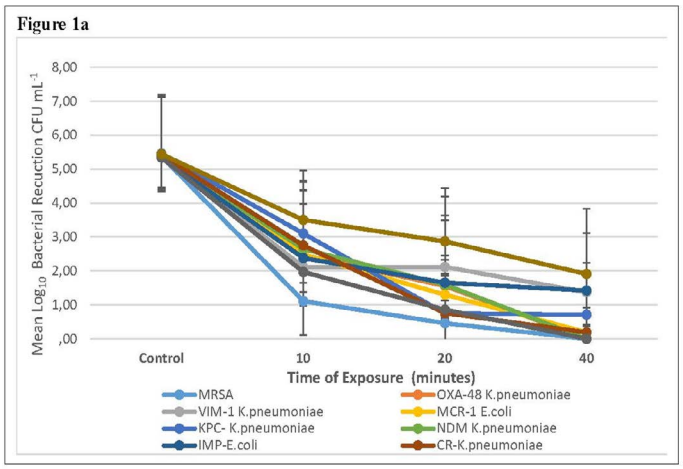
The greatest decrease was detected in methicillin-resistant *S. aureus* (MRSA) between control and 10 min. ozone treated group (TG) (Table 2). The decrease in MRSA was significantly higher than the decrease in all other bacteria (the statistical significance between *OXA-48 K. pneumoniae p=0.001, VIM-1 K. pneumoniae p=0.036, mcr-1 E.coli p=0.001, KPC-K. pneumoniae p=0.000, NDM- K. pneumoniae p=0.000, IMP E. coli p=0.001, CR-K. pneumoniae p=0.000, MDR-P. aeruginosa p=0.000 except CR-A. baumannii (p=0.110)* after first 10min ozone exposure. Nevertheless, the decrease in *CR-A. baumannii* was significantly higher than only the reduction in *KPC-K. pneumoniae* and *MDR-P. aeruginosa*. It was observed that the decreases in *KPC-K. pneumoniae* between 10 and 20 min were significantly higher than the decreases in MRSA, OXA-48 *K. pneumoniae*, VIM-1 *K. pneumoniae*, *IMP E. coli* and MDR-*P. aeruginosa*. The reduction in *CR-K. pneumoniae* was significantly higher than the decrease in VIM-1 *K. pneumoniae*.

In the measurement of the 10 min. ozone treatment, *KPC-K. pneumoniae* (p= 0.038) and *MDR-P.*

Table 2. Comparison of logarithmic bacterial reduction according to bacterial species depending on exposure time to ozone gas

Type of Bacteria	n	C- 10 min.		10 min.- 20 min.		20 min. – 40 min.	
		Mean	SD	Mean	SD	Mean	SD
MRSA	21	4.27	1.34	0.65	1.59	0.46	1.45
OXA-48 *K.pneumoniae*	21	2.92	1.08	0.91	1.47	1.56	2.07
VIM-1 *K.pneumoniae*	21	3.25	1.03	0.00	0.00	0.75	1.40
mcr-1 *E.coli*	21	2.94	0.95	1.17	1.53	1.11	1.83
KPC-*K.pneumoniae*	21	2.36	0.93	2.35	1.81	0.03	0.15
NDM-*K.pneumoniae*	21	2.73	0.90	1.06	1.33	1.59	1.90
IMP-*E.coli*	21	2.96	0.92	0.74	1.17	0.23	0.39
CR-*K.pneumoniae*	21	2.67	0.99	1.99	1.53	0.57	1.43
CR-*A.baumannii*	21	3.37	0.79	1.11	1.40	0.86	1.59
MDR-*P.aeruginosa*	21	1.94	0.99	0.64	1.03	0.95	1.39
Total	210	2.94	1.15	1.06	1.49	0.81	1.53
p	210	p< 0.001		p< 0.001		p= 0.008	

C: Control, CONC: Concentration, n: number, SD: Standard deviation



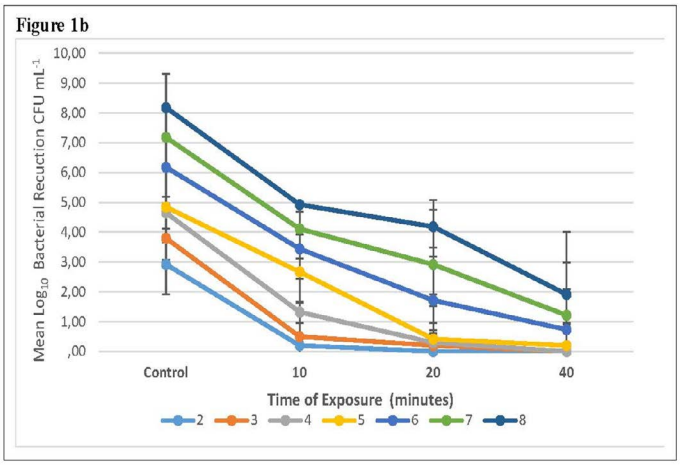


Figure1a. The Mean ± SD $\log_{10}$ reduction in bacterial cell counts in the culture medium was give in Figure1a. belonging to each different bacteria. Colony-forming units: CFU

Figure1b. Each experiment was repeated 3 times. The Mean ± SD $\log_{10}$ reduction in bacterial cell counts in the culture medium. The average $\log_{10}$ bacterial reduction at the different concentrations 10^2-10^8 CFU mL^{-1} with gaseous ozone of 40 µg/ml for 10, 20 and 40 minutes exposure. Colony-forming units: CFU

aeruginosa (p= 0.004) were found to be more resistant than MRSA. VIM-1 *K. pneumoniae* was also obtained to be more resistant than MRSA (p= 0.010), *OXA-48 K. pneumoniae* (p= 0.010), *NDM- K. pneumoniae* (p= 0.010) and *CR-A. baumannii* (p= 0.010). It was determined that some bacterial species did not show time-dependent killing continuity. For example, 3.24 $\log_{10}$ bacterial reduction was observed in the first 10 min of ozone exposure in VIM-1 *K. pneumoniae*, while no difference was observed between 10 min and 20 min. Additionally, approximately 1 $\log_{10}$ bacterial reduction was also appointed at the end of the 40 min (Table 3).

Gaseous ozone was showed bactericidal effect (>=3$\log_{10}$ bacterial reduction) on MRSA, VIM-1 *K. pneumoniae* and *CR-A. baumannii* in 10 min exposure time and the bacterial decresases were detected as 4.27 $\log_{10}$ CFU mL^{-1}, 3.24 $\log_{10}$ CFU $mL^{-1}$and 3.37 $\log_{10}$ CFU mL^{-1}, respectively. The least bacterial decrease (1.93 $\log_{10}$) in 10 min ozone exposure was observed in *MDR-P. aeruginosa*. At the 20 min ozone exposure, the bactericidal activity was detected on all tested bacteria except MDR-*P. aeruginosa* (2.57

Table 3. Determination of logarithmic bacterial inactivation at the different time exposure to ozone for each bacteria

Type of Bacteria	n	Control		10 min.		20 min.		40 min.	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD
MRSA	21	5.38	1.82	1.11	2.05	0.46	1.45	0.00	0.00
OXA-48 *K.pneumoniae*	21	5.39	1.79	2.47	2.25	1.56	2.07	0.00	0.00
VIM-1 *K.pneumoniae*	21	5.36	1.81	2.12	2.29	2.12	2.29	1.37	2.01
mcr-1 *E.coli*	21	5.42	1.76	2.48	2.06	1.30	1.91	0.19	0.87
KPC- *K.pneumoniae*	21	5.46	1.74	3.10	1.86	0.75	1.58	0.71	1.52
NDM *K.pneumoniae*	21	5.38	1.81	2.65	2.00	1.59	1.90	0.00	0.00
IMP-*E.coli*	21	5.34	1.78	2.38	2.00	1.65	1.98	1.42	1.70
CR-*K.pneumoniae*	21	5.42	1.75	2.75	1.72	0.76	1.61	0.19	0.87
CR-*A.baumannii*	21	5.34	1.83	1.97	2.00	0.86	1.59	0.00	0.00
MDR-*P.aeruginosa*	21	5.44	1.75	3.50	1.12	2.87	1.57	1.91	1.92
Toplam	210	5.39	1.75	2.45	2.01	1.39	1.91	0.58	1.36
p	210	p = 1.000		p = 0.017		p = 0.001		p < 0.001	

CONC: Concentration, n: number, SD: Standard deviation

log_{10} CFU mL^{-1} bacterial reduction). Finally, it was determined that the 40 min ozone exposure showed bactericidal effect on all tested isolates including *MDR-P. aeruginosa*.

DISCUSSION

As more antibiotics are losing their activity to MDR microorganisms, the major corcern should be altered to alternative therapies. It is essential to enhance reseach into new and natural strategies to deal with infectious disease. As well as it should be aimed at decreasing the expand and transmission ratio for these pathogens, via contact either between people or between people and settings /surfaces/medical equipments by new research in this pandemic world (16,17). The bactericidal impact of gaseous ozone is well recognized so far (10). Meanwhile, there is a lack of studies aiming the effectiveness of ozone against MDR pathogens, particularly carbapenemase-producing bacteria. In this assay, the antimicrobial activity of gaseous ozone against ten multi-drug resistant bacteria was investigated. The gaseous ozone showed its efficacy on MDR Gram negative and positive bacteria under the following conditions applied: 40 µg/ml at 10 min, 20 min and 40 min. The results demostrated that ozone was effective against all tested bacteria. Nevertheless, the optimal effect was observed with a dose of 40 µg/ml and within 20 min except *MDR-P. aeuginosa*.

Ozone is a potent biocidal agent, capable of inactivating several pathogens including Gram (-) and Gram (+) bacteria, fungi or viral capsids. The Ozone (O3) can be applied as a bactericidal agent in the forms of ozonized water or oil, ozone associated with other substances and more principally the gaseous O_3/O_2 gas mixture. The inactivation or reduction of microorganisms depends on ozone concentration, type of pathogens, initial bacterial load and time of exposure (18).

It is immensely significant to underline that the greater activity detected of the action of gaseous ozone in higher concentrations of bacterial inoculum in the presented study. This situation can be clarified that the greater is the inoculum, the higher is the colony forming unit in the control plates. Therefore, Log_{10} is greater at higher inoculum concentrations of microorganisms and is minor at lower concentrations. For this reason, it is required to interpret the results of Log_{10} and killing ratio together, pointing that the reduction was mostly 100%, regardless of the inoculum used (16).

Although the sensitivity of bacteria to ozone gas at the same concentration varies, it has been determined that meticilin resistant *S. aureus*, which is a Gram-positive bacterium, is more sensitive than Gram-negative bacteria, and the inactivation is provided faster and at a higher rate in this study. These findings are similar to those determined by Giuliani et al. (19) and Hirai (20) who, in their studies on the effect of ozonized water on various types of bacteria, described that the effect of the ozone applications was greater in the action on Gram-positive bacteria. Komanapalli et al. (21) notified that O_3 affects proteins easier and faster than lipids. Therefore, Gram positive bacteria may be more likely sensitive to ozone. The MRSA was inactivated at the level of >99% within 10 min in this study. On the contrary, Azuma et al. (22) reported that the MRSA was inactivated gradually: 36% after.1 min, 79% after 5 min, and 83% after 10 min. contact to ozone.

The mean colony counts for each exposure time of gaseous ozone were figured out and transformed to Log_{10}. The logarithmic inactivation of *MDR-P. aeruginosa* was lower than the results detected for other Gram negative bacteria at the all tested exposure time.

The bactericidal activity was generally observed within 20min. on tested isolates. However, the *MDR-P. aeruginosa* showed a relatively lower response to ozone and it was observed that the bacterial reductions within 10 min., 20min., 40min. as 1.93 Log_{10} CFU mL^{-1}, 2.57 Log_{10} CFU mL^{-1}, 3.53 Log_{10} CFUmL^{-1}, respectively. So, it was required more than 20min. exposure to ozone for the bactericidal effect. Although the bactericidal efficiency was reached to relatively adequate level, an average ~1.91 log CFU mL^{-1} *MDR-P. aeruginosa* population could survived after the 40 min. ozone application. These results suggested different mechanisms of pathogens to deal with the bactericidal effects of gaseous ozone. A previous assay indicated a selection of a robust bacterial population through ozonation, which is defined by a high guanine-cytosine (GC) content of their genomes (23). The weaker results obtained for MDR-*P. aeruginosa* in the current study may be related to high GC-contents >60% belonging to this species (24). Similiar to this work, Andreani et al. reported that *S. aureus, E. faecalis E. coli, S. mutans* and *S. typhi* were highly sensitive to ozone at a concentration of $1x10^2$ CFU mL^{-1}, presenting a decrease of viable cells varing from 45 to 80 % within 30 min of exposure to ozone. On the other hand, *P.*

aeruginosa was inactivated in the same conditions by only 25 % of the initial bacterial load (25).

The potential of our results is interesting. It was investigated whether the antibacterial activity of ozone was affected by the types of resistance genes carried by the bacteria. It was determined minimal difference in the time dependent inactivation among the three different type of carbapenemase producing (blaOXA-48, blaKPC or blaNDM^{-1}) *K. pneumoniae* isolates. No distinct differences were noted at the first 10min for these bacteria. However, the net log reduction of *KPC-K. pneumoniae* was significatly higher OXA-48 *K. pneumoniae* within 10 min-20 min. In addition that the bacterial decrease of OXA-48 *K. pneumoniae* was significantly higher than both of them within 20min-40min. Nevetheless, all of the three isolates were in activated at the level of >90% and the bactericidal effectiveness was detected within 20 min. A consistently a longer exposition time might conceivably end in a higher inactivation ratio (26). However, some bacterial species could not represent time-dependent inactivation contionusly (27). Taking the results of VIM-1 *K. pneumoniae* strains, no distinct differences were noted between the effects of exposure to gaseous ozone for 10 vs. 20 min.

No significant differences occured between IMP-1 producing *E. coli* and mcr-1 carring *E. coli* the effects of exposure to gaseous ozone; the average log reduction within 10 min was 2.93 and 2.96 log units, respectively. Meanwhile, the bactericidal activity was observed within 20 min both of them. The carbapenemase-producing *A. baumannii* isolates were also found considerably sensitive to ozone, reduction rates greater than 4 log units were revealed at first 20 min. Similiarly, Mark et al (28) reported that ozone could be a promising agent to perform disinfection of surfaces contaminated with carbapenemase producing *A. baumannii* under room conditions. Inactivation rates higher than 5 log units were observed on all stainless steel and ceramic carriers after ozon contact (80 ppm ozone; 60 min.). Song et al. (29) investigated the clinical safety and efficacy of topical ozone in two patients with MRSA skin infection. These authors reported that almost 100% MRSA and 100% *S. aureus* a were inactivated by ozonated water in 1 min. Yasheng et al. (30) perfomed a combination of ozonated water and conventional treatment on eighteen patients with chronic osteomyelitis and gained good clinical outcomes. In a study of Oh et al. (31) the ozone could decrease antibionicrobial resistant bacteria and their resistance genes by more than 90% even at 3 mg/L ozone concentration. In another study, All of carbapenem resistant *Enterobacteriaceae,* MRSA, vancomycin-resistant *Enterococcus spp., MDR Acinetobacter spp.* and *MDR P.aeruginosa* were inactivated at the level of >90% only within 10 min. Interestingly, antimicrobial sensitive bacteria (AMSB) represented similar patterns to ozonation in the same study (21). On the contrary, in a study of Lüddeke showed that antibiotic resistant *E. coli* and staphylococci virtually survived ozone exposure better than AMSB (32).

Overall, the bactericidal effects of ozone strongly depend on the bacterial species. Some facultative bacteria are capable of different levels of resistance to ozone oxidative stress to survive. Therefore, the factors affecting the sensitivity to ozone should be clarified by futher deep studies.

In general, the diversity of the available literature data, in addition to the various methodological strategies performed in the different assays, complicate the exact assesment of the efficiency of the ozone implementations. This study includes the following limitations: during the experiment ozone dose was stable, so we could not evaluate the ozone dose dependent efffect. The nature of the study, it is not clear evident how well our results may convert into clinical practice in which parameters such as variable blood flow, necrotic tissue, and great bacterial loads may play a significant role, especially in the soft tissue infections.

CONCLUSIONS

Given the results exposed, gaseous ozone showed adequate bactericidal activity on MDR bacteria and its effect increased dependently exposure time. The results of these studies clearly underline the necessity of properly optimizing the ozone practices (e.g. specific ozone dose, exposure time) considering both the bacterial species and related antibiotic resistance profiles, as well as physico-chemical properties, safety corcern to combat infections with multi-drug resistant bacteria. Bearing in mind the necessity for novel approaches for the control of microbial infections, the optimization of ozone treatment should be taken high priority and more further in vivo studies are needed to provide in depth understanding of the ozone effect on the inactivation of antibiotic resistant bacteria in the current pandemic world.

Informed consent

Nature of the study, it is not required patient

consent form.

This study conformed to the Helsinki Declaration. The study was approved by the ethic review board from Necmettin Erbakan University Faculty of Medicine (decision number: 2021/3161)

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NEÜ YAYINEVİ

Efficacy of Convalescent Plasma Therapy in COVID-19 Patients

COVID-19 Hastalarında Konvelesan Plazma Tedavisinin Etkinliği

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Öz

Amaç: Corona Virüs 2019 Hastalığı (COVID-19)'nda şu ana kadar spesifik antiviral ajan olmamasına rağmen tedavi için konvelesan plazma (CP) tedavisi tedavi için kullanılmıştır. Ancak CP tedavisinin prognoz ve mortalite üzerindeki etkinliği halen tartışma konusudur. Bu çalışmada COVID-19 hastalığında CP tedavisinin etkinliğine ilişkin deneyimlerimizin paylaşması amaçlandı.
Hastalar ve Yöntem: Çalışma Mayıs 2020-Şubat 2021 tarihleri arasında standart tedaviye ek olarak CP tedavisi alan 126 COVID-19 tanılı hastada gerçekleştirildi. 126 hasta ilk beş gün içinde (Grup A) ve beş günden sonra (Grup B) CP uygulananlar olarak iki gruba ayrıldı. Bu iki gruptaki hastalar laboratuvar parametreleri, klinik bulgular ve mortalite açısından değerlendirildi.
Bulgular: Toplam 126 hasta Grup A'da 86 hasta ve Grup B'de 40 hasta) tespit edildi. 119 (%94.4) hasta şifa ile taburcu olurken 7 (%5,5) hasta kaybedildi. Ortalama hastane yatış süresi Grup A'da 11.4±0.7, Grup B'de 18.4±1.7 gün olarak bulundu (p<0,001). Lenfosit, PLT, fibrinojen ve CRP'nin tedaviye bağlı ana değişim etkisi istatistiksel olarak anlamlıydı (p<0.001). Ancak, iki grup D-dimer açısından karşılaştırıldığında sonuçlar marjinal olarak anlamlıydı. Basit etki değerlendirildiğinde; Grup A'daki değişim anlamlı değilken, Grup B'deki değişim anlamlıydı. CP tedavisine 5 gün önce veya 5 gün sonra başlanması laboratuvar parametrelerini değiştirmedi. Ancak, D-dimer'daki değişim marjinal olarak anlamlıydı (p=0.058).
Sonuç: Çalışmamızda CP tedavisine erken başlamanın hastanede kalış süresini azalttığı ancak mortalite ve laboratuvar parametreleri üzerine etkisinin olmadığı gösterildi.

Anahtar Kelimeler: Konvelesen plazma, COVID-19, SARS-CoV-2

Abstract

Aim: Convalescent plasma (CP) therapy has been used for treatment, although it has not been Corona Virus 2019 Disease (COVID-19) specific antiviral agent so far. However, the effectiveness of CP treatment on prognosis and mortality is still a matter of debate. In this study, we aimed to share our experiences about the effectiveness of CP treatment in COVID-19.
Patients and Methods: The study was conducted in 126 patients diagnosed with COVID-19 who received CP treatment in addition to standard treatment between May 2020 and February 2021. 126 patients were divided into two groups as those who underwent SP within the first five days (Group A) and after five days (Group B). The patients in these two groups were evaluated in terms of laboratory parameters, clinical and mortality.
Results: A total of 126 patients were identified (86 patients in Group A and 40 patients in Group B). 119 (94.4%) patients were discharged with recovery, 7 (5.5%) patients died. The mean days of hospitalization were found to be 11.4±0.7 in Group A and 18.4±1.7 in Group B (p<0.001). Treatment-related lymphocyte, PLT, fibrinogen and CRP main effect of change was significant (p<0.001). However, the results were marginally significant when the two groups were compared in terms of D-dimer. When the simple effect is evaluated; Group A as not significant, while group B was significant. Starting CP treatment 5 days before or 5 days later did not change the laboratory parameters. However, D-dimer was marginally significant (p=0.058).
Conclusion: In our study, it was shown that early initiation of CP treatment reduced the hospitalization, but had no effect on mortality and laboratory parameters.

Key words: Convalescent plasma, COVID-19, SARS-CoV-2.

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INTRODUCTION

Corona Virus 2019 Disease (COVID-19), caused by a new type of beta coronavirus called SARS-CoV-2 in Wuhan, China, emerged as a new pandemic at the end of 2019 (1). More than 185 million people have been infected worldwide, and more than 4 million people have died due to this pandemic since then (2). The clinical signs and symptoms of the disease are very variable. Although it is mild in general, it is severe in approximately 14% of cases (dyspnea, hypoxia, severe lung involvement on imaging). Mortality rate is 3-4% (3, 4).

The treatments applied for COVID-19 disease does not have a specific treatment, these treatments are generally supportive treatments. Although different agents (favipiravir, azithromycin, anakinra, tocilizumab, remdesivir, lopinavir/ritonavir, high-dose steroid) are used in the treatment of COVID-19, their efficacy is not satisfactory (5, 6). Therefore, new treatment strategies are needed to alleviate symptoms and reduce mortality. Previous experience with SARS shows that convalescent plasma (CP) therapy elicits a directed neutralizing antibody response against the viral S protein. In addition, these antibodies prevent the entry of SARS-CoV-ACE2 (7). A retrospective study comparing the clinical results of high-dose steroid therapy and CP therapy in SARS patients showed that patients in the CP group had shorter hospitalization and lower mortality (8). Studies demonstrate that CP therapy is a safe method that improves passive immunity in COVID-19 patients (5, 6, 9-11). Despite the positive results of the use of CP in COVID-19 disease, the recently published randomized study shows that it does not have a significant effect on hospitalization and mortality (12). Therefore, the efficacy and safety of treatment, optimum volume, number of transfusions, the interval between transfusions, optimum neutralizing antibody titer should be determined (13, 14).

From this point of view, the changes in the clinical and laboratory parameters of the patients who received CP treatment at different times of the disease were evaluated retrospectively.

PATIENTS AND METHODS

The study was carried out with the ethics committee's approval (2021/133), by the Declaration of Helsinki, between May 2020 and February 2021. One hundred twenty-six patients who received standard therapy plus CP treatment were included in the study. The patients were divided into two groups as Group A and Group B. Group A, refers to the patients who started CP treatment within the first five days, and Group B, refers to the patients who started CP treatment after five days. The patients in these two groups were compared in terms of clinical and laboratory parameters and mortality.

Inclusion and Exclusion Criteria

It was applied to patients diagnosed with COVID-19 by RT-PCR method and did not have IgA deficiency in line with the CP usage criteria of the Turkish Ministry of Health Between May 2020 and February 2021 (15). Patients who were treated with CP in the first five days and patients who were treated with CP for more than five days were included in the study. A study file was created and demographic data of the patients (age, gender, comorbidity), laboratory results obtained at the time of admission (Lymphocyte, platelets (PLT), neutrophil, neutrophil/lymphocyte ratio, D-dimer, fibrinogen, ferritin, C-reactive protein (CRP), lactate dehydrogenase (LDH), hospitalization and survival were recorded. Patients who were diagnosed with COVID-19 and were not treated with CP were not included in the study.

Donor Selection and Plasmapheresis

CP donor selection was selected according to the CP guidelines of the Turkish Ministry of Health (16). Plasma was collected by the plasmapheresis for two doses of 200 ml from each donor. If CP was to be used immediately, irradiation was performed. The plasma that would not be used on the same day was stored at -25°C. The second CP treatment was performed at least 24 hours after the first CP application.

Statistical Analysis

The conformity of the data to the normal distribution was checked with the Shapiro-Wilk's test. Non-parametric tests were used for data that were not normally distributed. For descriptive statistics, it was expressed as Mean ± SEM (Standart error of mean) for continuous variables and as number (%) for categorical variables. Age and hospitalization parameters were compared between groups by independent t-test. A two-way repeated measure analysis of variance (ANOVA) was performed to test for the main effects corresponding to groups (Group A, Group B) and time (Before-After), as well as the interaction between the two (Groups and time: to see the effect of CP treatment on the Before-After change). In addition, a simple effect test was performed for each group. Total survival analyzes were evaluated using the Kaplan-Meier method. For comparison of survival curves between groups, log-rank test was used and

presented with 95% confidence intervals. All tests were applied in two tailed and p<0.05 was considered statistically significant. Analyzes was carried out with Jamovi ver. 1.2.27 software.

RESULTS

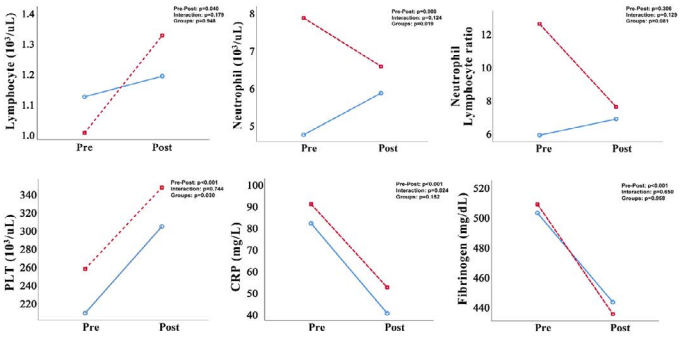
The male/female ratio of 126 patients was determined as 77/49. The mean time of CP treatment was 3.61±0.28 days from the hospitalization. One unit of CP was performed in 19 patients, two units in 107 patients, and three units in 5 patients. No comorbidity was found in 50 (39.68%) patients, while 76 (60.3%) patients had at least one comorbidity. The average hospitalization of the patients was 13.62±0.76 days. No complications were observed during and after CP treatment. Sixteen (12.7%) patients were taken to the intensive care unit, and 7 (5.5%) of these patients were intubated. One hundred-nineteen (94.4%) patients were discharged with recovery, 7 (5.5%) patients died (Table 1).

The patients were divided into two groups: those who received CP treatment within the first five days (Group A; n: 86) and those who received CP treatment after five days (Group B; n: 40). While the time of CP treatment was 1.87±0.14 days in Group A, it was 7.35±0.44 days in Group B. Treatment-related lymphocyte [F (1,124) =4.306, p=0.040, η2=0.034], PLT [F (1,124) =110.404, p<0.001, η2=0.471], fibrinogen [F (1,124) =19.189, p<0,001, η2=0.134] and CRP [F (1,124) =34.649, p<0.001, η2=0.134] main effect of change (before and after CP treatment) was significant. In contrast, the main effect of D-dimer and group interaction was marginally significant [F (1,124) =0.107, p=0.058, η2=0.029]. When the simple effect is evaluated; Group A [F (1,85) =0.602, p=0.440, η2 =0.007] as not significant, while group B [F (1, 39) =4.186, p=0.048, η2 =0.097] was significant. The mean of neutrophil [F (1, 124) =5.619, p=0.019, η2 =0.043] and PLT [F (1, 124) =4.791, p=0.030, η2 =0.037] were significant between groups. In addition, the neutrophil/lymphocyte ratio [F (1, 124) =3.096, p=0.081, η2 =0.043] was marginally significant between groups (Table 2) (Figure 1).

Table 1. Demographic and clinical characteristics of the all patients (CP: Convalescent plasma)

Parameters	COVİD-19 patients (n:126)
Gender	
Male	77 (61.1%)
Female	49 (38.9%)
Age (years)	63.75 ± 1.26
Comorbidity (n:76)	
Diabetes mellitus	21 (27.6%)
Hypertension	29 (38.15%)
Cadiovascular diseases	7 (9.2%)
Respiratory disease	12 (15.7%)
Chronic renal diasease	6 (7.8%)
Chronic liver diasease	1 (1.3%)
Malignancy	10 (13.15%)
Hospitalization (Day)	13.62 ± 0.76
CP application time (Day)	3.61 ± 0.28
Intensive care unit need	16 (12.7%)
Number of CP applied	
1 unit	19
2 unit	102
3 unit	5

Figure 1. Changes in hematological and biochemical parameters between groups (A-solid line and B-dash line) and time (Pre-Post).



Mean age was 62.15±1.6 years in Group A, while it was 67.2±1.9 years in Group B, and it was not statistically significant (p=0.061). Hospitalization was found to be 11.40±0.7 days in Group A and 18.4±1.7 days in Group B, and it was statistically significant (p<0.001) (Table 2). Patients who needed intensive care and died were patients in Group B.

While the median age of the patients who needed intensive care was 70.19±2.41, the median age of the patients who did not need intensive care was 62.82±1.38 and statistically marginal significant (p=0.050).

While the median age of the patients with comorbidity was 68.37±1.35, the median age of the patients without any comorbidity was 56.74±2.05 (p<0.001). Hospitalization was found to be 15.58±1.11 days in patients with comorbidity and 10.64±0.77 days in the other group, and it was statistically significant

Table 2. Evaluation of the characteristics of patients in Group A and Group B. According to repeated measurement analysis; †: Within-subjects effects p<0.05; ‡: interaction effect p<0.05; ¶: between-subjects effects p<0.05 and other parameters: p>0.05. Age (p>0.061) and hospitalizasyon (p<0.001). (PLT: Platelets; CRP: C-reactive protein; LDH: lactate dehydrogenase)

Parameters	Group A (n=86)		Group B (n=40)	
	Before Mean ± SEM	After Mean ± SEM	Before Mean ± SEM	After Mean ± SEM
Lymphocyte (10^3/uL) †	1.13±0.07	1.19±0.06	1.01±0.08	1.33±0.21
Neutrophil (10^3/uL) ¶	4.75±0.25	5.86±0.29	7.87±2.13	6.57±0.46
Neutrophil/ Lymphocyte □	5.89±0.6	6.86±0.65	12.59±5.6	7.6±0.94
PLT (10^3 /uL) †¶	210.1±8.3	305.4±11.8	258.8±22.3	348.3±27.4
Fibrinogen (mg/dL) †	502.8±13.9	443.1±13.8	508.7±20.9	435.2±18.2
D-dimer (ng/mL) ‡	693.2±224.1	582.4±103.7	526.2±108.9	873.8±246.7
CRP (mg/L) †	82.4±6.4	40.9± 4.1	91.4±9.9	52.94±7.12
Procalcitonin (ug/L)	0.23±0.04	0.24±0.05	0.3±0.05	0.2±0.02
LDH (U/L)	336.6±13.4	321.8±9.6	360.7±22.9	355.4±27.6
Ferritin (ug/L)	612.4±59.0	631.7±53.8	616.4±103.6	674.3±115.5
Age (years)	62.15±1.6	67.20±1.9		
Hospitalization (Day)	11.40±0.7	18.40±1.7		

(p=0.001).

All patients (n=122) had a mean of 27.8±0.92 (95% CI 25.9-29.6) days in the 30-day total survival analysis, and there was no difference between groups A and B when comparing the survival curves of those with CP. Median values of survival analysis by subgroups: Grup A (n=4) Survival: 26 ± 2.29 (95% CI 21.51-30.48); Grup B (n=2) Survival: 25 ±1.63 (95% CI 21.79-28.20); Overall: 26 ± 1.54 (95% CI 22.96-29.03) Chi-Square/ P: 0.021/0.886) (Table 3).

DISCUSSION

CP treatment came to the fore with the therapy to 5 severe COVID-19 patients who were resistant to steroid and antiviral treatment by Shen et al (17). It was started to reduce the mortality rate and the need for intensive care by collecting the plasma with anti-SARS COV-2 antibody from individuals diagnosed with COVID-19 and recovered by the plasmapheresis method. On the other hand, studies have started to be published showing that CP treatment in COVID-19 disease is beneficial in non-randomized studies and that it is not beneficial on the course of the disease in randomized studies (12). From this point of view, the effect of CP treatment on the clinical and laboratory findings of the patients was evaluated in this study.

Our study showed that initiation of CP treatment in the early period shortened the hospitalization but had no effect on survival. Liu et al. in a retrospective study of 39 patients, it was shown that the survival rate increased (18). In a randomized controlled study investigating clinical improvement up to 28 days after CP treatment, it was shown that 52% of patients who received CP treatment and 43.1% of the control group recovered, and no significant difference was observed in 28-day mortality (18, 19). In a randomized controlled trial of 464 COVID-19 patients conducted in India, 235 patients received CP therapy plus standard therapy, and 229 patients received standard therapy. It has been shown that CP treatment is not associated with disease severity and mortality rate (20). In a similar study of 241 patients, it was found that CP treatment did not significantly affect hospital stay and mortality (21). In the study of Cizmecioglu et al., which included 50 COVID-19 patients, it was shown that CP treatment performed in the first five

Table 3. Analysis of overall survival and comparison of survival times between groups.

Groups	Median		95% Confidence interval		Log rank
	Estimate	Std. error	Lower bound	Upper bound	Chi-Square / P value
Group A (n=4)	26.00	2.29	21.51	30.48	
Group B (n=2)	25.00	1.63	21.79	28.20	0.021 / 0.886
Overall	26.00	1.54	22.96	29.03	

decreased the hospitalization (22). In a recently published randomized trial involving 228 patients with severe COVID-19 pneumonia, it was shown that the use of CP compared to placebo in patients did not provide significant clinical benefit, did not affect 30-day mortality, and had no effect on other clinical and laboratory parameters (12). Some studies have shown that CP transfusion within the first 14 days results in good clinical results. A similar study determined that CP treatment in the first three days positively affected mortality (19, 23). Although definitive results regarding the effectiveness of CP have not been obtained in the literature, it has been shown in our study that it has no effect on mortality, but early application reduces the length of hospital stay.

The effect of the initiation time of CP therapy applied to COVID-19 patients on laboratory parameters had no effect in general. In other words, starting CP treatment 5 days before or 5 days later did not change the laboratory parameters. However, D-dimer was marginally significant. While D-dimer was 693.22 ng/mL before CP in Group A, it was 526.22 ng/mL in Group B. After-CP was found to be 582.45 ng/mL in Group A and 873.8 ng/mL in Group B. When these data were evaluated, it was thought that CP treatment had a negative effect on D-dimer if the onset time was above 5 days.

Studies on CP transfusion dose are planned with one unit (200 mL) for prophylaxis and one to two units for treatment. Although the duration of activity of antibodies is unknown, it is estimated to last from weeks to several months (24). In our study, one CP was applied to 19 patients, two CP to 102 patients, and three CP to 5 patients. Second unit CP therapy was required in the vast majority of patients. When 126 patients were evaluated, it was concluded that 1 unit of CP treatment was insufficient.

In the study, when the patients in need of intensive care were compared with the other patients, it was determined that the patients in need of intensive care were older and were found to be compatible with the literature. COVID-19 patients with co-morbidity have been shown to have a poor prognosis (25). Sixty percent of the patients in our study had at least one other disease, and seven patients who died were patients with the other disease. In addition, patients with comorbidities had longer hospitalization. In the group with additional disease, D-Dimer elevation and lymphopenia did not improve after CP. D-Dimer elevation and lymphopenia are associated with poor prognosis in COVID-19. It seems consistent with the literature that CP treatment did not affect laboratory parameters in the group with co-morbidity, except for the length of hospital stay (26).

The risks of CP treatment are similar to those of standard plasma. Risk of infection with another infectious disease agent (viral transmission or bacterial contamination), immunological reactions, non-hemolytic transfusion reactions (chills, fever, urticaria), transfusion-related overload (27). Our study shows that CP is a safe method without any complications during and after CP transfusion.

Our study has some limitations. Initially, other antiviral agents and steroid treatments were administered to the patients during their hospitalization. Secondly; The study was carried out retrospectively, and the antibody titer ratios of the CP used could not be studied for technical reasons. As a third, patients who did not receive CP treatment as a control group could not be included, so they were compared in terms of transfusion time and needed for intensive care.

As a result; the effectiveness of CP treatment, as in our study and other studies, is still the subject of study. Our study showed that although early CP treatment reduced the hospitalization, it did not affect survival. Although CP treatment seems effective in non-randomized studies, randomized studies show that CP use is ineffective. Therefore, we believe that randomized controlled studies are needed.

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Institutional Review Board Statement: The study was carried out with the Necmettin Erbakan University Ethics Committee's approval (2021/133), by the Declaration of Helsinki.

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NEÜ YAYINEVİ

Measurement of Epidermis, Dermis, and Total Skin Thicknesses from Six Different Face Regions

Altı Farklı Yüz Bölgesinden Epidermis, Dermis ve Toplam Cilt Kalınlıklarının Ölçümü

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Öz

Amaç: Bu çalışmanın amacı, yüzün altı farklı bölgesinin epidermis, dermis ve toplam deri kalınlıklarının belirlenerek cilt kalınlığı haritası oluşturmaktır.
Hastalar ve Yöntem: Yaşları 30-80 arasında değişen 90 kadın ve 90 erkek hastanın yüz derisi, saçlı deri, alın, yanak, kulak, burun ve dudak bölgelerinden 9-10 mm sağlıklı doku içeren örnekler retrospektif olarak çalışmaya dahil edildi. Örnekler incelendi ve ışık mikroskobu altında mikrometre ile epidermis ve dermis kalınlıkları ölçüldü.
Bulgular: Çalışmaya alınan 90 kadın katılımcının 6 yüz bölgesinin epidermis kalınlıkları 65,91±14,44 µm ile 120,91 ±44,74 µm, dermis kalınlıkları 1150±217,43 µm ile 1498,33±388,56 µm ve toplam deri kalınlıkları 1234,83± 217,6 µm ve 1599,33±492,2 µm idi. Çalışmaya alınan 90 erkek katılımcının 6 yüz bölgesinin epidermis kalınlıkları 79,08±13,88 µm ile 122,75±32,5 µm, dermis kalınlıkları 1106,66±389,82 µm ile 1942,5±464,06 µm ve toplam deri kalınlıkları 1756±503,75 µm ve 2022,5±460,24 µm arasında bulundu.
Sonuç: Kadın hastalarda en ince epidermis saçlı deriden, erkek hastalarda ise en ince epidermis yanaktan ölçüldü. En kalın epidermis kadın ve erkek hastalarda üst dudak üstü bölgedeydi. Ancak dermis kalınlığının en ince ve kalın olduğu bölgeler cinsiyete göre farklılık gösterdi. Daha ileri çalışmalarda, daha çok merkezli, çok ırklı materyaller kullanılarak yüzün daha fazla alt birime bölünmesiyle yüz derisi kalınlığının tam bir haritası elde edilebilir.

Anahtar Kelimeler: Yüz derisi, deri, kalınlık, epidermis, dermis, histometrik

Abstract

Aim: The aim of this study was to map the skin thickness by determining the epidermis, dermis and total skin thickness of six different regions of the face.
Patients and Methods: Samples containing 9-10 mm of healthy tissue from the facial skin, scalp, forehead, cheek, ear, nose and lip regions of 90 female and 90 male patients aged between 30 and 80 years were retrospectively included in the study, and epidermis and dermis thicknesses examined with a micrometer under a light microscope.
Results: Epidermis thicknesses of 6 facial regions of 90 female participants included in the study were between 65.91±14.44 µm and 120.91±44.74 µm, dermis thicknesses were between 1150±217.43 µm and 1498.33±388.56 µm, and total skin thicknesses were between 1234.83±217.6 µm and 1599.33±492.2 µm. Epidermis thicknesses of 6 facial regions of 90 male participants included in the study were between 79.08±13.88 µm and 122.75±32.5 µm, dermis thicknesses were between 1106.66±389.82 µm and 1942.5±464.06 µm, and total skin thicknesses were between 1756±503.75 µm and 2022.5±460.24 µm.
Conclusion: In the female patients, the thinnest epidermis was measured on the scalp and the thinnest epidermis in the male patients was measured on the cheek. The thickest epidermis was on the upper lip in the male and female patients. However, the regions with the thinnest and thickest dermis thicknesses differed according to gender. In further studies, a full map of facial skin thickness can be obtained by dividing the face into more subunits using more multicentre, multiethnic materials.

Key words: Facial skin, skin, thickness, epidermis, dermis, histometric

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INTRODUCTION

The skin is one of the largest organs of the body with its many components and layers (1). It is frequently affected by external factors and surgical interventions. Thousands of studies are carried out in a wide variety of fields related to human skin. It is essential for these studies to know the vascular networks, other components and layers of the skin as well as its histological structure as a whole. In general, the histological structure of the skin has been known for many years, but another known fact is that the size and density of these components and layers are variable according to body regions (2).

Knowing the mean values of the epidermis and dermis thicknesses in different body parts of the skin is important for many applications to the skin, from laser to peeling, and from vaccination to intradermal injections. For this reason, measurement results with various techniques have been reported in many different studies in the literature (1-5).

Although biopsy is the gold standard for determining skin thickness, it is ethically inconvenient to take a biopsy from a healthy person (4). Although imaging techniques such as ultrasound and magnetic resonance (MR) can also provide data, they cannot provide precise objective results as in microscopic examination (2). Although skin thickness can be measured microscopically in studies performed on fresh cadavers, measurement of skin thickness from surgical materials taken from living people stands out as a method that can be standardized and conformed to ethical rules, due to the difficulties in obtaining cadavers and the inability to obtain sufficient number of preparations (4).

Knowing the thickness of the epidermis and dermis in many skin rejuvenation methods that work with the logic of creating damage to the skin can increase the efficiency after the procedure and reduce the complications that may develop. Although the skin thickness of various regions of the face of people in different geographical regions is reported in the literature, the skin thickness of the facial region of people living in Anatolia is not known.

The aim of this study was to map the skin thickness by determining the epidermis, dermis and total skin thickness of six different regions of the face.

PATIENTS AND METHODS

Patients who were operated for malignant skin tumors in the Plastic Surgery clinic between 2016 and 2021 were retrospectively scanned. Ethical approval was obtained from the Ethics Committee of Necmettin Erbakan University, Meram Faculty of Medicine (2022/3947). Materials containing at least 9-10 mm of healthy skin adjacent to the surgical margin, without lesions, were included in the study. Materials that did not have a sufficient length of solid area between the lesioned area and the surgical margin, and that appeared congested due to fixation problems or that contained crosssection artifacts were excluded from the study. The patients were contacted by phone and their height and body weights were questioned. Patients with a body mass index of 20-20 were included in the study, while patients defined as thin or obese were excluded.

Subunits were scanned separately for male and female patients by regionbased search from the computer system. Facial subunits in which the data of at least 30 patients, 15 female and 15 male, could be measured, were determined. Facial regions with fewer samples than this number were excluded from the study (Figure 1).

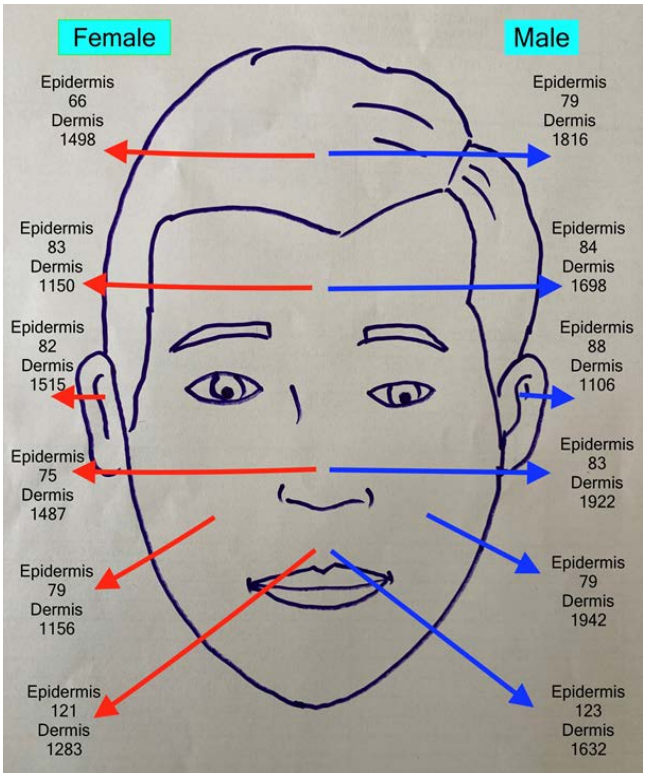

Figure 1. Epidermis and dermis thickness of the six regions in μm in female and male face skins; scalp, forehead, cheek, ear, upper lip and dorsum of the nose.

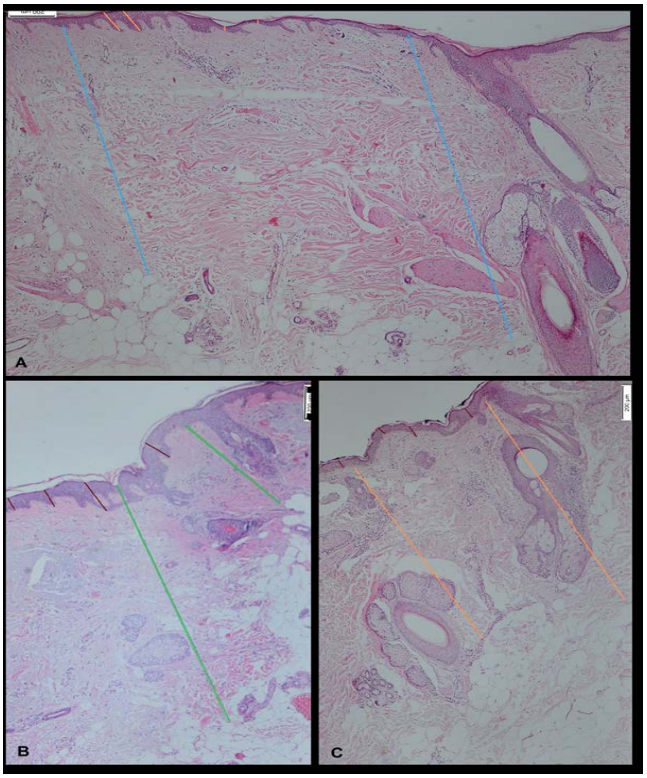

Figure 2. The mean dermis and epidermis and total skin thicknesses of each case were calculated by taking 4 measurements of different thicknesses for the epidermis and 2 measurements for the dermis, X40,H&E, A)Scalp B)Forehead C)Ear

H&E stained preparations of the listed materials were obtained from the archive of the Department of Medical Pathology.

The epidermis and dermis thicknesses of 15 skin tissues of male and female patients for each region were measured with a micrometer under an Olympus light microscope. The mean dermis and epidermis and total skin thicknesses of each case were calculated by taking 4 measurements of different thicknesses for the epidermis and 2 measurements for the dermis (Figure 2). All preparations were evaluated by the same pathologist and then checked by a second pathologist. Preparations with differences in measurements were excluded from the study.

Statistical Analysis

The mean, median, standard deviation, Q1 value and total skin thickness of epidermis and dermis thicknesses were calculated statistically using Microsoft Excel® (Microsoft Corporation, Redmond, Washington) program. Mean ± standard deviation values were evaluated for genders and regions.

RESULTS

In the study, the preparations of a total of 3584 patients were scanned. A total of 180 of these preparations meeting the criteria were included in the study. The six regions were evaluated because of the sufficient number of materials belonging to the scalp, forehead, cheek, ear, upper lip and dorsum of the nose on the facial skin. (Figure 1).

The mean age of the 90 female participants included in the study was 66. Epidermis thicknesses of the six facial regions ranged from 65.91±14.44 µm to 120.91±44.74 µm, dermis thicknesses ranged from 1150±217.43 µm to 1498.33±388.56 µm, and total skin thicknesses ranged between 1234.83±217.6 µm and 1599.33±492.2 µm. The thinnest epidermis was measured on the scalp, the thickest epidermis on the upper lip, the thinnest dermis on the forehead, and the thickest dermis on the ear skin. Total skin thickness was thinnest on the forehead and thickest in the ear (Table 1).

The mean age of the 90 male participants included in the study was 67. Epidermis thicknesses of the six facial regions ranged from 79.08±13.88 µm to 122.75±32.5 µm, dermis thicknesses ranged from

Table 1. Female epidermis, dermis and total skin thickness of six face regions

Female	Epidermis (µm)				Dermis (µm)				Total skin thickness (µm)			
	Mean	Median	SD	Q1	Mean	Median	SD	Q1	Mean	Median	SD	Q1
Forehead	83,83	75	24,84	66,25	1150	1150	217,43	1006,25	1234,83	1220	217,60	1087,50
Nose	75	70	18,80	57,50	1487,50	1375	505,12	1175	1563,50	1470	497,69	1262,50
Lip	120,91	117,50	44,74	80	1283,33	1250	447,08	981,25	1403,33	1360	446,32	977,50
Ear	82	75	20,45	63,75	1515	1537,50	486,28	1256,25	1599,33	1620	492,20	1315
Scalp	65,91	63,75	14,44	61,25	1498,33	1425	388,56	1275	1564,83	1487,50	387,77	1340
cheek	78,83	80	24,44	61,25	1155,83	1225	356,84	968,75	1244,66	1282,50	355,40	1005

Table 2. Male epidermis, dermis and total skin thickness of six face regions

Male	Epidermis (µm)				Dermis (µm)				Total skin thickness (µm)			
	Mean	Median	SD	Q1	Mean	Median	SD	Q1	Mean	Median	SD	Q1
Forehead	84,25	85	21,42	68,75	1698,33	1625	514,27	1368,75	1784,83	1707,50	512,61	1332,50
Nose	83,16	82,50	18,53	72,50	1922,50	1762,50	535,92	1531,25	2009,98	1870	525,11	1577,50
Lip	122,75	121,25	32,5	97,50	1631,66	1687,50	512,41	1399,37	1756	1772,50	503,75	1457,50
Ear	87,91	83,75	28,31	66,25	1106,66	1100	389,82	800	1200,33	1215	387,65	857,50
Scalp	79,41	76,25	17,04	66,25	1815,83	1850	432,71	1537,50	1899,83	1960	432,63	1510
Cheek	79,08	72,50	13,88	66,25	1942,50	1900	464,06	1731,25	2022,50	1977,50	460,24	1722,50

1106.66±389.82 µm to 1942.5±464.06 µm, and total skin thicknesses ranged from 1756±503.75 µm to 2022.5±460.24 µm. The thinnest epidermis was on the cheek area, while the thickest epidermis was on the upper lip. The thinnest dermis was measured on the ear skin, and the thickest dermis on the forehead skin. The thinnest total skin thickness was measured at the ear, while the thickest area was at the cheek (Table 2).

The areas with the thickest epidermis thicknesses were the same in the male and female patients. However, the regions with the thinnest and thickest dermis thicknesses differed according to gender.

DISCUSSION

The epidermis, dermis and total thicknesses of the skin may vary according to body regions, age, gender and even geographical region and ethnic origin (2-9). Knowing the measurement values of skin layers is important for many scientific studies, treatment applications, oncological staging and evaluation studies, in vitro skin models, some surgical techniques and reconstruction applications (3,4,6-13).

There are studies measuring facial skin thicknesses in the literature (2-5). Most of the studies were conducted using noninvasive but costly methods such as computed tomography (CT), magnetic resonance imaging (MRI), ultrasonographic (USG) evaluation and confocal microscopy, and histometric and invasive methods using cadaver skin, autopsy materials, or punch biopsy materials applied to living humans (3,7,9-12,17-19). The diversity of measurement methods causes a lack of standardization. MRI and CT are expensive techniques for measuring skin thickness and cannot provide as detailed information as microscopic examinations. There is a risk of radiation exposure with CT and a subjective assessment risk with USG (7,15). Confocal microscopy instrumentation methods can provide accurate measurements, but are still under development (15,17). There are studies measuring breast skin thickness on filmscreen mammograms in women (20). Although this method is also inexpensive and noninvasive, it is not a suitable method for measuring whole body skin areas due to direct exposure to radiation. The methods other than histometric measurement may not allow to measure epidermis and dermis thicknesses separately and it may be difficult to establish standardization.

In the method we defined, intact skin parts of retrospective surgical materials are used and it reflects the results of living people with an ethical method. We think that the values obtained by this method are more standardized than the values obtained from autopsy or other methods. On the other hand, data on thousands of patients can be obtained using this technique. Comparative studies on this subject can contribute to the literature.

In studies measuring facial skin thicknesses (3,7,10,18,21), it is seen that values obtained by USG are used, as well as histometric methods obtained from punch biopsy and cadaver. To the best of our knowledge, a study that measures the epidermis, dermis and total skin thicknesses of various parts of the facial skin histometrically as a whole was not found in the literature. In a study of Lee et al., in which the skin thickness of Korean adults was measured using punch biopsy materials, skin thicknesses of several facial regions such as the eyelid and chin were reported, and it is seen that the measurements in the facial region were limited to 2-3 regions due to the invasiveness of the method (3). On the other hand, the number of specimens measured in these regions varied between 6 and 14. Whereas, in our study, more reliable data were obtained by examining

15 specimens for each region. In this study, it was reported that the mean thickness of the epidermis and dermis at the forehead region were 93.6 and 788 μm. In our study these thicknesses were 84 and 1424 μm. They reported that the mean thickness of the epidermis and dermis at the cheek region were 98.2 and 1076 μm. In our study these thicknesses were 79 and 1562 μm. In both regions, the thickness of the epidermis of the Turkish people was thinner than that of the Korean society, and it is seen that the thickness of the dermis was much higher. We could not compare the skin thicknesses of the other regions we measured in our study, since we could not find any studies that measured different ethnic origins.

In a study, a significant relationship was reported between dermis thickness and hypertrophic scar (6). According to the data obtained in our study, it can be thought that there will be more scars in areas with high dermis thickness in men and women. When evaluated from this point of view, it can be thought that most of the incisions in men will be on the forehead. Incisions to be made parallel to the forehead lines can eliminate this disadvantage.

The limitations of our study are that the facial skin is limited to some main regions and the average age is high. However, this is due to the fact that we use the available archive data. Further studies to be conducted in large centers where the excision materials of facial skin lesions are much more or multicenter studies can provide contribution to the literature.

In many studies in the literature, it has been reported that the thinnest skin on the face is in the eyelid (3, 23). Although this region was not examined in our study, it was observed that the thinnest epidermis was on the cheek in men among the examined regions. In procedures such as laser, peeling, dermabrasion to the cheek area of men, performing the procedure more superficial than other parts of the face can reduce possible hyperpigmentation. In both sexes, the processes that damage the epidermis on the upper lip, where the thickest epidermis is determined, can be performed deeper than the other regions.

The thickness of the epidermis and dermis we determined in our study is quite different from the studies in the literature. In particular, the application depth to be determined in rejuvenation methods that work with the logic of damaging the upper layers of the skin should be chosen in different thicknesses according to each society. Otherwise, the possibility of encountering unwanted complications such as hyperpigmentation or scarring may increase.

In further studies, a full map of facial skin thickness can be obtained by dividing the face into more subunits using more multicentre, multiethnic materials.

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SELÇUK TIP DERGİSİ
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Olgu Sunumu / Case Report

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Use of Mechanically Isolated Stromal Vascular Fraction in Different Wound Types

Farklı Yara Tiplerinde Mekanik Yolla İzole Edilen Stromal Vasküler Fraksiyonun Kullanımı

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Öz

Yara iyileşmesi hemostaz, inflamasyon, proliferasyon ve maturasyon evrelerinden oluşan fizyolojik bir süreçtir. Bu iç içe geçmiş evrelerin herhangi bir aşamasında meydana gelen bozulma, klinisyenin karşısına için kronik bir yara olarak çıkar. Lipoaspiratın enzimatik veya mekanik sindiriminden sonra sulu fraksiyonun bir parçası olarak izole edilen stromal vasküler fraksiyon, aynı zamanda önemli bir mezenkimal kök hücre rezervidir. İçerdiği preadipositler, endotelyal öncüler, immün hücreler, hematopoietik hücreler, fibroblastlar ve perisitler nedeniyle heterojen bir doku kokteyli olarak kabul edilir. Proanjiyogenik, antiapoptotik, antifibrotik, immünomodülatör ve antiinflamatuar aktivitelerinin yanı sıra izolasyondaki avantajları nedeniyle, stromal vasküler fraksiyon son zamanlarda mevcut yara tedavisinde önem kazanmıştır. SVF tek başına yara iyileşmesi için etkili olmasa da, uygun yara bakımı ve rekonstrüksiyon öncesi yara yatağı hazırlığı için yeterli hücre sayısı ve canlılığı sağlayan ve cerrahi başarısızlığı en aza indiren yeni ve etkili bir teknolojidir.

Anahtar Kelimeler: Kronik yara, stromal vasküler fraksiyon, yağ grefti, yara iyileşmesi

Abstract

Wound healing is a physiological process consisting of hemostasis, inflammation, proliferation, and maturation phases. The disruption that occurs at any stage of these intertwined phases is presented to the clinician as a chronic wound. The stromal vascular fraction, isolated as a part of the aqueous fraction after enzymatic or mechanical digestion of lipoaspirate, is an important mesenchymal stem cell reserve, as well as. It is considered a heterogeneous tissue cocktail due to the preadipocytes, endothelial precursors, immune cells, hematopoietic cells, fibroblasts and pericytes it contains. Because of its proangiogenic, antiapoptotic, antifibrotic, immunomodulatory, and anti-inflammatory activities, as well as its advantages in isolation, stromal vascular fraction has lately acquired prominence in current wound therapy. Although SVF alone is not effective for wound healing, it is a new and effective technology that provides adequate cell count and viability for proper wound care and wound bed preparation prior to reconstruction and to minimize surgical failure.

Key words: Chronic wound, fat grafting, stromal vascular fraction, wound healing

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INTRODUCTION

Wound healing is a physiological process consisting of hemostasis, inflammation, proliferation, and maturation phases. The disruption that occurs at any stage of these intertwined phases is presented to the clinician as a chronic wound. An ideal method could not be determined due to the high cost of chronic wound management, the inability of current therapeutic strategies to affect all healing phases alone, and potential adverse effects. After it was discovered in 2001 that stem and stromal cells could be obtained from adipose tissue, more research studies were designed about the use of this cell groups with regenerative properties (1). The stromal vascular fraction (SVF), isolated as a part of the aqueous fraction after enzymatic or mechanical digestion of lipoaspirate, is an important mesenchymal stem cell reserve, as well as. It is considered a heterogeneous tissue cocktail due to the preadipocytes, endothelial precursors, immune cells, hematopoietic cells, fibroblasts and pericytes it contains (2,3). SVF has two primary characteristics that distinguish it from mesenchymal stem cells in terms of tissue regeneration. The first of these is that thanks to the different cell components it contains it offers greater benefits in aspects such as immunomodulation, anti-inflammatory, and angiogenesis than compared to the application of stem cells alone, and the second is that SVF is easier to produce than stem cell isolation (4). Because of its proangiogenic, antiapoptotic, antifibrotic, immunomodulatory, and anti-inflammatory activities, as well as its advantages in isolation, SVF has lately acquired prominence in current wound therapy. The goal of our research was to present the outcomes of mechanically produced SVF in patients with different wounds caused by a variety of factors.

Application Procedure

Patients were operated in the operating room with spinal anesthesia with sedation or general anesthesia. Fat is harvested from any area of excess subcutaneous fat and/or areas of patient preference if sufficient fat is available. The patient who have malignancy, coagulation disorder, pregnancy and connective tissue disease is accepted unsuitable candidate for lipoaspirate. Tumescent anesthesia material, which is approximately 35% more than the amount of fat planned to be harvested, was given to the donor area from which lipoaspirate was to be harvested, and it was waited for an average of 5-10 minutes. Tumescent anesthesia includes 500 mL of Ringer lactate with 25 mg lidocaine and 1 vial of epinephrine [1:1000]. With a blunt cannula with maximum dimensions of 20 mm length, 3 mm diameter and 2 mm aspiration holes, lipoaspirate was transferred to the closed system bag as milli-fat. For the separation of Milli fat tumescent anesthesia in the bag, 60 cc of SF was added and suspended for 5 -7 minutes to ensure decantation. After separation, the tumescent anesthesia material was removed from the bag by means of a 3-way cock. The washed Milli fat remaining in the bag was reduced to micron level by mechanically refining with the help of knives in 2 separate Lipocube SVF black cubes, respectively. Autologous fat, which was reduced to micron degree, was separated in a patented piston tube with 4 separate concave gaskets (black piston injector) and Lipocube special software variable speed Celldrive (Centrifuge) device for 9 minutes. 'Stromal Vascular Matrix' was obtained by combining Extracellular Matrix and Stromal Vascular cell collection in the separated fat. The resulting Stromal Vascular Matrix was injected intralesional and perilesional to the wound of the patients

CASE

Case 1

A 62-year-old male patient was admitted to the wound care clinic with a tissue defect accompanied by ulcerated areas which is hyperemic and edematous on the left cruris diagnosed as a venous ulcer (Figure 1). Venous doppler ultrasound showed no thrombosis

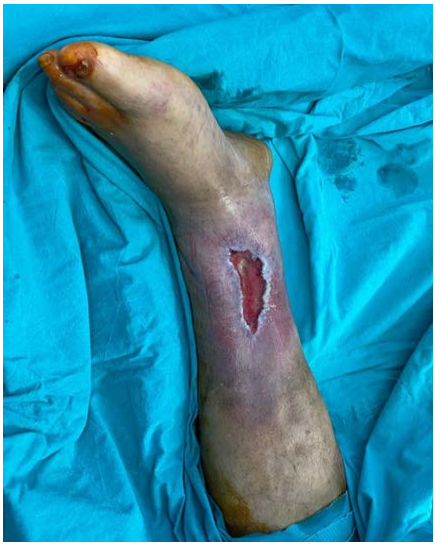

Figure 1. The tissue defect accompanied by ulcerated areas which is hyperemic and edematous on the left cruris diagnosed as a venous ulcer

on the venous system. He has a history of diabetes mellitus, coronary artery disease and hypertension. At the time of admission, the patient's body temperature was 36.10C. The erythrocyte sedimentation rate was 55 mm/h and the CRP value was 31.6 mg/L. Revascularization was not considered necessary after the Ankle/Arm index was found to be 0.9. Debridement was performed, and cultures were taken. Upon the growth of P. Aeuriginosa in deep tissue culture, piperacilin/tazobactam was applied in accordance with the culture antibiogram, and antibiotic therapy was applied for 10 days. In the wound care follow-up process, Negative Pressure Wound Therapy was applied for 9 days. Fat-derived Mechanical Stromal Vascular Matrix application was applied to the patient whose granulation tissue progression slowed down during this period. After the operation patient had hyperbaric oxygen therapy for 20 sessions. Pre-procedure sedimentation rate was 32 mm/h and CRP was 9 mg/L. Wound dimensions were measured as 6 cm x 6 cm x 3 cm (108 cm^3) during the application. After the SVM application, the follow-up was continued with the hydrocolloid-containing passive wound dressing and offloading. Photographing and wound care were performed at 1st, 3rd, and 6th week follow-ups. At the end of the sixth week, 96.7% success was achieved with a wound volume of 1.5 cm^3 (Figure 2-3).

Case 2

A 43-year-old male patient was admitted to the wound care clinic with a detachment of the anterolateral thigh flap on the anterior and inferomedial side on the

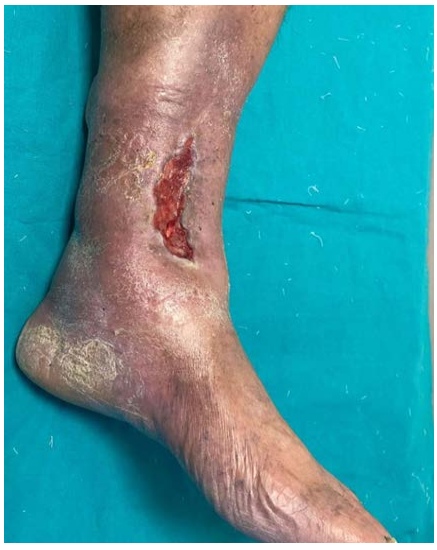

Figure 2. Postoperative third week of venous ulcer on left medial malleolus

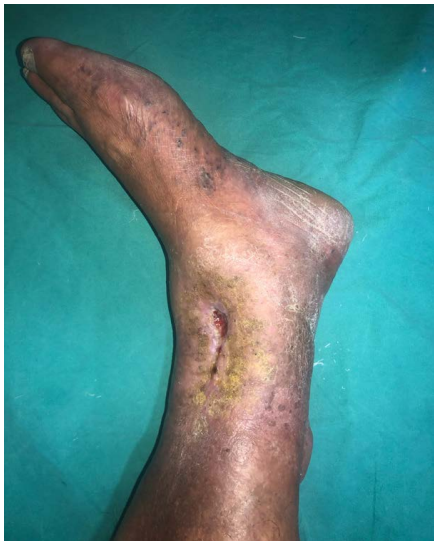

Figure 3. Postoperative sixth week of venous ulcer on left medial malleolus

postoperative third months. Tissue defect that was present on the previous operation was due to the car accident. There was no early complication during the follow-up. Patient was discharged 7th day of operation with no detachment or infection. Patient has no significant medical history. At the time of admission, the patient's body temperature was 36.4oC. The CRP value was 32.4 mg/L. Debridement was performed, and cultures were taken. No significant growth of any bacteria in deep wound culture. On the wound care follow-up process, Negative Pressure Wound Therapy was applied for 9 days. Fat-derived Mechanical Stromal Vascular Matrix application was applied to the patient. After the SVM application, the follow-up was continued with the hydrocolloid-containing passive wound dressing and offloading Photographing and wound care were performed at 1st, 3rd, and 6th week follow-ups. At the end of the sixth week, no more tissue defect between the flap and nearby tissue.

Case 3

A 47-year old man patient was admitted to the wound care clinic with a total necrosis of D1 and D2 at the level of metatarsophalangeal joint (Figure 4). An arterial doppler ultrasonography showed that the anterior tibial artery had biphasic flow pattern on the affected side. The patient consulted to the orthopedy and traumatology department. Orthopedy department planned a ray amputation. On postoperative 13th day, patient came to our clinic with detachment and bed-smell at the incision site. Debridement was performed, and cultures were taken. Upon the growth

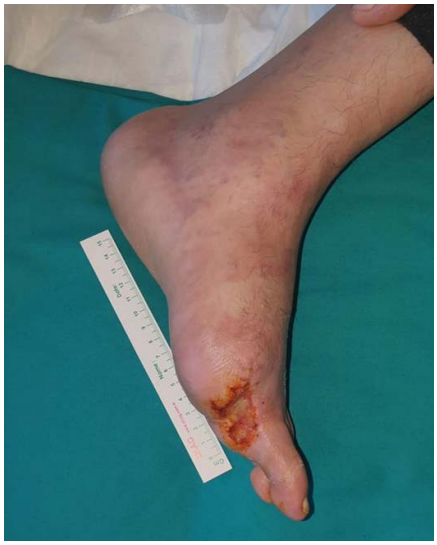

Figure 4. The tissue defect on the level of D1 and D2 metatarsophalangeal joint

of A. Baummani in deep tissue culture, tigesiklin was applied in accordance with the culture antibiogram, and antibiotic therapy was applied for 18 days. At the end of the sixth week of SVM application, no more tissue defect between the flap and nearby tissue (Figure 5).

Case 4

A 62-year old man patient was admitted to the wound care clinic with a diabetic ulcer that is hyperemic, sniffy and purulent discharge on the dorsal surface of the left foot. Peripheral pulses were palpable. Arterial and venous doppler ultrasonography showed no significant changes on the vascular system bilaterally. On the X-Ray; there was no sign of osteomyelitis or any kind of osseous pathologies. CRP value was 256 g/dL. Three sessions of debridement were performed. Upon the growth of A. Baummani and P. Aeruginosa in deep tissue culture, tigecycline and piperacillin/tazobactam were applied in accordance with the culture antibiogram, and antibiotic therapy was applied for 13 days. Fat-derived Mechanical Stromal Vascular Matrix application was applied to the patient. After the SVM application, on the postoperative 30th day; we reconstruct the tissue defect with the split-thickness skin graft.

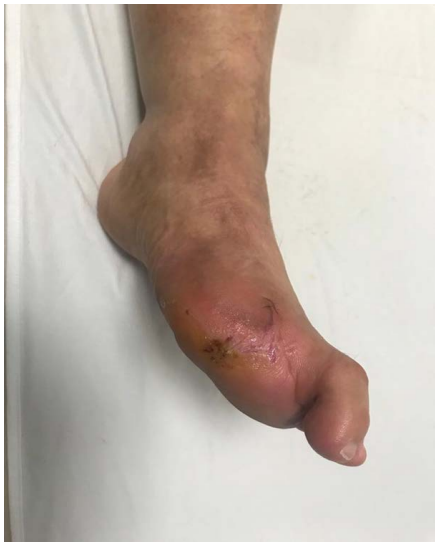

Figure 5. Postoperative sixth week without no more tissue defect

DISCUSSION

Chronic wounds, which are difficult to heal and complex, cause patients to stay in the hospital for a long time periods and create a workload for the health workforce. Although wound healing follows the same process in all tissues, wounds due to different etiological reasons can become complicated. Even though progress has been achieved in chronic wound management with the developing technology, complex procedures are still required in the management of severe wounds treatments despite modern wound dressings (5). Since there is no method that can provide success in chronic wound management alone, many procedures are performed before reconstruction in order to optimize wound healing and achieve success in chronic wound management (6). The stromal vascular fraction included in these procedures is an application that is obtained from adipose tissue through several mechanisms and has a positive contribution to wound healing (8). Isolation methods that can be used to obtain SVF can be basically divided into three as enzymatic methods, automatic devices, and mechanical separation (9). Despite the fact that enzymatically generated lipoaspirate can contain around 100000-1300000/gr cells, its high cost and long manufacturing time (average 120 minutes) limit its application (7). In addition, it has been evaluated by the FDA within the scope of drug research since it is argued that the enzymatic SVF production causes deterioration of the original content and tissue integrity of the adipose tissue. On the other hand, automatic devices have an advantage over enzymatic isolation as they provide isolation in closed environment, limit contamination, and standardize in clinical practice. However, the cost of the devices

has been the major limitation of this method (8). Due to these disadvantages, mechanical methods such as shaking, vibration, centrifugation, and sonication have been tried in recent years to obtain SVF. SVF obtained via this way is called 'Tissue-like SVF' (10). They contain extracellular matrix fragments and cells because they are microfragmented adipose tissue. When different mechanical isolation methods were compared to enzymatic techniques, it was observed in the study by Tiryaki that, while the cell number in the SVF population obtained by mechanical methods was low, the cell division rate and type 1 collagen gene expression were higher in the SVF population isolated by mechanical methods (11). Increased type 1 collagen gene expression indicates the acceleration of collagen production and the secondary enhanced wound healing as a result. The physiological activities and functionality of cells are thought to be enhanced by cellular forces during mechanical isolation. The most significant advantage of this method is that it can be obtained easily and quickly even in the operating room, since no enzymes are used during mechanical isolation; nevertheless, obtaining fewer cells is seen as a disadvantage when compared to enzymatic methods. Banyard et al. suggested in a study that cell number is less important in SVF activity than previously thought, and that cell activity is more important (12). In addition, Fraser et al. determined that mesenchymal stem cells were 500 times more abundant in SVF than in bone marrow. Thus, it can be argued that SVF, which is easily obtained from adipose tissue, can be an alternative to bone marrow-derived mesenchymal stem cells (13). Another advantage regarding the use of SVF for wound healing is that it contains not only stem cells, but also a regulatory T cell (Treg), which is a cell of the immune system and has anti-inflammatory properties. According to another study, it was argued that there are more Tregs in the SVF compared to the peripheral regions, and that the anti-inflammatory effect to be obtained by SVF application can be increased in this way (14). There is not enough information on when it should be used in the chronic wound method, however in our study, it was preferred to be applied after showing the absence of microbial growth and osteomyelitis in the wound culture. Furthermore, it would be appropriate to use it in chronic wound management after correcting the pathologies that need to be treated depending on the etiology. Although SVF alone is not effective for wound healing, it is a new and effective technology that provides adequate cell count and viability for proper wound care and wound bed preparation prior to reconstruction and to minimize surgical failure.

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