Mehmet Akif Ersoy Üniversitesi

Sağlık Bilimleri Enstitüsü Dergisi

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I- Mehmet Akif Ersoy Üniversitesi Sağlık Bilimleri Enstitüsü Dergisi Genel Bilgiler

Mehmet Akif Ersoy Üniversitesi (MAKÜ) Sağlık Bilimleri Enstitüsü Dergisi, Mehmet Akif Ersoy Üniversitesi Sağlık Bilimleri Enstitüsü'nün yayın organıdır. Derginin kısaltılmış adı "MAKÜ Sag. Bil. Enst. Derg" dir. Yılda 2 kez yayınlanır. MAKÜ Sağlık Bilimleri Enstitüsü Dergisi sağlık bilimleri, (veteriner, tıp, diş hekimliği, hemşirelik ve spor bilimleri) alanlarında temel ve klinik hakemli bilim yazılarının yayınlandığı hakem-denetimli bir dergidir. Derginin dili İngilizce'dir. Dergiye gönderilen yazıların başka herhangi bir dergide yayınlanmamış, yayına kabul edilmemiş ya da yayınlanmak üzere değerlendirme aşamasında olmaması gerekir. Bu kural bilimsel toplantılarda sunulan ve özeti yayınlanan bildiriler için geçerli değildir. Ancak, bu gibi durumlarda bildirinin sunulduğu toplantının adı, tarihi ve yeri bildirilmelidir. Makalelerin formatı "Uniform Requirements for Manuscripts Submitted to Biomedical Journals: Writing and Editing for Biomedical Publication (http:/www.icmje.org/)" kurallarına göre düzenlenmelidir.

Gönderilen yazılar yayın kuruluna ulaştıktan sonra öncelikle, yazım kurallarına uygunluğu yönünden değerlendirilir; sonucu yazara dört hafta içinde bildirilir. Yazının, gerek teknik özellikleri gerekse genel kapsamı açısından derginin genel yayın ilkelerine uygun bulunmaması durumunda yazı reddedilir. Ya da, gerekirse, yazar(lar)ın yazıyı yazım kurallarına uygun biçimde yeniden göndermeleri istenebilir. Yeniden gönderilen yazılar benzer bir teknik incelemenin ardından yazım kurallarına uygun ise danışman denetimi sürecine alınır. Yazı, editör ve yardımcı editörler ile yazının başlık sayfasını görmeyen en az iki danışmana gönderilerek incelenir. Yazı, yayın kurulunun belirlediği ve bilimsel içerik ve yazım kuralları açısından değerlendirilir. Editör ve yardımcı editörler gerek gördüğünde makaleyi üçüncü bir danışmana gönderebilir. Hakem belirleme yetkisi tamamen editör ve yardımcı editörler ve yayın kuruluna aittir. Danışmanlar belirlenirken derginin uluslararası yayın danışma kurulundan isimler seçilebileceği gibi yazının konusuna göre ihtiyaç duyulduğunda yurt içinden veya yurt dışından bağımsız danışmanlar da belirlenebilir. Daha sonra, danışman raporları dikkate alınarak ve gerekirse yazar(lar)la tekrar iletişim kurularak yayın kurulunca son redaksiyon yapılır. Yazıların kabulüne editör karar verir.

Editör yayın koşullarına uymayan yazıları; düzeltmek üzere yazarına geri gönderme, biçimce düzenleme veya reddetme yetkisine sahiptir. Yazılarını geri çekmek isteyen yazarlar bunu yazılı olarak editöre bildirmek durumundadır. Editör görülen lüzum halinde bazı makaleler hakkında yayın yürütme kurulunun görüşüne başvurur. Bu değerlendirme süreci dergiye gönderilen yazı türlerinden araştırma yazılarını, olgu sunumlarını ve özgün yazıları kapsar. Diğer yazı türlerindeki yazılar doğrudan yayın kurulunca değerlendirilir. Dergiye gönderilen yazılar yayınlansın ya da yayınlanmasın geri gönderilmez. Tüm yazarlar bilimsel katkı ve sorumluluklarını ve çıkar çatışması olmadığını bildiren toplu imza ile yayına katılmalıdır. Araştırmalara yapılan kısmi de olsa nakdi ya da ayni yardımların hangi kurum, kuruluş, ilaç-gereç firmalarınca yapıldığı dip not olarak bildirilmelidir. Dergide yayınlanan yazılar için herhangi bir ücret ya da karşılık ödenmez.

Yayın kurulu yazar(lar)ın dergiye gönderdikleri yazıları değerlendirme süreci tamamlanmadan başka bir dergiye göndermeyeceklerini taahhüt ettiklerini kabul eder. İnsanlar ve hayvanlar üzerinde yapılan deneysel araştırmaların bildirildiği yazıların gereç ve yöntem bölümünde, bu araştırmanın yapıldığı gönüllü ya da hastalara uygulanan işlemler anlatıldıktan sonra kendilerinin onaylarının alındığını (informed consent) gösterir bir cümle bulunmalıdır. Yazar(lar), bu tür araştırmalarda, uluslararası alanda kabul edilen kılavuzlara (2002 yılında revize edilen 1975 Helsinki Deklarasyonu- http://www.wma.net/e/policy/b3.htm, Guide for the care and use of laboratory animals - www.nap.edu/catalog/5140.html), T.C. Sağlık Bakanlığı tarafından getirilen, 29 Ocak 1993 tarih ve 21480 sayılı Resmi gazetede yayınlanan "İlaç Araştırmaları Hakkında Yönetmelik" ve daha sonra yayınlanan diğer yönetmeliklerde belirtilen hükümlere uyulduğunu belirtmeli ve kurumdan aldıkları Etik Kurul Onayı'nın bir kopyasını göndermelidir. Metin içinde standart kısaltmalar kullanılır, bunlar ilk geçtikleri yerde açık olarak yazılır. İlaç adları kullanımında ilaçların jenerik adları Türkçe okunuşlarıyla yazılır. Ölçüm birimleri metrik sisteme uygun olarak verilir; örneğin, "mg" olarak yazılır, nokta kullanılmaz; ek alırsa („) ile ayrılır. Laboratuar ölçümleri Uluslararası Sistem (US; Systéme International: SI) birimleri ile bildirilir.

Bilimsel sorumluluk

Makalelerin tüm bilimsel sorumluluğu yazarlara aittir. Gönderilen makalede belirtilen yazarların çalışmaya belirli bir oranda katkısının olması gereklidir. Yazarların isim sıralaması ortak verilen bir karar olmalıdır. Sorumlu yazar, yazar sıralamasını "Yazar Sorumluluk ve Yayım Hakkı Devir Formu'nu" doldurarak tüm yazarlar adına kabul etmiş sayılır. Yazarların tümünün ismi makale başlığının altındaki bölümde yer almalıdır.

Yayın Ücretleri

Bu dergide yayın tamamen ücretsizdir. Yayın ücreti, başvuru ücreti, makale işleme ücreti ve bir figürün, rakamın veya tamamlayıcı verinin uzunluğuna göre ek ücret ödenmesi gerekmez. İçerik öğeleri (Editörler, Düzeltmeler, İlaveler, Geri Çekmeler, Mektuplar, Yorumlar vb.) tamamen ücretsizdir.

Etik sorumluluk

Makalelerin etik kurallara uygunluğu yazarların sorumluluğundadır. Hayvanlar üzerinde yapılan deneysel çalışmalarda, çalışma protokolünün çalışmanın yapıldığı kurumdaki hayvan deneyleri etik kurulu tarafından onaylandığı belirtilmelidir. Yazarlar etik kurul onayını makale ile birlikte göndermelidir. Eğer makalede daha önce yayımlanmış alıntı yazı, tablo, resim vs. var ise yazarlar; yayım hakkı sahibi ve yazarlarından yazılı izin alarak bu durumu makalede belirtmek zorundadır. Makalenin değerlendirilmesi aşamasında yayın kurulunun gerek görmesi halinde, makale ile ilgili araştırma verilerinin ve/veya etik kurul onayı belgesinin sunulması yazarlardan talep edilebilir.

İntihal politikası

Mehmet Akif Ersoy Üniversitesi Sağlık Bilimleri Enstitüsü Dergisi'ne (MAKÜ Sag. Bil. Enst. Derg.) Gönderilen yazılar intihal açısından değerlendirilir. Her gönderilen makale, iThenticate ve Turnitin yazılımı ile intihal için kontrol edilir. Makalenin benzerlik oranı %20'nin üzerinde ise, revize edilmesi için ilgili yazara geri gönderilir. Eğer makalenin yayınlanmasından sonra intihal kanıtlanırsa, bu makale derhal web sitesinden kaldırılır ve ilgili yazarlara makalelerinin MAKÜ Sag. Bil. Enst. Derg.'de yayınlanmasının uygun olmadığı bildirilecektir.

II- Dergiye Gönderilecek Yazı Türleri ve Özellikleri

a) Araştırma Makaleleri: Bu yazılar daha önce yayınlanmamış özgün araştırma verilerinin değerlendirildiği net anlam taşıyan bilimsel çalışmaları kapsar. Araştırma makaleleri "Öz, Giriş, Gereç ve Yöntem, Bulgular, Tartışma ve Kaynaklar" bölümlerinden oluşmalıdır. Dergide yayınlanmak üzere gönderilen araştırma makaleleri kapak sayfası hariç en fazla 20 sayfa olmalıdır. Araştırma makalelerinde kullanılacak tablo, çizim ve resim sayısı toplam 10'u geçmemelidir. Yazarlar gerek duydukları takdirde "Tartışma" bölümünden sonra "Teşekkür" bölümü açarak gerekli açıklamaları yapabilirler.

b) Derleme Makaleleri: Derleme makaleleri dergi editör/yayın kurulu tarafından "çağrılı derlemeler" başlığı altında oluşturulan alnında katkı sağlama potansiyeli olan yazıları içerir. Kaynakça bölümü en fazla 30 kaynakçadan oluşturulmalıdır. Derlemelerde kullanılacak tablo, çizim ve resim sayısı toplam 10'u geçmemelidir. Kapak sayfası hariç en fazla 20 sayfa olarak hazırlanmalıdır. Derlemelerde mutlaka "Öz, Giriş, Sonuç ve Kaynaklar" bölümleri bulunmalıdır.

c) Olgu Sunumları: Yazarların, herhangi planlanmış bir araştırmaya dayanmayan ancak karşılaştıkları yeni veya ender gözlemlenen olguların ele alındığı, bilimsel değere sahip bilgileri içeren eserlerdir. Bu eserlerde gereksiz uzatmaları önlemek amacıyla en fazla 15 kaynak kullanılmalı ve bu kaynakların güncel olmasına özen gösterilmelidir. Kapak sayfası hariç en fazla 5 sayfa olmalı; "Öz, Giriş, Olgu, Tartışma ve Kaynaklar" bölümlerinden oluşmalıdır.

d) Kısa Araştırma Raporu: Dar kapsamlı ele alınmış (sınırlı sayıda örneğin analiz edildiği çalışmalar vb.) ancak önemli ve yeni bilgiler sunan bilimsel araştırmaya dayalı makalelerdir. Kısa bildiriler araştırma makalesi formatında hazırlanmalı ve kapak sayfası hariç en fazla 10 sayfa olmalıdır. Bu eserlerde kullanılacak tablo ve şekil sayısı beşi geçmemelidir.

e) Özel Bölümler:

1. Editöre mektuplar: Dergide yayınlanan yazılara ilişkin değerlendirme ve eleştirileri içeren yazılardır. Mümkün olduğunca eleştirilen yazının yazar(lar)ınca verilen yanıtlar ile birlikte yayınlanır. Editöre mektuplar 3 sayfayı geçemez.

2. Toplantı haberleri/izlenimleri: Derginin yayın alanıyla ilgili konularda yapılmış ya da yapılacak olan bilimsel toplantıları tanıtıcı yazılardır. 1 sayfayı geçemez.

3. Dergi haberleri: Derginin yayın alanıyla ilgili konularda yayınlanmakta olan bilimsel dergileri tanıtıcı yazılardır; 1 sayfayı geçemez.

4. Web siteleri tanıtımı: Derginin yayın alanıyla ilgili konulardaki web sitelerini tanıtıcı yazılardır; 1 sayfayı geçemez.

5. Kitap/tez tanıtımı: Derginin yayın alanıyla ilgili konularda yayınlanmış bulunan kitapları/tezleri tanıtan yazılardır; 3 sayfayı geçemez.

III- Makalelerin Düzenlenmesi

Dergiye gönderilecek yazılar türlerine göre, başlık sayfası, İngilizce ve Türkçe özetler, ana metin, kaynaklar, tablo/şekil/resim bölümlerini içerir. Dergiye yayınlanması için gönderilen makalelerde aşağıdaki biçimsel esaslara uyulmalıdır: Yazı Microsoft Word programında Times New Roman yazı stilinde 12 punto büyüklüğünde, siyah renkte, 1,5 satır aralığında hazırlanmalıdır. Kenarlardan 2,5 cm boşluk bırakılmalıdır. Her sayfaya satır numarası eklenmelidir.

Anatomik terimler Latincede yazıldığı gibi kullanılmalıdır. Günlük tıp diline yerleşmiş terimler ise okundukları gibi Türkçe yazım kurallarına uygun olarak yazılmalıdır. İngilizce veya başka bir yabancı dildeki şekli ile yazılan terimler tırnak içinde belirtilmelidir. Yazının başlık sayfasında, yazının Türkçe ve İngilizce başlığı ve sayfa üstünde kullanılmak üzere boşluklar da dahil 40 karakteri aşmayacak şekilde Türkçe ve İngilizce kısa başlık önerisi bulunmalı. Çalışmaların yapıldığı klinik, anabilim dalı/bilim dalı, enstitü ve kuruluşun adı belirtilmelidir.

a) Başlık Sayfası: Gönderilen makalenin kategorisini, başlığını (Türkçe-İngilizce ve sadece ilk sözcüğün baş harfi büyük), yazarların adlarını (sadece baş harfleri büyük yazılır), çalıştıkları kurumları (rakamla dipnot olarak belirtilmeli), yazışmaların yapılacağı sorumlu yazarın adı, açık adresi, telefon ve faks numaraları ile e-posta adresini içermelidir. Sorumlu yazar yıldız (*) ile belirtilir. Makale daha önce bilimsel bir toplantıda sunulmuş ise toplantının adı, tarihi ve yeri belirtilerek yazılmalıdır.

b) Ana Metin Bölümü: Yazının ana metni Öz ve Anahtar Kelimeler, Giriş, Gereç ve Yöntem, Bulgular ve Tartışma başlıkları içinde düzenlenir. Özler ve anahtar sözcükler: Türkçe ve İngilizce olmak üzere iki dilde yazılır ve yazının başlığını da içerir.

Öz 200 kelimeyi geçmemeli, çalışmanın ana noktaları olan amacını, hayvan ve örnek popülasyonunu, metodunu ve önemli sonuçlarını, çalışmadan elde edilen çıkarımı klinik olarak uygulanabilirliğini içermelidir. Yayını okumadan okuyucular için anlaşılır olmalıdır ve özet içinde kaynaklara atıf yapılmamalıdır. Türkçe ve İngilizce özetler ayrı sayfalarda yazılmalı ve özetlerin sonunda her iki dilden en az 3, en çok 5 anahtar sözcük yer almalıdır. Anahtar kelimeler Index Medicus Medical Subject Headings (MeSH)'e uygun olmalıdır. Anahtar kelimeler için www.nlm.nih.gov/mesh/MBrowser.html adresine başvurulmalıdır.

Giriş bölümünde yazının dayandığı temel bilgilere ve gerekçelere kısaca değinildikten sonra, son paragrafında amaç açık bir anlatımla yer alır. Gereç ve yöntem bölümü gerekirse araştırma/hasta/denek grubu, araçlar, uygulama ve istatistik değerlendirme gibi alt başlıklara göre düzenlenebilir. Bu bölüm çalışmaya katılmayan birisinin de rahatlıkla anlayabileceği açıklıkta yazılmalıdır. Bulgular bölümü çalışmanın sonuçlarını özetler ve temel bulgular gerekirse tablo ve şekillerle desteklenir. Tartışma bölümünde çalışmanın bulguları ilgili yurt içi ve yurt dışı çalışmaların sonuçları bağlamında tartışılır; genel bir gözden geçirmeyi değil, özgün bulguların tartışılmasını içerir. Yayın sisteme yüklenirken ana metin bölümü ana dosya olarak yüklenmelidir.

c) Teşekkür: Yazarlar çalışmalarında vermek istedikleri ek bilgiler ile katkı sağlayan destekçi kurumlara ve/veya şahıslara teşekkür yazılarını bu bölümde belirtebilirler.

d) Kaynaklar: Kaynaklar listesi alfabetik sıraya göre yazılmalıdır. Sadece yayınlanmış veya yayına kabul edilmiş kaynaklar yer almalıdır. Kabul edilmiş ancak henüz yayınlanmamış kaynaklar için "baskıda" ifadesi kullanılmalıdır. Yazarlar kaynaklar listesinde bulunan bütün kaynakların metin içinde kullanılmış olduğunu kontrol etmelidirler.

Yayındaki bütün kaynaklar kullanılmalıdır. Makale içinde referans kullanma şekline örnekler.

Metin içinde doğrudan atıf yapılırken yazar veya yazarların soyadından sonra parantez içinde kaynağın yayın yılı belirtilmelidir.

Örnekler: Bell (2005) tarafından; Nielsen ve Engberg (2006) tarafından; Doyle ve ark. (2007) tarafından

Cümlenin sonunda atıf yapıldığında ise yazar ismi ve yayın yılı parantez içinde belirtilmelidir.

Örnekler: ...bildirilmiştir (Bell, 2005); ....bildirilmiştir (Nielsen ve Engberg, 2006); .....bildirilmiştir (Doyle ve ark., 2007).

Birden çok kaynağa atıf yapılması durumunda kronolojik sıralama yapılmalıdır.

Örnekler: ….bildirilmiştir (Bell, 2005; Nielsen ve Engberg, 2006; Doyle ve ark., 2007).

Aynı yazarın aynı yıl yayınları söz konusu ise her biri "a" harfinden başlayarak küçük harflerle işaretlenmelidir.

Örnek: …. (Bell, 2005a; Bell, 2005b; Bell, 2005c …). Atıf yapılırken aşırı kaynak kullanımından kaçınılmalıdır.

Kaynaklar listesinin düzenlenmesi:

Mendeley programı kullanan yazarlar aşağıda linki verilen dergi format stilini kullanarak çalışmalarını düzenleyebilir:
https://csl.mendeley.com/styles/529990351/makusagbilensderg

Kaynaklar listesinde yazar isimleri ve yayın yılı koyu harflerle yazılmalıdır. Kaynak listesi şu şekilde hazırlanmalıdır:

i) Kaynak makale ise

Yazarların soyadları ve adlarının ilk harfi yazılmalıdır. Devamında sırasıyla makalenin yayın yılı, makalenin adı, yayınlandığı derginin açık adı, cilt, sayı ve sayfa numaraları belirtilmelidir.

Örnekler:

Cohen, N.D., Vontur, C.A., Rakestraw, P.C., 2000. Risk factors for enterolithiasis among horses in Texas. Journal of the American Veterinary Medical Association 216, 1787-1794.

Rajmohan, S., Dodd, C.E., Waites, W.M., 2002. Enzymes from isolates of *Pseudomonas fluorescens* involved in food spoilage. Journal of Applied Microbiology 93, 205-213.

Ono, K., Yamamoto, K., 1999. Contamination of meat with *Campylobacter jejuni* in Saitama, Japan. International Journal of Food Microbiology 47, 211-219.

Yayınlanmak üzere kabul edilen ve DOI numarası bulunan, ancak henüz basılmamış makaleler için; makale künyesinin sonunda DOI numarası belirtilmelidir.

McGregor, B.A., Butler, K.L., 2014. The value of visual fleece assessment in addition to objective measurements in identifying Angora goats of greater clean mohair production. Small Ruminant Research, in press (DOI: 10.1016/j.smallrumres.2014.04.001).

ii) Kaynak kitap ise

Yazarların (veya editörün) soyadları ve adlarının ilk harfi yazılmalıdır. Devamında sırasıyla kitabın yayın yılı, adı, yayınevi veya yayınlayan kuruluş ve yayınlandığı yer belirtilmelidir. Kaynak, kitaptan bir bölüm ise bölüm yazarlarının isminden sonra sırasıyla kitabın yayın yılı, bölümün adı, editörün soy ismi ve adının ilk harfi, bölümün alındığı kitabın adı, yayınevi veya kuruluş, yayınlandığı yer, bölümün sayfa numaraları yazılmalıdır.

Örnekler:

Combs, G.F., 1992. The Vitamins: Fundamental Aspects in Nutrition and Health. Academic Press, San Diego.

Concannon, P.W., 1986. Physiology and Endocrinology of Cannine Pregnancy. In: Marrow, D.A. (Ed.), Current Therapy in Theriogenology. Philadelphia, W.B. Saunders Company, pp. 491-497.

Perkins, J.B., Pero, J., 2002. Vitamin biosynthesis. In: Sonenshein, A., Hoch, J., Losick, R. (Eds.), Bacillus subtilis and Its Closest Relatives: from Genes to Cells. ASM Press, Washington D.C., pp. 271-286.

Kramer, J.M., Gilbert, R.J., 1989. Bacillus cereus. In: Doyle, M.P. (Ed.), Foodborne Bacterial Pathogens. Marcel Dekker, New York, pp. 22-70.

iii) Kaynak bir tez ise

Tezi yazan kişinin soyadı ve adının ilk harfi koyu olarak yazılmalı, kabul edildiği yıl, tezin başlığı, tezin cinsi (yüksek lisans veya doktora), üniversitesi ve enstitüsü belirtilmelidir.

Örnek:

Bacınoğlu, S., 2002. Boğa spermasında farklı eritme süreleri ve eritme sonrasında oluşturulan soğuk şoklarının spermatolojik özelliklere etkisi. Doktora Tezi, İstanbul Üniversitesi Sağlık Bilimleri Enstitüsü, İstanbul.

iv) Kaynak internette bulunan bir web sitesi ise

Yazarların soyadları ve adının ilk harfi (Yazar adı yoksa web sitesinin veya kaynağın adı) yazılır. Daha sonra sırasıyla yılı, makalenin adı, varsa yayıncı, internet adresi ve erişim tarihi belirtilir.

Örnekler:

FDA, 2001. Effect of the use of antimicrobials in food-producing animals on pathogen load. Systematic review of the published literature. http://www.fda.gov/cvm/antimicrobial/PathRpt.pdf (Erişim 14.12.2001)

Cleveland, C.W., Peterson, D.S., Latimer, K.S., 2005. An Overview of Canine Babesiosis. Clinical Pathology. College of Veterinary Medicine, The University of Georgia: http://www.vet.uga.edu/vpp/clerk/Cleveland (Erişim 17.12.2005).

Thierry, F., 2006. Contagious equine metritis: a review. Equine Reproductive Infections: http://www.equinereproinfections.com (Erişim 07.07.2006].

FSAI, 2008. Report of the Implementation Group on Folic Acid Food Fortification to the Department of Health and Children. Food Safety Authority of Ireland: http://www.fsai.ie/assets/0/86/204/cc3c2261-7dc8-4225-bf79-9a47fbc2287b.pdf (Erişim 20.06.2008)

v) Kaynak bilimsel toplantıda sunulmuş bir bildiri ise

Yazarların soyadı ve adının baş harfinden sonra sırasıyla toplantının yılı, bildirinin başlığı, toplantının adı, toplantı yeri, bildiri kitabındaki sayfa no yazılmalıdır.

Örnekler:

Cardinali, R., Rebollar, P.G., Mugnai, C., Dal Bosco, A., Cuadrado, M., Castellini, C., 2008. Pasture availability and genotype effects in rabbits: 2. development of gastro-intestinal tract and immune function of the vermiphorm appendix. In: Proc. 9th World Rabbit Congress, Verona, Italy, 1159-1164.

Mauget, R., Legendre, X., Comizzoli, P., 1998. Assisted reproductive technology in sika deer: a program to preserve endangered deer subspecies. In: Proc. 4th Int. Deer Biology Congress, Kaspovar, 185-186.

e) Tablolar: Kullanım sırasına göre numaralandırılmalı, kısa başlıklarla ifade edilmeli ve metin içinde tablo numarası verilerek (örneğin Tablo 1) atıfta bulunulmalıdır. Tablo başlıkları tablonun üst bölümüne yazılmalıdır. Tabloda kullanılan kısaltmalar ve gerekli açıklamalar tablo altında verilmelidir.

f) Şekil ve Resimler: Metinde kullanılan fotoğraflar, grafikler ve çizimler metin içinde şekil adı ile kullanılmalıdır. Şekiller kullanım sırasına göre numaralandırmalı ve kısa başlıklarla ifade edilmeli, metin içinde şekil numarası verilerek (örneğin Şekil 1) atıfta bulunulmalıdır. Şekil başlıkları şekillerin altında yer almalıdır. Şekillerde istenilen noktaya dikkat çekmek amacıyla; üzerlerine işaret konulmalı ve başlıklardan sonra yer alacak olan şekil altı notta kullanılan işaretler belirtilerek gerekli açıklamalar yapılmalıdır.

IV- Makale Süreci (Kör hakemlik)

Makale başvurusu yalnızca online olarak http://dergipark.gov.tr/maeusabed adresi üzerinden kabul edilmektedir. Sorumlu yazar, makale ile birlikte göndereceği tüm dosyaları yukarıdaki internet adresinde bulunan yeni makale gönder ikonunu tıklayarak sisteme ekleyebilir. Yazarlar dergiye gönderi yapmadan önce kayıt olmalıdır. Kaydolduktan sonra, ana sayfadaki Mehmet Akif Ersoy Üniversitesi Sağlık Bilimleri Enstitüsü Dergisi ikonuna tıklayarak; yazım kurallarına göre düzenlenmiş bilimsel çalışmayı dergi panelindeki Makale Gönder kısmından 4 basamaklı (başlarken, yükleme, kaynaklar, önizleme&gönder) gönderi işlemini yapabilir. Gönderilen makalede ön değerlendirme aşaması sırasında yazar künyeleri, çalışmanın yapıldığı kurum, etik kurul ya da özel izin adres bilgileri gibi tanıtıcı bilgiler içermemelidir. Ön değerlendirmeden (bilimsel nitelik, dil, yazım kuralları kontrolü, İntihal kontrolü iThenticate ve Turnitin programı,) geçen bilimsel çalışmaların hakem ataması yapılır. Sorumlu yazar makalenin hangi aşamada olduğunu sistem panelindeki Süreçteki Makaleler kısmından takip edebilir. Atanan hakemlere, kör hakemlik kuralları çerçevesinde çalışmanın tam metni, şekil, tablo, grafik ve resimleri sistem üzerinden yüklenerek e-posta aracılığıyla makale değerlendirme talebi gönderilir. Hakemler e-posta aracılığıyla gönderilen linke tıklayarak talebi kabul ya da reddederler. Kabul eden hakemler, kararlarını sistem üzerinden en fazla 1 ay içinde sebeplerle birlikte yüklemelidirler. Hakemin önerdiği düzeltme var ise tekrar yazara gönderilir. İstenilen düzeltmeler 1 ay içinde tamamlanıp gönderilmediği takdirde makale otomatik olarak iptal edilecektir. Editör, makalelerin yayın değerliliği ve hakemlerin görüşlerine dayanarak yayına kabul veya red kararını verir. İstenilen düzeltmeler yapıldıktan sonra makale yazar tarafından sisteme tekrar yüklenir. Derginin gizlilik bildiriminde belirtildiği gibi, yazarların kimlik bilgileri ve e-posta adresleri hiçbir şekilde başka amaçlar için kullanılmayacaktır.

Bu dergi; bilimsel araştırmaları halka ücretsiz sunmanın bilginin küresel paylaşımını artıracağı ilkesini benimseyerek, içeriğine anında açık erişim sağlamaktadır.

Mehmet Akif Ersoy University Journal of Health Sciences Institute
INSTRUCTIONS TO AUTHORS

I- Mehmet Akif Ersoy University Journal of Health Sciences Institute General Information

Mehmet Akif Ersoy University Journal of Health Sciences Intitute (MAKU J. Health Sci. Inst.) is the publication of Mehmet Akif Ersoy University Health Sciences Institute. It is published two times annualy. The journal is a peer-reviewed scientific journal in which basic and clinical scientific articles in the field of medical sciences (veterinary, medicine, dentistry, nursing and sports sciences) are published. The language of the journal is English. Papers submitted to the journal should not have been previously published, accepted for publication or be in the process of evaluation for publication in any other journal. This rule does not apply to articles presented as bulletins in scientific meetings and whose summaries are published. In such cases, however, the name, date and place of the meeting in which the paper was presented should be notified. The format of the article should be in accordance with the rules of "Uniform Requirements for Manuscripts Submitted to Biomedical Journals: Writing and Editing for Biomedical Publication (http:/www.icmje.org/)".

On receipt of the paper by the Editorial Board, the paper is evaluated for compliance with the format rules and the authors are informed about the result in four weeks. In the event that the paper is not found to comply with the general publication principles of the journal from the standpoint of either technical characteristics or general scope, the paper is rejected. Alternatively, the author(s) may be asked to re-submit the paper in accordance with the writing requirements. Papers resubmitted are passed through a similar technical examination and, if found to comply with the rules, are passed on for peer review. The paper is sent, without the title, to two reviewers selected by the board, who then assess the paper for scientific content and format compliance. When necessary the Editorial Advisory Board can send the paper to third reviewers. The selection of reviewers is ultimately at the discretion of the editor, associate Editors and/or the editorial board. The appropriate reviewers can be selected from journal's international database of reviewers listing or, if needed; independent reviewers can be determined from inland or abroad. Thereafter the Editorial Advisory Board carries out the final editing, taking the reports of the reviewers into consideration, and, when necessary, communicating with the author(s).

The Editor gives the final decision about the acceptance of the manuscript. The Editorial Board is authorized to publish the paper, return it for correction, or reject it. The assessment process involves research articles, case reports and original articles submitted to the journal. Other types of articles are evaluated directly by the Board. Papers submitted to the journal will not be returned whether they are published or not. The Editor and the Editorial Board have the right to reject, to require additional revision or to revise the format of manuscripts which do not follow the rules. The authors should inform the editorial board if they decide to withdraw the manuscript. The editor may consult editorial executive board about a manuscript if (s) he deems necessary. All the authors should submit a collectively signed statement that there is no conflict of interest regarding scientific contribution or responsibility. The association, establishment, and medication-material supply firms which have given financial, even partial, or material support to the research should be mentioned in a footnote. No fee or compensation will be paid for articles published in the journal.

The Editorial Board assumes that the author(s) are obliged not to submit the paper to another journal before completion of the assessment process. In the "method" section of articles concerned with experimental research on humans or animals, a sentence showing that the informed consent of patients and volunteers has been obtained following a detailed explanation of the interventions carried out on them. In such studies, authors should clearly state the compliance with internationally accepted guidelines (1975 Helsinki declaration revised in 2002 http://www.wma.net/e/policy/b3.htm, Guide for the care and use of laboratory animals-www.nap.edu/catalog/5140.html) issued by the Republic of Turkey Ministry of Health and published in the Official Journal dated 29 January 1993 number 21480 "Regulations Concerning Drug Research", and other more recently published rules laid out in governing statutes. They should forward a copy of the Ethic Committee Approval received from the relevant institution. Standard abbreviations used in the text are written in full when first mentioned. In the use of drugs, the generic names should be written in their Turkish pronunciation spelling

form. Measurement units are given according to the metric system; e.g. written as “mg”, no punctuation is used, in the case of extensions („) is used as a separator. Laboratory measurements are reported in International System Units (US; Systeme Internationale; SI).

Scientific responsibility

All scientific responsibility of the articles belongs to the authors. The authors of the submitted article must have a specific contribution to the work. Authors' name ordering should be a joint decision. Corresponding author is considered to accept the author sorting by filling in "Author Responsibility and Publication Transfer Form" on behalf of all authors. All of the authors should be listed under the title of article.

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Ethical responsibility

The authors are responsible for their compliance with the ethical rules. In experimental studies on animals, it should be noted that the study protocol has been approved by the animal experiment ethics committee at the institution where the study was conducted. Authors should submit the ethics committee's approval with the article. If there are previously published text, tables, pictures, etc. in the article, the authors have to get written permission from the copyright holder and the authors should specify and indicate the used material in the manuscript. In the course of the manuscript evaluation, the authors may be requested to submit the research data and / or the ethics committee approval document if deemed necessary.

Plagiarism policy

Manuscripts submitted to Mehmet Akif Ersoy University Journal of Health Sciences Institute is evaluated in terms of plagiarism. Every submitted article is checked for plagiarism through iThenticate and Turnitin software. When Smilarity Index of the article is above %20, it is sent back to the corresponding author to revise it. If plagiarism is proved after publication of the article, that article will be immediately removed from the website and the concerned authors will be considered ineligible for publication of their articles in Mehmet Akif Ersoy University Journal of Health Sciences Institute.

II- Types and Characteristics of Papers to be Submitted to the Journal

a) Research Articles: These articles are prepared in full accordance with the writing style definitions given below, in which previously unpublished original research data are evaluated. The main text section of the research articles should include (Title, Introduction Materials and Methods, Results, Discussion and Conclusion) sections and (excluding title page, bibliography, tables/figures/pictures) should not exceed 20 pages. If some parts of the research data given in these articles have previously been discussed in another paper, this must be notified without fail when sending the paper and, in addition, reference should be made to the relevant paper within the bibliography.

b) Review Articles: Review Articles should cover subjects falling within the scope of the journal which are of active current interest. They may be submitted or invited. Invited reviews will normally be solicited by the Review's Editor, but suggestions for appropriate review topics may be sent to editor.

c) Case Reports: These are articles which present and discuss the characteristics of one or more cases which have special features and scientific importance from the clinical evaluation, observation or other standpoint. Case presentations include the title page, summary, main text (includes introduction, case and discussion), bibliography,

table/figure/picture sections; subtitles in the main text are organised according to the text content. Abstracts of the case presentations should have 150 words. The main text (excluding title page, bibliography, table/figure/picture) should not exceed 10 pages.

d) Brief Reports: These are articles in which original ideas dealing with important theoretical or practical problems related to a specific subject are presented and discussed. Original articles include a title page, summary,

main text, bibliography, table/figure/picture sections; subtitles in the main text are organised according to the text content. The main text of original articles (excluding title page, bibliography, table/figure/picture) should not exceed 10 pages.

e) Special Sections:

1. Letters to the Editor: These articles include evaluation and criticisms of articles published in the journal. These are published together with the responses of the author(s) of the paper concerned where possible. Letters to the Editor may not exceed 5 pages.

2. Meeting news/notes: These articles introduce scientific meetings held or to be held on subjects within the scope of the journal. The paper may not exceed 1 page.

3. Journal news: These articles introduce scientific journals being published within the scope of the journal. The paper may not exceed 1 page.

4. Introduction of websites: These articles introduce websites relevant to the scope of the journal. These articles may not exceed 1 page.

5. Book/Thesis Section: These articles introduce books/theses published on subjects related to the scope of the journal and may not exceed 3 pages.

III- Preparation of Manuscripts

Papers to be submitted to the journal include the sections of title page, abstract, main text, references and tables/figures/pictures. Articles submitted for publication in the journal should follow the following formal principles: The text should be prepared in Microsoft Word program in Times New Roman font style with a font size of 12 font, black and 1.5 line. All side of the paper, page margins should be as 2.5 cm. Line numbers should be added to the beginning of the page.

Anatomical terms should be used as written in Latin. Running title (not exceed 40 characters) of the manuscript should add to title page. The name of the clinic, department / science, institute and institution should be stated.

a) Title Page: should contain the category, the title (only first letter capital), the names of the authors (only the first letters capital), the institution (s) where they work (indicated with numbered footnotes), corresponding author (address, phone, fax numbers and e-mail address). Corresponding author is indicated by an asterisk (*). If the article was previously presented at a scientific meeting, the name, date and place of the meeting must be stated.

b) Main Text: The main text of the paper is organised under the subtitles of Abstract and Keywords, Introduction, Materials and Methods, Results and Discussion.

Abstract and Keywords: This is written in two languages, Turkish and English, and also includes the title of the paper. The abstract is consists of 200 words. The abstract should bring out the main points of the manuscript and should include the following information: objective, the animals or sample population involved, design, the materials and methods used, the main results, a brief conclusion and clinical relevance, where applicable. They should be comprehensible to readers before they have read the paper, and abbreviations and reference citations should be avoided. At the end of the abstract, at least 3, at most 5 keywords in both languages are included.

In the introduction, following a brief statement of basic information and justifications which constitute the basis of the paper, the objective is clearly given in the last paragraph. If necessary, the "method" section may be organised according to sub-titles such as research/patient/ test group, instruments, application and statistical analysis. This section should be written with clarity so that a person not involved in the study may easily understand. Results summarize the findings of the study and, when necessary, basic findings are supported with tables and figures. In the discussion section, the findings of the study are discussed in the light of relevant national and international studies; this section includes discussion of original findings, not a general review.

c) Acknowledgements: When considered necessary, author(s) may add brief acknowledgements in a few sentences to those whose contributions to the paper are not at author level but deserve to be mentioned. Here, the contributions of those acknowledged (e.g. financial or equipment aid, technical support etc) are clearly stated (e.g. "scientific counseling", "editing of the draft", "data collection", "participation in clinical research" etc).

d) Bibliographic References:

All citations in the text should refer to: the year of publication of the reference should be indicated in parentheses after the surname of the author or authors.

Examples: Bell (2005), Nielsen and Engberg (2006), Doyle et al. (2007) were indicated that.....

The name of the author and the year of publication should be stated in parentheses at the end of the sentence.

Examples: ...were detected as 23% of the samples (Bell, 2005); ....were detected as 23% of the samples (Nielsen and Engberg, 2006); ...were detected as 23% of the samples (Doyle et al., 2007).

In case of more than one reference, references should be arranged chronologically.

Examples: ....were reported that... (Bell, 2005; Nielsen and Engberg, 2006; Doyle et al., 2007).

More than one reference from the same author(s) in the same year must be identified by the letters 'a', 'b', 'c', etc., placed after the year of publication.

Examples: (Bell, 2005a; Bell, 2005b; Bell, 2005c …)

The authors can use below formatted style link in mendeley:

http://csl.mendeley.com/styles/529990351/sagbilensderg

References should be written in alphabetical order. Reference style, the authors' names and year of publication should be written in bold. Source list should be prepared as follows:

i) Examples of journal articles:

Cohen, N.D., Vontur, C.A., Rakestraw, P.C., 2000. Risk factors for enterolithiasis among horses in Texas. Journal of the American Veterinary Medical Association 216, 1787-1794.

Rajmohan, S., Dodd, C.E., Waites, W.M., 2002. Enzymes from isolates of *Pseudomonas fluorescens* involved in food spoilage. Journal of Applied Microbiology 93, 205-213.

Ono, K., Yamamoto, K., 1999. Contamination of meat with *Campylobacter jejuni* in Saitama, Japan. International Journal of Food Microbiology 47, 211-219.

For articles that are accepted for publication and have a DOI number but not yet published; DOI number must be specified at the end of the article.

McGregor, B.A., Butler, K.L., 2014. The value of visual fleece assessment in addition to objective measurements in identifying Angora goats of greater clean mohair production. Small Ruminant Research, in press (DOI: 10.1016/j.smallrumres.2014.04.001).

ii) Books:

Combs, G.F., 1992. The Vitamins: Fundamental Aspects in Nutrition and Health. Academic Press, San Diego.

Concannon, P.W., 1986. Physiology and Endocrinology of Cannine Pregnancy. In: Marrow, D.A. (Ed.), Current Therapy in Theriogenology. Philadelphia, W.B. Saunders Company, pp. 491-497.

Perkins J.B., Pero, J., 2002. Vitamin biosynthesis. In: Sonenshein, A., Hoch, J., Losick, R. (Eds.), Bacillus subtilis and Its Closest Relatives: from Genes to Cells. ASM Press, Washington D.C., pp. 271-286.

Kramer, J.M., Gilbert, R.J., 1989. Bacillus cereus. In: Doyle, M.P. (Ed.), Foodborne Bacterial Pathogens. Marcel Dekker, New York, pp. 22-70.

iii) Thesis:

Bacınoğlu, S., 2002. Boğa spermasında farklı eritme süreleri ve eritme sonrasında oluşturulan soğuk şoklarının spermatolojik özelliklere etkisi. Doktora Tezi, İstanbul Üniversitesi Sağlık Bilimleri Enstitüsü, İstanbul.

iv) Web site or author is an institution:

FDA, 2001. Effect of the use of antimicrobials in food-producing animals on pathogen load. Systematic review of the published literature. http://www.fda.gov/cvm/antimicrobial/PathRpt.pdf (Accessed: 14.12.2001)

Cleveland, C.W., Peterson, D.S., Latimer, K.S., 2005. An Overview of Canine Babesiosis. Clinical Pathology. College of Veterinary Medicine, The University of Georgia: http://www.vet.uga.edu/vpp/clerk/Cleveland (Accessed: 17.12.2005).

Thierry, F., 2006. Contagious equine metritis: a review. Equine Reproductive Infections: http://www.equinereproinfections.com (Accessed: 07.07.2006).

FSAI, 2008. Report of the Implementation Group on Folic Acid Food Fortification to the Department of Health and Children. Food Safety Authority of Ireland:http://www.fsai.ie/assets/0/86/204/cc3c2261-7dc8-4225-bf79-9a47fbc2287b.pdf (Accessed: 20.06.2008).

v) Paper presented at a scientific meeting

Cardinali, R., Rebollar, P.G., Mugnai, C., Dal Bosco, A., Cuadrado, M., Castellini, C., 2008. Pasture availability and genotype effects in rabbits: 2. development of gastro-intestinal tract and immune function of the vermiphorm appendix. In: Proc. 9th World Rabbit Congress, Verona, Italy, 1159-1164.

Mauget, R., Legendre, X., Comizzoli, P., 1998. Assisted reproductive technology in sika deer: a program to preserve endangered deer subspecies. In: Proc. 4th Int. Deer Biology Congress, Kaspovar, 185-186.

e) Tables: Each table is printed on a separate page and numbered according to the sequence of referral within the text (Table 1). Each table has a title and, when necessary, explanations are given under the table (e.g. abbreviations given in the table). Each table should be understandable without need for referral to the text. Each table should be referred to in the text..

f) Figures and Pictures: Figures should be numbered according to the order of use and should be expressed with short titles.Figures should be numbered in the text (Figure 1). Letters, numbers and symbols within the figure should be clear and readable when downsized for printing.Each figure should be referred to in the text..

IV- Submission of Articles (Blind Peer-Review)

The article submission is only accepted online via 'http://dergipark.gov.tr/maeusabed' The Corresponding authors, all the files can be added to the system by clicking the submit new article icon at the above address. Authors must register on Dergipark system before submitting a manuscript. After signing up, clicking Mehmet Akif Ersoy University Journal of Health Sciences icons on the main page, the manuscript written according to the guide for authors is submitted in 4 steps (start, submission, reference, preview & submit). The submitted manuscript must not contain any identifying information, such as author information, institution, ethics committee or special permit address, during the preliminary evaluation phase. The manuscript that pass the preliminary evaluation (paper scientific qualification, language, conformity to Guide for author and checking plagiarism via

iThenticate and Turnitin program,) are assigned to the Reviewers. The corresponding author can follow the article evaluation process from the section on the Articles in the Process. According to the blind peer-review rules, the main text, tables, graphics and pictures of the manuscript are uploaded via the system and sent to the appointed reviewers for an article evaluation request via e-mail. The reviewers accept or reject the request by clicking on the link sent via e-mail. The reviewers who accept it have to upload their decisions together with the reasons within a maximum of 1 month via the system. If the correction requested by the Reviewer is sent back to the author. If the requested corrections are not completed within 1 month, the article will be automatically canceled. After the desired corrections are made, the article is uploaded back to the system by the author. The editor makes decisions to accept or reject papers based on their opinion of the papers' publication worthiness and reviewers' comments. As stated in the privacy statement, authors' identity information and e-mail addresses will not be used for any other purpose.

MEHMET AKİF ERSOY ÜNİVERSİTESİ SAĞLIK BİLİMLERİ ENSTİTÜSÜ DERGİSİ

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Sağlık Bilimleri Enstitüsü
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Araştırma Makaleleri / Research Articles

Olgu Sunumları / Case Reports

MAKU J. Health Sci. Inst. 2023, 11(2): 212-220.
doi: 10.24998/maeusabed.1297698

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Journal of Health Sciences Institute
http://dergipark.gov.tr/maeusabed

Research Article / Araştırma Makalesi

Comparison of Effects of Water-Based and Land-Based High-Intensity Interval Training on Aerobic Capacity and Spinal Stabilization

Su içinde ve Karada Yapılan Kısa Dönem Yüksek Şiddetli Aralıklı Egzersiz Eğitiminin Aerobik Kapasite ve Spinal Stabilizasyon Üzerine Etkilerinin Karşılaştırılması

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Abstract: The purpose of the study is to examine the effects of a two-week period of high-intensity interval training (HIIT) in water and land-based running on aerobic capacity and spinal stabilization. Forty-one healthy young participants were divided into water-based exercise, land-based exercise, and control groups. Aerobic capacity was measured with 20-m Shuttle Run Test, and spinal stabilization was tested with 60° Flexion test and Sorensen test. Water and land-based exercise groups performed six sessions of HIIT program for two weeks. A significant improvement in aerobic capacity and spinal stabilization was found between the baseline and after two-week evaluations in both the water and land based HIIT exercise groups ($p<0.05$). The increase in aerobic capacity and extensor spinal muscle endurance were found similar in both exercise group yet increase in flexor spinal stabilization endurance was statically more significant in land exercises ($p= 0.038$). This study shows that the two-week period of HIIT exercises, both in water and on land, can be safely used to improve aerobic capacity and spinal stabilization, even in the short term.

Keywords: Aerobic capacity, Aquatic exercise, Core stability, HIIT.

Öz: Çalışmanın amacı, suda ve karada yapılan iki haftalık yüksek şiddetli aralıklı egzersizin (YŞAE) aerobik kapasite ve spinal stabilizasyon üzerindeki etkilerini incelemektir. Kırk bir sağlıklı genç katılımcı, su içi egzersiz, kara egzersizleri ve kontrol grubu olarak ayrıldı. Aerobik kapasite 20 m Mekik Koşusu Testi ile, spinal stabilizasyon ise 60° Fleksiyon testi ve Sorensen testi ile ölçüldü. Su ve kara egzersiz grupları iki hafta boyunca altı seanslık YŞAE programı gerçekleştirdi. Hem su hem de kara YŞAE egzersiz gruplarında başlangıç ile iki haftalık değerlendirmeler arasında aerobik kapasite ve spinal stabilizasyonda anlamlı bir iyileşme bulundu ($p<0.05$). Aerobik kapasite ve ekstansör spinal kas enduransındaki artış her iki egzersiz grubunda benzer bulunurken, fleksör spinal stabilizasyon enduransındaki artış kara egzersizlerinde istatistiksel olarak daha anlamlıydı ($p= 0.038$). Bu çalışma, hem suda hem de karada iki haftalık YŞAE egzersizlerinin kısa sürede bile aerobik kapasiteyi ve omurga stabilizasyonunu geliştirmek için güvenle kullanılabileceğini göstermektedir.

Anahtar Kelimeler: Aerobik kapasite, Su içi egzersizler, Core stabilite, YŞAE.

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Introduction

Physical inactivity has an essential role in contemporary health problems. Moreover, it also causes many systemic diseases and infections (Lee et al., 2012). The various barriers to physical activity have been the subject of many studies to date; however, the most common cause of physical inactivity is 'timelessness' (Trost, Owen, Bauman, Sallis, & Brown, 2002).

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High-intensity interval training (HIIT) is a repetitive exercise stage consisting of severe exercise periods interspersed with recovery intervals. Depending on the severity of the exercise, the severe exercise period can extend from a few seconds to a few minutes, with recovery periods consisting of rest or low-intensity exercises (Gibala & McGee, 2008). HIIT has been mainly used for athletes; however, recent studies show that HIIT is used in clinics and also in healthy populations (Broman, Quintana, Lindberg, Jansson, & Kaijser, 2006; Gillen et al., 2014; Nagle, Sanders, & Franklin, 2017; Wilber, Moffatt, Scott, Lee, & Cucuzzo, 1996).

The essential characteristic of HIIT from other aerobic exercises is that it is effective in periods as short as two weeks. People report seeing similar effects on their body composition as they would over a longer time period (Gillen et al., 2014).

Although a growing body of research shows that HIIT is an effective, safe, time-efficient, and enjoyable exercise, there is no consensus on training protocol or exercise principles. It is possible to do HIIT in water as well as on land. Despite the publication of a number of studies of water-based HIIT exercises, we found none focused on the comparison of water-based and land-based HIIT in short term (Broman et al., 2006; Mohr et al., 2014; Nagle et al., 2017; Rebold, Kobak, & Otterstetter, 2013; Wilber et al., 1996).

Accordingly, we aimed to investigate the comparison of the effects of deep water running (DWR) as HIIT and land-based running on aerobic capacity and spinal stabilization over a two-week period.

Materials and Methods

This study was conducted between February 2016 and May 2016, at the Near East University, Department of Physiotherapy and Rehabilitation. The Olympic swimming pool on the university campus used for water-based exercise. Two specialist physiotherapists were involved, one applied the evaluation procedure, and the other carried out the training sessions. Both the participants and therapists were blinded in the trial. Written informed consent was obtained from all participants. The Ethical Committee approved the study protocol of Dokuz Eylül University in Izmir, Turkey (Approval No. 2016/06-40).

Participants

The power analysis was calculated by G*power software and based on the effect size 0.65 obtained, assuming a power of 0.8 at an alpha level of 0.05. The sample size computed was 13 or more subjects per group. Considering dropout's probability 15 volunteers were recruited in each group.

Forty-five healthy young voluntary participants, the ages of 18-25 years were included to this study. They were divided into three groups: water-based exercise, land-based exercise, and controls randomly.

After the elimination of the applicants, computer-generated randomization was used for the grouping of the individuals. The volunteers were excluded if they had cardiovascular or musculoskeletal disease, any obstacle for exercise, chlorine allergy, or difficulties with water immersion. The characteristics of the participants are summarized in Table-1.

Outcomes

Participants performed the 20 m Shuttle Run Test (SRT) to determine the aerobic capacity (VO_{2max}). In this test, participants run back and forth on a 20 m course, touching the 20 m line each time; at the same time, a sound signal is emitted from a pre-recorded tape. The frequency of the sound signals is increased by 0.5 km h^{-1} each minute from a starting speed of 8.5 km h^{-1}. When the subject can no longer keep the pace, the last stage number announced is used to predict maximal oxygen uptake (VO_{2max}) (S. Ahmaidi, K. Collomp, C. Caillaud, & C. Prefaut, 1992; Hızal, Açıkada, Hazır, & Tınazcı, 1997).

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Table 1. The characteristics of the participants

Variables	Land-based exercise $\bar{x}$ ±SD	Pool-based exercises $\bar{x}$±SD	Control $\bar{x}$±SD	p
Age (year)	21.66±2.97	22.00±2.97	20.35±1.08	0.21
Height (cm)	171.00±8.10	170.46±10.03	168.14±9.14	0.62
Weight (kg)	66.26±11.18	68.76±13.80	61.78±10.41	0.33
BMI (kg/m²)	22.54±2.41	23.66±4.45	21.70±1.65	0.35

Kruskal Wallis Variance Analysis, X:Mean, SD: Standart Deviation, BMI: Body Mass Index

VO2max was calculated by placing the result calculated with the age of the participant, and velocity determined by using the distance covered in 30 seconds during the last stage of the test (S Ahmaidi, K Collomp, C Caillaud, & C Prefaut, 1992).

VO2max = 31.025 + (3.238 x velocity) - (3.248 x age) + (0.1536 x age x velocity)

Spinal stabilization was tested a 60° Flexion test and Sorensen test.

60° Flexion test: Participants sat on the test bench and placed their upper body against a support with an angle of 60° from the testbed. Both the knees and hips were flexed to 90°. The arms were folded across the chest, hands were placed on opposite shoulders, and toes were tucked under the toe straps. Participants maintained this position while the researcher moved the supporting wedge back 10 centimeters from the subject's back to begin the test. Time until the upper body fell below 60° was recorded (Anderson, Barthelemy, Gmach, & Posey, 2013).

Sorensen test: Participants lied on an examination table in prone position with their lower body was strapped to the bench. The edge of table was aligned with the anterior–superior iliac spines. They were asked to hold the position as long as they could while their hands were folded across their chest. The test was terminated if they had no difficulty longer than 4 minutes.

Interventions

After a five-min warm-up (stretching and jogging), participants wore a waist belt attached to pool wall with rope and practiced high knee style DWR in a "5 m Olympic pool". They performed six sessions of HIIT program over two weeks. This training includes five sets of one-minute high-intensity running at 16-20, and three-minutes moderate-intensity running at 11/20, according to Borg Scale (Borg, 1982; Killgore, Wilcox, Caster, & Wood, 2006). Borg scale was explained and demonstrated to the participants in a separate session and was continually monitored during exercise sessions.

The same HIIT exercise protocol was used for the land-based group in the exercise room. There was no exercise training performed in the control group, who were asked to maintain their regular physical activity level. Assessments were performed by the same researcher at the beginning and the end of the two-week period.

Statistical analyses

SPSS Version 22.0 (IBM Corporation, Armonk, NY, USA) was used for all statistical analyses. The Shapiro–Wilk test indicated that the variables did not have a normal distribution. Kruskal Wallis variance analysis test was used to compare the three groups. Wilcoxon tests were applied to determine the results from dependent groups. The Mann-Whitney U test was used to compare the differences between two independent groups. Significant results were then analyzed by post-hoc tests (Wilcoxon signed ranks test with Bonferroni correction) in which the significance level was set at 0.017 (0.05/3) after Bonferroni correction.

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Results

Four participants were failed to show up to the last assessment therefore, forty-one participants with the mean age of 21.33±2.29 years and average BMI 22.61±3.04 kg/m² were concluded the study. There were no significant baseline differences between groups ($p>0.05$) (Table 1).

After two weeks of HIIT, aerobic capacity increased significantly in both water and land-based groups ($p<0.05$). There was no difference was found in aerobic capacity of control group.

Spinal stabilization was significantly increased in both exercises groups. There was no statistically significant difference regarding Sorenson Tests scores between two exercise groups ($p= 0.038$). However, 60° Flexion test scores were statistically higher in land exercise group than water exercise group ($p< 0.001$) (Table 2).

A comparison of baseline and two weeks later measurements revealed significant differences between the water and control groups, and also between land-based exercise and control groups ($p<0.05$).

Discussion

The results of our study show that both water and land based HIIT exercises are effective in the improvement of aerobic capacity and spinal stabilization even in a period as short as two weeks. Also, compared to water-based HIIT, land-based HIIT was more effective on spinal stabilization. However, there is no significant effect on the aerobic capacity results between the two groups. To our knowledge, this is the first study aiming to compare HIIT exercises in water and on land. The results showed that HIIT is safe and effective in increasing aerobic capacity and spinal stabilization, even over a two-week period.

In previous studies, it is well documented that HIIT is an effective method in improving aerobic capacity in water and land (Nagle et al., 2017; Talanian, Galloway, Heigenhauser, Bonen, & Spriet, 2007), and the results of our study were consistent with these.

Comparing exercise responses between water and land conditions is a complex subject as there are many influencing factors. The intensity of the aerobic exercise is generally monitored by heart rate; however, heart rate changes not only by immersion in water but also depending on water temperature (B. E. J. P. Becker, 2009). On the other hand, exercise intensity changes according to whether subjects are stationary or moving in the water. Due to viscosity, faster movements in water create greater resistance (B. E. Becker, 2020; Cole & Becker, 2004). Therefore, any change in the experiment design directly effects the exercise responses, which complicates the comparison of land and water exercises.

Comparisons of DWR and treadmill exercises in previous studies (Butts, Tucker, & Greening, 1991; Butts, Tucker, & Smith, 1991) showed that treadmill exercises require higher maximum oxygen uptake (VO_{2max}), and ventilation volume. These studies restricted participants by attaching them to the pool wall, and exercise load on land was determined by the speed and inclination of the treadmill, whereas cadence was used in the water run. Authors noted that adjusting the workload in water was inefficient and awkward.

They also reported the heart rate was lower in DWR, yet they did not use the immersion heart rate lower than land. Michaud et al compared water and land running at the same exercise intensity (75% of VO_2 Peak) and found that blood lactate and perceived of excursion were significantly higher, whereas heart rate was significantly lower in DWR (Michaud, Rodriguez-Zayas, Andres, Flynn, & Lambert, 1995).

Town and Bradley, observed that both running in shallow water and treadmill HIIT increased VO_{2max} more than deep water running (Town & Bradley, 1991). Kruel et al found that treadmill exercises increased VO_{2max} and muscle activity more than DWR (Kruel et al., 2013).

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Table 2. The comparison of exercise effectiveness between the three groups.

	Aquatic Group (n=16) Median (Quartiles)		**Land Group (n=13)** Median (Quartiles)		**Control Group (n=17)** Median (Quartiles)		
	Before	**After**	**Before**	**After**	**Before**	**After**	**p value**
VO2max (ml/kg/dk)	28,7 (2250/31.3)	31.0*c (28.3/34.2)	28,4 (26.5/31.0)	30.8*b (27.4/35.2)	25.7 (24.2/26.6)	25.3 (23.7/26.5)	
Δ	2.2 (0.7/2.2)		2,3 (0.8/4.5)		-0.4 (-0.8/0.5)		**p<0.001#**
60° flexion test (sec)	51.0 (41.7/62.2)	70.4* c (55.6/85.7)	43.8 (36.9/50.7)	105.0*a b (80.8/121.9)	40.2 (33.3/48.1)	39.7 (31.4/47.9)	
Δ	19.4 (8.9/30.1)		61.2 (40.5/86.2)		-0.5 (-1.2/2.8)		**p<0.001#**
Sorensen Test (sec)	59.6 (47.4/71.8)	80.7* c (68.1/92.2)	35.6 (25.3/41.2)	86.7* b (66.7/102.4)	38.2 (29.1/47.2)	37.5 (8.3/46.8)	
Δ	21.0 (13.2/29.3)		51.0 (28.7/79.8)		-0.7 (2.3/3.5)		**p<0.001#**

VO2max: Maximum Oxygen Uptake; Kruskal Wallis Test #p<0.05; Wilcoxon Signed Rank Test *p<0.05 within groups;
a-c (post-hoc Mann–Whitney U test between groups test with Bonferroni correction resulting in a significance level of p< 0.017).
a Aquatic versus land; b Land versus control; c Aquatic versus control.

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The researchers underline two factors which limit VO_{2max} and spinal muscle activity improvement during DWR. The first is the lack of contact between the extremities and the floor. The other is the support of the belt, which takes the bodyweight during DWR (Masumoto, Applequist, & Mercer, 2013; Oddsson, 2019).

Masumuto et al. applied EMG tests on four muscles, and they showed that treadmill exercises increased muscle activity more than DWR. They observed that muscle activity was affected by the mode of DWR (Masumoto et al., 2013; Masumoto, Horsch, Agnelli, McClellan, & Mercer, 2014; Masumoto, Mefferd, Iyo, & Mercer, 2018; Masumoto, Takasugi, Hotta, Fujishima, & Iwamoto, 2005). Also, DWR and treadmill exercises gave similar results in rate of perceived exertion when applied at an equal heart rate. Unlike these studies, we used DWR and running as HIIT, thus found similar results. To our knowledge, no study has investigated the effects of water based HIIT on spinal stabilization. Bressel et al. gave 11 spinal stabilization exercises in shallow water (Eadric Bressel, Dolny, & Gibbons, 2011), and reported higher EMG activity of spinal stabilization muscles except for erector spines compared to land-based exercises. These results can be explained with the effect of hydrostatic pressure and buoyancy, which suggests that trunk muscles provide less stabilization in water. Pöyhönen and Avela explained the decrease of EMG activity as being due to hydrostatic pressure causing the presynaptic inhibition in inter-neuronal pathways by stimulating the body's mechanoreceptors (Poyhonen & Avela, 2002).

Meanwhile, according to their study results, the muscular temperature affected the frequency and magnitude in shallow water. Studies using EMG and computer techniques have shown that muscle endurance depends on the co-activation of all trunk muscles, and is not more effective on one muscle than on another (E. Bressel, Dolny, Vandenberg, & Cronin, 2012). Masumuto et al. examined the effect of land-walking and water-walking exercises on the trunk and lower extremity muscle activation (Masumoto et al., 2005). Also, they observed land exercise EMG results, which were found to be higher in all muscles except paraspinal muscles. They also argue that water exercise against a current increases muscle activity more than other types of water exercises. Our study results showed that 2 weeks HIIT DWR increased spinal muscle endurance, land based exercises were more effective for trunk flexors than water based exercises. It is due to the elimination of the gravitational force, which creates resistance on land during running, in the water. There were no statistically significant difference regarding trunk extensors between on land and in water this could be the result of increased paraspinal muscle activation (Masumoto et al., 2005) balancing elemination of gravitational forces.

As a result of our study, we can say that in parallel with the literature, the two-week HIIT training improves the aerobic capacity with the increase of the VO_{2max} level. In different populations, HIIT results were found to be similar. Studies have shown that HIIT increases VO_{2max} more than other exercise modalities in healthy people (Cicioni-Kolsky, Lorenzen, Williams, & Kemp, 2013; Helgerud et al., 2007), those with the cardiometabolic disease (Weston, Wisloff, & Coombes, 2014), athletes (Etxebarria, Anson, Pyne, & Ferguson, 2014; Yang, Lee, Hsu, & Chan, 2017) and adolescents (Costigan, Eather, Plotnikoff, Taaffe, & Lubans, 2015). According to the results of the lumbar stabilization tests in our study, we can say that short-term HIIT on land was more effective than water based HIIT. Bayraktar et al (Bayraktar et al., 2016) gave training to core stabilization in water and land for three days a week for eight weeks. The featured result was the improvement in the core stabilization parameters of both groups, and there was no difference between them. In literature, no study investigated the effect of HIIT on the core muscles. Another study about HIIT on low-back pain concluded that these exercises retained motivation leading to better rehabilitation (Verbrugghe et al., 2018). Our study was

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performed in healthy young individuals, compatible with literature.

Moreover, this study is distinct from the literature as it compares the effects of water-based and land-based HIIT. Although the examples of HIIT were previously studied in the water and on land using different groups, no studies were found comparing water and land to assess which is the most effective exercise environment. Our research shows that water and land based HIIT have similar results, and both are beneficial. The number of studies about the effects of HIIT in clinical populations is constantly increasing day, and this approach is widely used in the athletic group. However, there are no HIIT guideline for clinical or healthy populations (Taylor et al., 2019). The intention of the study was to compare the short-term effects of HIIT in water and on land that is why long-term effects could not be interpreted by this paper. There is therefore a need for studies to compare the effects of HIIT on land and in water on individuals in different diagnostic groups.

Another important aspect of our work is the comparison of land and water based HIIT as short-term effects. This study shows that HIIT has potential to increase cardiovascular fitness in healthy people. There are some limitations in our study: muscle strength could not be assessed objectively due to equipment failure, and aerobic capacity was calculated indirectly by the shuttle run test.

Conclusions

In summary, according to the results of this study, six sessions of HIIT have produced positive results in the core stabilization with no adverse effect. The two-week HIIT appears to increase spinal stabilization both in the water and on land, confirming our hypothesis. Another finding of this study was HIIT is widely used in the clinic because it increases aerobic capacity and spinal stabilization on land and in water, even when applied for a relatively short time. It provides motivation, time-saving, and enjoyment. HIIT will be a solution, especially for those who complain about lack of time.

For this reason, it is potentially a valuable option for the clinician in the athletic, clinical, and healthy population. This is an advantage when HIIT is performed in water, and especially as group exercises. A significant increase in muscle stabilization in the short term of HIIT may be considered as an important contribution to the literature. In view of the limited number of studies with water based HIIT, there is a need for further studies on HIIT in water and on land.

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Research Article / Araştırma Makalesi

Determination of Phlebitis Rate with Visual Infusion Phlebitis Diagnostic Scale: An Observational Study

Görsel İnfüzyon Flebit Tanılama Skalası ile Flebit Oranının Belirlenmesi: Gözlemsel Bir Çalışma

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Abstract: Phlebitis is the inflammation of the tunica intima layer of the vein and is a common preventable complication of peripheral intravenous catheters. The aim of this study was to determine the rate of phlebitis development in patients with peripheral intravenous catheters implantation using the Visual Infusion Phlebitis Diagnostic Scale and to determine the associated factors. The study was conducted in the internal and surgical clinics of a state hospital hospital between September 1, 2022 and February 1, 2023. The 'Patient Information Form', 'Peripheral Intravenous Catheter Evaluation Form' and 'Visual Infusion Phlebitis Diagnosis Scale' were used to collect the study data. In 30.3% of the patients with PIC, phlebitis developed and 48.9% of the phlebitis cases were grade 1. The variables such as sex and presence of a chronic disease did not affect the development of phlebitis. Phlebitis development was more common in patients in the 40-64 age group, on the wrist, on the left arm and on the actively used arm, in sites where the peripheral intravenous catheters was inserted in two or more attempts, in patients in whom 16 Fr or 18 Fr catheters were inserted, and in patients taking antibiotics.

Keywords: Phlebitis, intravenous cathat, Phlebitis Diagnostic Scale, Intravenaous Access.

Öz: Flebit; venin tunika intima tabakasının inflamasyonudur ve yaygın görülen önlenebilir bir periferik intavenöz katater komplikasyonudur. Çalışmada periferik intavenöz katater takılı olan hastalarda flebit gelişme oranlarının ile belirlenmesi ve ilişkili faktörlerin ortaya konması amaçlanmıştır. Çalışma, 1 Eylül 2022-1 Şubat 2023 tarihleri arasında bir devlet hastanesinin dahili ve cerrahi kliniklerinde yürütülmüştür. Veriler 'Hasta bilgi formu', 'Periferik İntravenöz Kateter Değerlendirme Formu' ve 'Görsel İnfizyon Flebit Tanılama Skalası' ile toplanmıştır. Hastalara uygulanan 122 PİK girişiminin %30.3'ünde flebit geliştiği ve gelişen flebitlerin % 48,9'unun 1. Derece flebit olduğu belirlenmiştir. Cinsiyet ve kronik hastalık varlığının flebit oluşumunu etkilemediği, 40-64 yaş grubunda, el bileğinde, sol tarafta ve aktif olarak kullanılan tarafta, ikinci veya tekrarlı denemelerde, 16 ve 18 numara kateterle takılan periferik intavenöz kataterlerde ve antibiyotik kullanan hastalarda flebit oluşumunun arttığı belirlenmiştir.

Anahtar Kelimeler: Flebit, İntravenöz katater, Flebit tanılama skalası, İntravenöz girişim.

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Introduction

Peripheral intravenous catheters (PIC) are used to administer some drugs, to replace fluid and electrolyte loss, to regulate acid-base balance, to administer blood and blood products, and to provide total parenteral nutrition (Craven et al., 2015). Although PIC practices are among the frequently used nursing practices, they lead to some complications. Among these complications, phlebitis and infiltration are the most common, but they can be prevented if the care of the PIC area is inserted is performed regularly and properly (Gallant and Schultze, 2006).

Phlebitis is the inflammation of the tunica intima layer of the vein and is a common preventable complication of PIC (Craven et al., 2015). Post-infusion phlebitis develops within 24-96 hours after the termination of PIC application (Urbanetto et al., 2017). Phlebitis development rates differ from one sample group to another and

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there are many factors associated with the development of phlebitis. Among these factors are the type of the catheter, drug irritation, antibiotic use, the person's age and sex, fluid flow rate, frequency of drug use, anatomical region the catheter is inserted, and other infections in the patient (Urbanetto et al., 2017).

Nurses assume an important role in the prevention of the development of PIC-related phlebitis and infiltration, and in the provision of appropriate care if phlebitis has developed. The development of PIC complications causes patients to be exposed to unnecessary diagnostic procedures and treatment, and prolonged hospitalization, to experience stress, and increases the workload of health personnel and the cost of health expenditures. In order to prevent PIC complications, nurses should adopt good and preventive care practices and be able to make an urgent decision on care in case a complication occurs.

In the present study, the authors aimed to determine the rate of phlebitis development in patients with PIC by administering the Visual Infusion Phlebitis Diagnostic Scale and to reveal the related factors.

Materials and Methods

Design and setting

This cross-sectional study was conducted in the internal and surgical clinics of a public hospital between September 1, 2022 and February 1, 2023.

Sample

The study included 122 catheterized patients hospitalized in internal and surgical clinics between the above mentioned dates. Patients hospitalized in intensive care clinics were excluded from the study due to multiple interventions and the presence of central catheters. They were monitored for phlebitis development for 3 days after catheter insertion. After the study was completed, post power analysis was performed, which demonstrated that the sample had adequate power (80%). Standardized effect sizes determined by Cohen were used to determine the power of the sample size (Cohen et al., 2007).

Data Collection Tools

The 'Patient Information Form', 'Peripheral Intravenous Catheter Evaluation Form' and 'Visual Infusion Phlebitis Diagnosis Scale' were used to collect the study data.

***Patient Information Form**:* The form prepared by the researchers based on the literature is administered to determine patients' socio-demographic characteristics such as age and sex. It is also used to determine whether the patient has a chronic disease.

***Peripheral Intravenous Catheter Assessment Form**:* The form developed by the authors of this study is used to question the catheter size, the catheter insertion site, the body area where the catheter is inserted, the body side that the patient actively uses, the success at the first attempt, and the use of antibiotics. It is also used to question whether the previous insertion was performed on the same vein.

Visual Infusion Phlebitis Diagnosis Scale (GIFTS): The GIFTS was developed by Gallant and Schultze in 2006. The language and content validity study of the Turkish version of the GIFTS was performed by Paşalıoğlu and Kaya (2014).The GIFTS used to observe the catheter for possible risks and/or the relevant symptoms in case phlebitis develops in patients in whom PIC is inserted includes the following five grading steps:

Grade 1: At this grade, there are no signs of phlebitis such as pain, redness, or edema. The only recommendation is to observe the catheter.

Grade 2: At this grade, early signs of phlebitis such as redness smaller than 2.5 cm around the catheter and pain on palpation occur. It is recommended to replace the catheter with a new one.

Grade 3: It is the middle stage of phlebitis. At this grade, there is redness which is greater than 2.5

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cm, but smaller than 5 cm around the intravenous (IV) region. There is also pain on and/or around the IV region, and stiffness around it when the region is palpated. It is recommended to replace the catheter with a new one, to notify the physician and to provide the treatment ordered by the physician.

Grade 4: It is the advanced phlebitis or initial stage of thrombophlebitis. At this grade, there is redness of 5 cm or more in the IV region, pain and stiffness on or around the IV region when the region is palpated. It is recommended to replace the catheter with a new one, to notify the physician and to provide the treatment ordered by the physician.

Grade 5; It is the advanced stage of thrombophlebitis. At this grade, there are signs of grade 4 phlebitis and signs of purulent drainage. It is recommended to replace the catheter with a new one, to notify the physician and to provide the treatment ordered by the physician.

Data analysis

The statistical analysis of the data obtained in the present study was performed on the computer using the Statistical Package for the Social Sciences (SPSS) 16.0 (SPSS Inc., Chicago, IL, USA) program. Descriptive statistics were given as numbers and percentages, and the Chi-square test was used to compare the presence of phlebitis according to independent variables.

Ethical consideration

Before the study was conducted, ethical approval from Burdur Mehmet Akif Ersoy University Non-Interventional Research Ethics Committee (Date: 11/05/2022 and Decision no: GO 2022/715), permission to conduct the study from Burdur State Hospital where the study was to be conducted and written informed consent from the patients or their relatives who agreed to participate in the study were obtained.

Table 1. Patient characteristics

	n	%
Age		
18-39	24	19.7
40-64	70	57.4
65 and older	28	23.0
Sex		
Female	70	57.4
Male	52	42.6
Chronic disease		
Yes	89	73
No	33	27
Clinic		
Internal	48	39.3
Surgical	74	60.7

Results

In the study, 122 patients with PIC were included. Of them, 57.4% were women and in the age group of 40-64 years, 73% had at least one chronic disease and 60.7% were hospitalized in surgical clinics (Table 1). In all the patients, nurses preferred the upper extremity to insert the PIC. The PIC was mostly inserted in the 30.3% forearms of the patients, in the left arm of 76.2% of the patients, and in the arm that was not actively used in 73% of them. The PIC was inserted in the first attempt in 59.8% of the patients, and 42.6% of the PICs were inserted in the previous insertion site. The 22 Fr catheters were inserted in 38.5% of the patients, and 37.7% of the patients were administered antibiotics via PIC (Table 2).

According to the analysis of the phlebitis development in terms of the clinic where the patients were hospitalized, their sex, and the presence of a chronic disease demonstrated that there was no statistically significant difference between them (Table 3, $p>0.05$).

Table 2. Peripheral intravenous catheterization and related factors.

Variables	n	%
Catheter insertion site		
Dorsal face of the hand	29	23.8

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Wrist	34	27.9
Forearm	37	30.3
Antecubital region	22	18.0
Catheter inserted side		
Right	29	23.8
Left	93	76.2
Actively used arm		
Yes	33	27
No	89	73
Success in the first attempt	73	59.8
Yes	49	40.2
No		
Previous insertion in the same vein	52	42.6
Yes	70	57.4
No		
Catheter number (Fr.)		
16	2	1.6
18	31	25.4
20	42	34.4
22	47	38.5
Antibiotic administration		
Yes	46	37.7
No	76	63.3
Phlebitis No		
Grade 1	85	69.7
Grade 2	23	18.9
Grade 3	10	8.2
Grade 4	4	3.3
Grade 5	0	

According to the analysis of the phlebitis development in terms of the age groups of the patients, phlebitis development was higher in the 40-64 age group than it was in the other age groups and the difference was statistically significant (Table 3, $p<0.05$).

According to the analysis of the phlebitis development in terms of the sites where the PIC was inserted, phlebitis development was more common on the wrist, left arm and actively used arm, and the difference was statistically significant (Table 3, $p<0.05$).

According to the analysis of the phlebitis development in terms of the number of PIC insertion attempts, phlebitis development was more common if there were two or more attempts, or if the previous insertion had been performed on the same vein. The difference was statistically significant (Table 3, $p<0.05$).

According to the analysis of the phlebitis development in terms of the size of the catheters, phlebitis developed in all the patients in whom 16 Fr or 18 Fr catheters were inserted (Table 3, $p<0.05$).

Discussion

In our study, 122 peripheral intravenous catheter insertions were analyzed and the factors affecting the development of phlebitis were determined. In 30.3% of the patients with PIC, phlebitis developed and 48.9% of the phlebitis cases were grade 1. Our review of studies revealed that the incidence of phlebitis ranged between 1% and 77.5% (Couzigou et al., 2005; Lanbeck et.al., 2004; Palefski and Stoddard, 2001; Curran et al., 2000; Lundgren et. al., 1996). Phlebitis developed in 28.2% of 195 PIC insertions in Berse et al.'s (2020) study, in 43.2% of 110 patients in Braga et al.'s (2018) study, in 10% of 361 PIC insertions in Urbenotto and May's (2016) study, and in 31.8% of 317 PIC insertions in Atay et al.'s (2018) study. The Infusion Nurses Society states that if the incidence of phlebitis is less than 5% it is acceptable: however, the rate of phlebitis is above this limit in studies in the literature (INS, 2019). This high incidence of phlebitis probably stems from the different factors originating from the patient and the application. In several studies, most of the phlebitis cases were grade 1 (Berse et. al., 2020; Urbanetto and May, 2019; Atay et. al., 2018; Braga et. al., 2018;). Our study findings are consistent with those in the literature. Most of the phlebitis cases are grade 1 because phlebitis can be noticed early. The Infusion Nurses Society recommends that PICs should be checked at least every 8 hours.

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According to the results of our study the sex variable had no effect on phlebitis development, which was consistent with the results of other studies (Berse et. al., 2020; Uslusoy and Mete, 2008). However, in some other studies in the literature, it is reported that phlebitis is more common in women (Wallis et. al., 2014; Saini et. al., 2011). Thus, the effect of sex on the development of phlebitis is controversial.

As was indicated in several studies, the presence of a chronic disease affected the development of phlebitis (Atay et. al., 2018; Erdoğan and Denat, 2016); however, it did not affect the development of phlebitis in our study.

In the literature, it is stated that phlebitis is more common in older people, especially when it is accompanied by chronic diseases, due to the deterioration of the vascular structure and weakening of the immune system; however, in our study, the rate of phlebitis was higher in the 40-64 age group. While phlebitis was more common in individuals over 65 years of age in Berse et al.'s 82020) study, in Uslusoy and Mete's (2008) study, no relationship was determined between age and phlebitis development.

In the present study, the nurses preferred the forearm, left arm and arm that was not actively used to insert PIC, and phlebitis development was more common on the wrist and actively used arm. Contrary to our findings, in Berse et al.'s (2020) study, the site where the catheter was inserted had no effect on the development of phlebitis, but phlebitis developed more on the inactive arm. On the other hand, Atay et al. (2018) did not determine any difference between the actively and inactively used arms in terms of phlebitis development and the site where the catheter was inserted. However, in Buzatto et al.'s (2016) study, there was no difference between the right and left arms in terms of phlebitis development. As in our study, in several studies, phlebitis developed more in the actively used arm and in the joint areas because catheters inserted in these sites would be exposed to trauma more, and the use of the cubital vein and forearm veins reduced the risk of phlebitis development (Cicolini et. al., 2014; Do Rego Furtado, 2011; Uslusoy and Mete, 2008).

In our study, phlebitis development was more common in sites where the PIC was inserted in two or more attempts, or if the previous insertion was performed on the same vein. Simin et al. (2019) reported that two or more unsuccessful attempts increased the occurrence of phlebitis. In the insertion of peripheral intravenous catheter, failure of blood flow to the PIC during the insertion, failure to administer fluid, occurrence of swelling or pain in the insertion area, and the need for a new catheter are considered as the indications of unsuccessful intervention. Unsuccessful attempts suggest that either the patient's vessel walls are thin and delicate, or the nurse's knowledge and experience are inadequate (Aydın and Arslan 2018; Kuş and Büyükyılmaz, 2018). In addition, as the number of attempts made to the same vein for the insertion of PIC increases, the vein is exposed to trauma more and becomes more vulnerable to infection, and the risk of phlebitis increases as the intima layer of the vein is damaged (Webster et. al., 2015; Saini et. al., 2011).

In our study, phlebitis development was observed in all of the patients in whom 16 Fr or 18 Fr catheters were inserted. In several studies in the literature, it is reported that thick and long catheters increase the risk of phlebitis development as they cause more trauma to the vessel (Nyika et.al., 2018; Do Rego Furtado 2011). However, in some other studies, the size of the catheter is indicated to have no effect on phlebitis formation (Berse et al., 2020; Buzatto et. al., 2016; Salgueiro-Oliveira et. al., 2012). In the literature, it is recommended that the diameter of a PIC should not exceed 45% of the vessel diameter if the catheter is inserted in the vessels in the upper extremities because these vessels are thinner and delicate (Turkish Society for Hospital Infections and Control, 2019; Sharp et. al., 2015; Chopra et. al., 2014).

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Table 3. Comparison of phlebitis development according to patient characteristics and some factors related to PIC.

	Phlebitis development		**Test X^{2*}**	**p**
Variables	**No**	**Yes**		
	n (%)	n (%)		
Age				
18-39	22 (91.7)	2 (8.3)		
40-64	43 (61.4)	27 (38.6)	7.787	0.020**
65 and older	20 (71.4)	8 (28.6)		
Sex				
Female	49 (70)	21 (30)	0.008	0.927
Male	36 (69.2)	16 (30.8)		
Chronic disease				
Yes	62 (69.7)	27 (30.3)	0.000	0.997
No	23 (69.7)	10 (30.3)		
Clinic				
Internal	36 (75)	12(25)	1.063	0.303
Surgical	49 (66.2)	25 (32.8)		
Catheter insertion site				
Dorsal face of the hand	26 (89.7)	3 (10.3)		
Wrist	2 (5.9)	32 (94.1)	91.409	0.000**
Forearm	35 (94.6)	2 (5.4)		
Antecubital region	22 (100)	0 (0)		
Catheter inserted side				
Right	26 (98.7)	3 (10.3)	7.189	0.007**
Left	59 (63.4)	34 (36.6)		
Actively used arm				
Yes	0	33 (100)	103.920	0.000**
No	85 (95.5)	4 (4.5)		
Success in the first attempt				
Yes	69 (94.5)	4 (5.5)	53.111	0.000**
No	16 (32.7)	33 (67.3)		
Previous insertion in the same vein				
Yes	19 (36.5)	33 (63.5)	47.087	0.000**
No	66 (94.3)	4 (5.7)		
Catheter number (Fr.)				
16	0 (0)	2 (100)		
18	0(0)	31 (100)	104.873	0.000**
20	38 (90.5)	4 (9.5)		
22	47 (100)	0 (0)		
Antibiotic administration				
Yes	12 (26.1)	34 (73.9)	66.387	0.000**
No	73 (96.1)	3 (3.9)		

*x^2 = Chi-Square test, **$p<0.05$.

In our study, the use of antibiotics by the patients increased the development of phlebitis. Similarly, in their study, Urbanetto et al. (2016) determined that antibiotics led to the development of phlebitis, probably due to the fact that the pH of antibiotics is different from that of the blood and that they cause chemical trauma to the tunica intima.

Conclusion

In our study, the rate of phlebitis development was above the rate determined by the Infusion Nurses

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Society, most of them were grade 1 phlebitis, and the variables such as sex and presence of a chronic disease did not affect the development of phlebitis. Phlebitis development was more common in patients in the 40-64 age group, on the wrist, on the left arm and on the actively used arm, in sites where the PIC was inserted in two or more attempts, in patients in whom 16 Fr or 18 Fr catheters were inserted, and in patients taking antibiotics. To reduce the rate of phlebitis development, we recommend that reliable measurement tools should be used, that the signs and symptoms in the PIC region should be regularly and continuously observed, that nurses should regularly participate in applied trainings in order to improve their knowledge and skills, and that future studies to be carried out to determine different risk factors should include larger samples.

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Research Article / Araştırma Makalesi

The Effect of Different Body Possition on Calf Blood Pressure: A Cross-Sectional Study

Bacaktan Kan Basıncı Ölçümünde Farklı Vücut Pozisyonlarının Kan Basıncına Etkisi: Kesitsel Bir Çalışma

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Abstract: The calf area is an alternative measurement site when blood pressure cannot be measured at the arm. To determine the effects of different body positions on calf blood pressure readings in healthy young students. A total of 100 healthy young students who agreed to participate in the study were randomly selected. The study was carried out in Aydın Adnan Menderes University Nursing Faculty. In all participants, blood pressure was measured: supine, prone, right lateral, and left lateral recumbent. All measurements were performed three times at 1-min intervals, and the results were compared. There was no statistically significant difference between the blood pressure measurements in the supine and prone positions ($p>0.05$). The systolic blood pressure value was significantly lower in the left lateral recumbent position than in the other positions ($p<0.05$). The diastolic blood pressure was significantly higher in the right lateral recumbent position than in the other positions ($p<0.05$). When assessing calf blood pressure, it is important to consider the body position of the patient. It should be known that different results can be obtained in lateral recumbent positions.

Keywords: Blood pressure measurement, Calf, Vital sings.

Öz: Bacakta baldır bölgesi, kan basıncının koldan ölçülemediği durumlarda alternatif bir ölçüm yeridir. Bu çalışmada sağlıklı genç öğrencilerde farklı vücut pozisyonlarının bacaktan ölçülen kan basıncına etkisini belirlemek amaçlanmıştır. Araştırma rastgele seçilen ve araştırmaya katılmayı kabul eden toplam 100 sağlıklı genç öğrenci ile yapıldı. Çalışma Aydın Adnan Menderes Üniversitesi Hemşirelik Fakültesi'nde gerçekleştirildi. Tüm katılımcılarda kan basıncı: sırtüstü, yüzüstü, sağ yan ve sol yan yatar pozisyonda ölçüldü. Tüm ölçümler 1 dakikalık aralıklarla üç kez yapıldı ve sonuçlar karşılaştırıldı. Sırtüstü ve yüzüstü pozisyonda yapılan kan basıncı ölçümleri arasında istatistiksel olarak anlamlı fark yoktu ($p>0.05$). Sistolik kan basıncı değeri sol yan yatar pozisyonda diğer pozisyonlara göre daha düşüktü ($p<0.05$). Diyastolik kan basıncı sağ yan yatar pozisyonda diğer pozisyonlara göre daha yüksekti ($p<0.05$). Bacakta baldırdan kan basıncını değerlendirirken, hastanın vücut pozisyonunu dikkate almak önemlidir. Yan yatış pozisyonlarında farklı sonuçlar alınabileceği bilinmelidir.

Anahtar Kelimeler: Kan basıncı ölçümü, Baldır, Vital bulgular.

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Introduction

Vital signs provide essential information about the homeostatic balance and body's physiological state. Blood pressure is an important vital sign that indicates the general health status of individuals and plays a role in determining the treatment process of the patient (Cardona-Morrelli et. al., 2016). Therefore, it is necessary to measure blood pressure correctly. Although blood pressure measurement is one of the most routine examinations, it has a high probability of error. Accurate blood pressure measurements and diagnosis of hypertension are important in treating patients, regulating drug modifications, and determining cardiac risk factors (Netea et. al., 2003). Incorrect measurements cause unnecessary examinations and initiation or continuation of unnecessary drugs, which are detrimental to both the individual and the country's economy (Mok et. al., 2015).

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The upper arm is typically used for blood pressure measurements. However, an alternative measurement site is required if the arm cannot be used, especially in patients undergoing orthopaedic and plastic surgery, those recovering from a mastectomy or stroke, those with upper extremity fractures or burns, and those who have received multiple intravenous injections. Also, choosing an alternative site for measuring blood pressure is required in patients with upper limb deformities or amputations. Therefore, as an alternative to the arm, the calf region of the leg is the most commonly used area for blood pressure measurement (Sareen et. al., 2012).

Calf blood pressure can be measured in the supine position, with the leg slightly bent at the knee (Craven et.al., 2009) or in the prone position (Enç, 2015). Generally, electronic blood pressure instruments are used for these measurements, which are calibrated at regular intervals (Craven et.al., 2009).

Certain studies comparing blood pressures measured at the arm, leg, and ankle reported that the ankle might be a suitable alternative region to the arm for assessing blood pressure. (Block & Schulte, 1996; Korhonen, 2006; Lee et.al., 2020; Wilkes et.al., 2004). Some studies have revealed that the calf can also be an alternative to the arm for measuring blood pressure. The blood pressure measurements made at the ankle were, on average 4 mmHg higher than measurements made at the calf (Moore et.al., 2008).

Patients with movement restrictions in supine, prone, or lateral recumbent positions may need calf blood pressure measurements. Although the calf region may be suitable for blood pressure measurements, the patients may be positioned differently during the procedure. However, no evidence-based study shows which positions are more reliable for measuring calf blood pressure. Further studies are needed to determine which position can reliably measure calf blood pressure. The study aimed to test the effects of different body positions on calf blood pressure measurements in healthy young Turkish students.

Materials and Methods

Study Design

This is a cross-sectional study. This study was approved by the Aydın Adnan Menderes University Nursing Faculty Non-Interventional Clinical Research Ethics Committee (No: E-76261397-050.04.04-88340). The aim of the study was explained to the students, they were informed of the study procedure, and their verbal consent was obtained. This study was conducted in accordance with the Declaration of Helsinki of 1964, as revised in 2013.

Study Population

This study included 100 healthy young students randomly selected from among students from the Aydın Adnan Menderes University Faculty of Nursing between January, 2022 and June, 2022. Inclusion criteria were (1) being 18 years of age or older and (2) not having any adverse condition (open wound, burns, fracture, lymphatic drainage problems, etc.) that could affect blood pressure measurement in the legs. Patients with anatomical disorders and those who could not lie in the prone or lateral recumbent positions were excluded from the study.

Sample

The study's sample size was calculated using the software G*Power version 3.1.9.2. The study by Lee et al. (2020) was used as a reference to estimate the effect size (Lee et.al., 2020). Considering the effect size (d=0.462), 5% margin of error (α=0.05), and 80% power, the sample size was determined to be 56. A total of 100 participants (79 women and 27 men) were included in the study using a random sampling method, taking into account possible data losses.

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Study Procedures and Data Collection

A questionnaire and blood pressure record form were used to collect the research data. The questionnaire included questions about sex, age, body mass index, drug use, smoking and presence of chronic disease. The blood pressure was measured supine, prone, and lateral recumbent positions were recorded on the blood pressure record form. An electronic sphygmomanometer of the Fortune brand (for adults) was used for blood pressure measurement. The sphygmomanometer used in the research is a valid device. A medical supplier calibrated the blood pressure monitor after every 20 measures. Before its use in the study, the sphygmomanometer was tested on 10 individuals, and its standardisation was evaluated.

In the research, the questionnaires were completed through in-person interviews. During blood pressure measurement, each participant's calf was adequately exposed, and the blood pressure measurements were initiated by having them lie on a comfortable bed.

Blood Pressure Measurements

Blood pressure measurements were made 30 min after smoking, consuming caffeinated beverages (tea and coffee), eating, and exercising. Individuals rested for 10 min before the measurements were made. Sphygmomanometer cuffs compatible with the circumference of the legs and the participant's weight were utilised. The cuff was placed 2–3 cm above the popliteal artery for the measurements. All measurements were performed on the right leg. The first measurement was made with the participants in the supine position, with the head supported by a pillow at shoulder level. The second measurement was made in the prone position, with the head and shoulder supported by a pillow and the arms extended to both sides.

Other measurements were performed with the participants in the right and left lateral recumbent positions. In these positions, the individual's head was supported by a pillow that did not extend past the shoulder level. In the right lateral recumbent position, the upper leg was slightly bent at the knee, the right leg was pulled back, and a thin pillow was placed under the left leg. Similarly, while in the left lateral recumbent position, the blood pressure was measured by slightly bending the upper right leg at the knee. A 1-min break was given between each measurement; the measurements were repeated three times in each position, and the average of all three measurements was recorded as the blood pressure value.

Statistical analysis

The statistical package for the social sciences version 25.0 (SPSS Inc., IL, USA) program was used to analyse the data. The data conformity to the normal distribution was analysed using the skewness test, and it was determined that the data conformed to the normal distribution (0.348±0.241–0.183±0.478). Data were expressed as numbers, percentages, and mean ± standard deviation. Repeated measures analysis of variance was used to compare the means of measurement made at different positions. Adjustment for multiple comparisons was performed using the Bonferroni correction. Differences were considered statistically significant at $p < 0.05$, unless otherwise stated.

Results

The mean age of the participants in the study was 20.27±1.21 years (range:18–26 years); 75% were 19–21 years old, and 79% were women. While 33% of the participants were smokers, 10% had a chronic disease, and 6% used drugs continuously. The mean body mass index of the participants was 21.71±3.12 kg/m^2 (range:16.96–33.13 kg/m^2). The calf blood pressure values (systolic and diastolic) of the participants in the four positions are presented in Table 1. The difference between systolic and diastolic blood pressures in all positions was statistically significant ($p<0.05$).

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Table 1. Calf blood pressure recordings for all positions

	Supine	Prone	Right lateral recumbent	Left lateral recumbent	F/p
Mean systolic BP	122.89±13.46	122.88±12.63	125.09±16.66	118.09±13.66	*F=12.26 **<0.01**
Mean diastolic BP	64.98±6.87	64.66±8.15	68.42±8.96	63.35±7.36	*F=14.14 **<0.01**

BP; blood pressure,*: Repeated measures analysis of variance.

Table 2. Pairwise comparisons between positions in systolic and diastolic calf blood pressure.

	Supine	Prone	Right Side Lying	Left Side Lying
Sistolic BP				
Supine				*
Prone				*
Right lateral recumbent				*
Left lateral recumbent	*	*	*	
Diastolic BP				
Supine			*	
Prone			*	*
Right lateral recumbent	*	*		*
Left lateral recumbent			*	

* p<0.05 statistically significant, Bonferroni test.

The pair-wise comparisons made between the positions for diastolic and systolic blood pressure (Table 2). Systolic blood pressure measured in the left side lying position was significantly lower (p<0.05). Diastolic blood pressure was higher in the lateral recumbent position. In the left lateral recumbent; was quite low compared to the prone position (p<0.05).

The limits of agreement for systolic and diastolic blood pressure measurements according to the left and right lateral recumbent positions are presented in Table 3. Accordingly, it was determined that the greatest difference in systolic blood pressure was between the right and the left lateral recumbent positions (6.99 mmHg more). In the diastolic blood pressure measurement, it was determined that the most significant difference was again in the right and left lateral recumbent positions (5.06 mmHg lower).

Discussion

The study aimed to determined the effects of different body positions on calf blood pressure readings in 100 healthy young Turkish students. A review of relevant literature reveals studies comparing the blood pressure measurements made at the arm, wrist, and leg. However, no study has evaluated the relationship between calf blood pressure measurements and the body position. The study's results indicated a difference between blood pressure values in all the four body positions. Based on the Bonferroni test, it was determined that there was no difference between the blood pressure measured in the supine and prone positions. While systolic blood pressure was

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the lowest in the left lateral recumbent position, diastolic blood pressure was the highest in the right lateral recumbent position.

Table 3. Limits of agreement for SBP, DBP (mmHg)

	Mean difference	**% 95 limits of agreement**
Systolic BP		
Supine - Left lateral recumbent	4.79	2.73 to 6.85
Prone - Left lateral recumbent	4.81	2.51 to 7.07
Right lateral recumbent-Left lateral recumbent	6.99	4.38 to 9.60
Diastolic BP		
Supine - Right lateral recumbent	-3.43	-4.95 to -1.91
Prone - Right lateral recumbent	-3.76	-5.42 to -2.10
Left lateral recumbent-Right lateral recumbent	-5.06	-6.83 to -3.29

BP - Blood pressure SBP - Systolic blood pressure, DBP - Diastolic blood**.**

Studies have reported that the calf and ankle may be viable alternatives to the arm for blood pressure measurement (Lakhal et.al., 2011; Moore et.al., 2008). However, in these studies, all blood pressure measurements were made at the calf in the supine position. In a study comparing blood pressure measured at the arms and legs in the supine position, calf blood pressure was, on average 4 mmHg higher than arm blood pressure, while the blood pressure measurements made at the wrist were 8 mmHg higher than the arm measures. The results of this study suggest that the calf could be used as an alternative region for measuring blood pressure (Sareen et.al., 2012).

In our study, systolic blood pressure measured in the left lateral position was lower than in all positions. There were studies reporting results similar to our study (Almeida et.al., 2009; Armstrong et.al., 2011; Ribeiro et.al., 2016; Tran et.al., 2014). According to another study, the blood pressure determined in the right lateral recumbent position was 16 mmHg lower than other positions (Bein et.al., 1996).

The reasons for these differences in blood pressure measurements and hydrostatic effects have been reported. Pressure in any vessel below the heart level tends to be high, whereas pressure in any vessel above the heart level tends to be low due to the influence of gravity (Park and Park, 2002). However, because all measurements were performed in the recumbent position in our study, there was no significant difference between leg and heart alignment. In our study, the difference between the lateral recumbent and other positions was much smaller than in other studies. These differences can be attributed to individual characteristics unrelated to the effects of the recumbent body position or the position of the leg. However, because all measurements in our study were performed in the recumbent position In addition, this difference may suggest that differences in body position do not result in significant haemodynamic changes. In addition, these results may have been obtained in blood pressure measurements due to individual variances in the calf muscle structure.

As a limitations, the study was conducted at a single centre and the number of patients was small.

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The study was performed in healthy young students and cannot necessarily be extrapolated to other populations. Calf blood pressure in the lying position was evaluated, and measurements were repeated in different positions. Blood pressure was measured only in the right leg.

In conclusion; the study's results indicate that different body positions affect the measurement of blood pressure in healthy young students. This study confirms the literature-suggested practise of taking blood pressure from the leg in the supine or prone posture. It was determined that the systolic blood pressure value was lower in the left lateral recumbent position. Also, the diastolic blood pressure was higher in the right lateral recumbent position and lower in the left lateral recumbent position than in the prone position. According to the results of the study, it is recommended that the blood pressure measurements of the patients be made in the prone or supine position if possible, and if it must be measured in the lateral recumbent position. It is recommended to evaluate the blood pressure values of the patient by being aware of these differences in the measurement results. In addition, the body position in which the blood pressure measurements were performed should be documented.

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Research Article / Araştırma Makalesi

Dietary Fiber Analysis of D-Allulose Added Cakes and Determination of Microbiological Changes During Storage

D-Alluloz İlaveli Keklerin Diyet Lifi Analizi ve Depolama Süresince Mikrobiyolojik Değişikliklerin Belirlenmesi

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Abstract: D-Allulose is a monosaccharide that is rarely found in foods and can be obtained commercially by enzymatic reactions and isolation from bacteria. It can not be absorbed because of its chemical composition's resistance to digestive enzymes. It is referred to as a "energy-free natural sweetener" for this reason. These findings has led to an increase in D-allulose usage in the food industry. In this study, the total dietary fiber of cakes with additional D-allulose was determined, as well as the microbiological changes that occurred during storage. Three distinct cakes were made for the trial as the control (sugar), 50% sugar plus 50% D-allulose, and 100% D-allulose. In the baked cakes, dietary fiber analysis was done on day 1 and microbiological analyses were done on days 0, 7, 14, and 21. On the first day of analysis, there was no difference between the groups in the total spor-forming bacteria counts of the cake groups, but on the 7th, 14th, and 21st days of analysis, there were statistically significant differences between the groups ($p<0.05$). There was no statistically significant difference between the AL100 group and the control and AL50 groups when the total thermophilic bacterial counts of the cake samples were compared according to the storage durations ($p>0.05$). The control group's total dietary fiber content were 5.93 g/100 g, and AL 100 group's content were 10.70 g/100 g. D-Allulose is regarded as a natural sweetener with significant antimicrobial properties and a high dietary fiber content.

Keywords: Cake, D-Allulose, Dietary fiber, Anti-microbial effect.

Öz: D-alluloz besinlerde nadir olarak bulunan ve ticari olarak enzimatik reaksiyonlar ve bakterilerden izolasyon ile elde edilebilen bir monosakkarittir. Kimyasal yapısına bağlı olarak sindirim enzimlerine direnç göstermekte ve emilememektedir. Bu nedenle enerji içermeyen doğal tatlandırıcı olarak isimlendirilmektedir. Bu etkisi nedeniyle D-allulozun besin endüstrisinde kullanımı giderek artmaktadır. Bu çalışmanın amacı D-alluloz ilave edilen keklerin toplam diyet lifinin belirlenmesi ve depolama süresince mikrobiyolojik değişikliklerin belirlenmesidir. Çalışmada kontrol (şeker), %50 şeker +%50 D-alluloz ve %100 D-alluloz olmak üzere 3 farklı kek üretilmiştir. Üretilen keklerde 1. gün diyet lifi analizi, 0, 7, 14 ve 21. günlerde mikrobiyolojik analizleri yapılmıştır. Kek gruplarının toplam sporlu bakteri sayımlarında analizin ilk gününde gruplar arası fark bulunmazken 7, 14 ve 21. gün analizlerinde gruplar arasında istatistiksel olarak anlamlı farklılıkar saptanmıştır ($p<0,05$). Kek örneklerinin depolama sürelerine göre toplam termofilik bakteri sayımlarına bakıldığında ise AL100 grubunda istatisitksel olarak anlamlı bir farklılık saptanmazken ($p>0,05$), kontrol ve AL50 grubunda istatistiksel olarak anlamlı farklılıklar bulunmaktadır. Kontrol grubunun toplam diyet lifi 5,93 g/100 g ve AL 100 grubunda 10,70 g/100 g'dır. D-Allulozun yüksek diyet lifi içeren ve antibakteriyel etkileri olan doğal bir tatlandırıcı olduğu düşünülmektedir.

Anahtar Kelimeler: Kek, D-Alluloz, Diyet lifi, Anti-mikrobiyal etki.

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Introduction

Cakes are baked products with a sweet flavor and a dense, soft texture that are appreciated by people all over the world. The primary factors that determine of the cakes' quality are the ingredients, wheat flour, eggs, sugar, fat or oil, and leavening agents, used in the recipe as well as the methods of combining and baking used to prepared (Dizlek, 2013). Sucrose (sugar) has a wide range of roles in baked goods, including those of sweeteners, softening and moisturizing, and browning agents. It is an essential ingredient that contributes to taste, flavor, and texture. Nevertheless, it has been demonstrated that reducing sugar in bakery products lowers the gelatinization temperature of starch and encourages the development of gluten proteins, which has a negative impact on the quality of cakes and inhibits the browning response (Kweon et al., 2009). Because sugar provides an important function in baked goods, sucrose substitutes are difficult to achieve identical attributes from. The qualitative properties of sugar alcohols or polyols, which are frequently used as sucrose substitutes in bread items, have been studied in a variety of ways (Mariotti et al., 2012).

Since sucrose is the primary sweetener used on a worldwide basis, it is an essential nutrients in the food industry. However, because to its high glycemic index, people with diabetes should be careful to consume sucrose in moderate (less than 10% of daily energy intake) as excessive consumption might result in dental caries and diet-related diseases (WHO, 2015). Cakes are the baked good that has the most sugar among of all the rest of the baked goods. Therefore, it's crucial for health that this cake's sugar content be kept to a minimum or replaced with a low-calorie component.

The hydroxyl group position of C-3 is transformed in the conversion of D-fructose to D-allulose, commonly known as D-psicose, by the activity of the ketose 3-epimease enzyme (Izumori, 2006). D-Allulose has increased in value as a result of its high level of food safety, utility, and physicochemical similarities to sucrose. The US Food and Drug Administration recently classified D-allulose as "generally safe" and it is currently in the "relatively harmless" category, which denotes the lowest toxicity (Mu et al., 2012). Although D-allulose has a sweetness that is 70% as sweet as sucrose, it is categorized as an ultra-low-calorie sugar since it only offers 0.2 kcal/g of usable energy, or 5% of sucrose's energy. Also, D-allulose, unlike sucrose and D-fructose, has almost no effect on blood sugar levels due to its negligible glycemic index (Lamorte et al., 2019). Therefore, D-allulose is suitable for consumption by diabetic patients, making it a potentially ideal substitute for sucrose and is a nutritionally better option compared to other free sugars (such as D-fructose) (Belitz et al., 2004).

It is aimed to determine the effect of D-allulose, which will be added to the structure of the cake, which is a bakery product we will produce within the scope of the project, on the microbiological quality of the product, as well as the effect of D-allulose, which will be added to the structure of the cake, which is a bakery product that we will produce within the scope of the project, dietary fiber amount of the product. For these purposes, it is aimed to produce functional cakes from D-allulose added at different rates and to determine the shelf life of the cakes produced by microbiological analyzes to be made on different days.

Materials and Methods

Materials

D-Allulose was obtained from a D-allulose manufacturer. Wheat flour, sugar, eggs, milk, salt, margarine, vanilla and baking powder used in cake production were purchased from local markets in Burdur, Türkiye.

Preparation of Experimental Cakes

The ratios of D-allulose to be added to the cakes were determined on the basis of studies using D-allulose as a sugar substitute. 100% sucrose, 0% D-

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allulose (Control); Three types of cakes were produced: 50% sucrose, 50% D-allulose (AL50) and 0% sucrose, 100% D-allulose (AL100) (Table 1).

Table 1. Cake recipe to be used in the study

Components	Control	AL50	AL100
Wheat flour (g)	100	100	100
Sugar (g)	80/0	40/40	0/80
Egg (g)	60	60	60
Milk (mL)	65	65	65
Salt (g)	0.5	0.5	0.5
Baking powder (g)	4	4	4
Margarine (g)	60	60	60
Vanilla (g)	9	9	9

Appropriate baking temperature and determined in cake recipes, all components were kept at room temperature for 45 minutes before starting to work. In the study, firstly, wheat flour, baking powder and salt were mixed. In a separate bowl, it was whipped for 5 minutes at the 5th revolution with the help of an electric mixer until the sugar and oil were well mixed, and vanilla was added to it and whisking was continued for more minute. Eggs were added to this mixture and pre-mixed wheat flour, baking powder, salt mixture and milk were added to ensure that they were mixed well.

The doughs obtained were baked in the oven (Arçelik / 9620 Mi), which was previously set at 175°C. At the end of the baking time, the cakes were removed from the oven and allowed to cool for 1 hour at room temperature. They were then packaged until analyzed and stored at +4°C until the 21st day. Microbiological analyzes were performed on days 0, 7, 14 and 21 of production, total dietary fiber and soluble and insoluble dietary fiber analysis on first day.

Microbiological analyzes

Total spor bacteria count: Plate Count Agar (PCA) was taken from a dilution of 1 mL and inoculated with the spread plate method, and after the petri were incubated for 48 hours at 28±1ºC, total spor bacteria counts were made.

Total thermophilic bacteria count: Plate Count Agar was taken from a dilution of 1 mL and inoculated with the spread plate method, and the total thermophilic bacteria formed after the petri were incubated at 45±1ºC for 48-72 hours were counted.

Coliform bacteria count: Violet Red Bile Agar was taken from a dilution of 1 mL and inoculated with the double layer pouring method, and the typical colonies formed after incubating the petri at 30±1ºC for 24 hours were counted.

Yeast and Molds count: 1 mL was taken from the dilutions and inoculated on Potato Dextrose Agar (PDA) medium by the cast plate method, and the colonies were counted by incubating the petri for 5 days at 22±1ºC.

Determination of the total amount of dietary fiber: The total, soluble and insoluble dietary fiber amounts of the cakes were determined according to the AOAC 991.43 method as a service purchase in the TÜBİTAK Marmara Research Center Food Laboratory.

Statistical analyzes

Data will be analyzed using SPSS (version 26) package software. One Way ANOVA test was applied to compare the differences between more than two groups. Tukey Post Hoc multiple comparison test was used to determine the difference between groups. $p<0.05$ was considered statistically significant. In order to determine the difference between days, ANOVA analysis was used in repeated measures, which is the equivalent of One Way ANOVA in dependent groups.

Results

In the study, 3 different cakes were produced using standard (control [100% sugar]), AL50 (50% D-allulose) and AL100 (100% D-allulose) and

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immediately after production and in the refrigerator (at +4°C) 7, 14 and after 21 days of storage, total spor-forming bacteria, total thermophilic bacteria (TTB), coliform group bacteria and yeast-mold counts are shown in Table 2. When the total spor-forming bacteria counts of the cake groups were examined, there was no difference between the groups on the first day of the analysis, but statistically significant differences were found between the groups in the analyzes of the 7th, 14th and 21st days ($p<0.05$). It was determined that this difference was due to the count difference between all groups on the 7th day, lower in the AL50 group on the 14th day, and higher bacterial count in the control group on the 21st day.

Table 2. Microbiological analyzes of cake samples

Parametres	Days	Cake groups (Mean±SD)		
		Control	AL50	AL100
Total Spor-Forming Bacteria	0	$5,77\pm0,1^{aA}$	$5,63\pm0,4^{aA}$	$5,81\pm0,2^{aA}$
	7	$6,8\pm0,7^{aB}$	$5,4\pm0,7^{bA}$	$5,77\pm0,7^{cA}$
	14	$6,48\pm0,1^{bB}$	$5,63\pm0,0^{aA}$	$6,63\pm0,4^{bB}$
	21	$7,51\pm0,2^{bC}$	$6,37\pm0,1^{aA}$	$6,30\pm0,2^{aB}$
Toplam Thermophilic Bacteria	0	$7,51\pm0,4^{aA}$	$6,63\pm0,3^{aA}$	$7,51\pm0,2^{aA}$
	7	$7,93\pm0,7^{aB}$	$6,88\pm0,7^{bA}$	$7,63\pm0,7^{cA}$
	14	$8,44\pm0,3^{aB}$	$8,57\pm0,2^{aB}$	$7,48\pm0,4^{aA}$
	21	$8,80\pm0,1^{bC}$	$8,92\pm0,0^{bB}$	$7,83\pm0,0^{aA}$
Coliform Bacteria	0	-	-	-
	7	-	-	-
	14	-	-	-
	21	-	-	-
Yeast-Mold	0	-	-	-
	7	-	-	-
	14	-	-	-
	21	-	-	-

*Statistical differences between groups are shown in lower case letters. Statistical differences between days are shown in capital letters.

When the bacterial counts depending on the shelf life of the cake groups were examined, it was determined that there was a statistical difference depending on the day in the control group and this difference was more pronounced on the 21st day. While there was no statistical significance with storage in the AL50 group, statistically significant differences were found in the AL100 group on the 14th and 21st days. While there was no statistically significant difference in total thermophilic bacteria counts on days 0 and 14 of the analysis ($p>0.05$), significant differences were found on days 7 and 21 ($p<0.05$). On the 7th day, it was determined that the reason for this difference was higher bacterial counts in the control group, and on the 21st day, it was due to the lower bacterial counts in the AL100 group compared to the AL50 and control group. When the total thermophilic bacteria counts of the cake samples were examined according to the storage times, there was no statistically significant difference in the AL100 group ($p>0.05$), while there were statistically significant differences in the control and AL50 groups. While this difference was more pronounced on the 21st day of storage

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in the control group, it was found to be associated with an increased bacterial count from the 14th day in the AL50 group. During the preservation of the cake groups, coliform bacteria and yeast-molds were not found in all groups. Total soluble and insoluble dietary fiber of the control group was 5.93 g/100 g, soluble dietary fiber was <0.65 g/100 g, and insoluble dietary fiber was 5.93 g/100 g. In the AL 100 group, these values were determined as 10.70 g/100 g, <0.65 g/100 g and 10.70 g/100 g, respectively (Table 3). Total soluble and insoluble dietary fiber amounts of AL 50 group were not determined.

Table 3. Total dietary fiber values of cake samples

	Total dietary fiber	Soluble dietary fiber	Insoluble dietary fiber
Control	5.93 g/100 g	<0.65 g/100 g	5.93 g/100 g
AL100	10.70 g/100 g	<0.65 g/100 g	10.70 g/100 g

Discussion

Microbiological Analysis

Typically, the primary factor limiting the shelf life of baked prodcuts is microbiological deterioration. Microbial growth disruption results in economic loss for both producers and consumers. Numerous specific factors, including packaging, hygienic production procedures, storage conditions, and product turnover, might contribute to these losses. Yeast is a concern, particularly in baked goods. Osmophilic yeast contamination of items is typically brought on by dirty tools and equipment. As a result, osmophilic yeast contamination is reduced to a minimum when excellent production processes are followed (Goranova et al., 2020). Mold formation in bakery products is a serious problem that causes economic losses. Mold spores are generally destroyed by baking in fresh bread and other baked goods (Malkki and Rauha, 2000). Therefore, for the product to mold, it must be contaminated from the air, oven surfaces, equipment, food processors or raw materials during post-cooking cooling, slicing or packaging operations. This means that all spoilage problems caused by molds must occur after cooking (Knight and Menlove, 2006). In warmer weather and more humid storage conditions, the number of mold spores is higher in summer than in winter, due to airborne contamination. In addition, moisture condensation on the surface of a product due to packaging before it is completely cooled can cause mold growth (Saranraj and Geetha, 2012).

Lactic acid bacteria can prevent yeast-mold formation in bakery products due to their antifungal activities. It is also known that many members of lactic acid bacteria produce bacteriocins. The antibacterial effect was defined by antibiotic and antibiotic-like substances such as acidophilin and lactocidin produced by *Lactobacillus acidophilus*, lactolin produced by *Lactobacillus plantarum* or *nisin* produced by *Lactococcus lactis*. Depending on the type of bacteriocin, they can inhibit food-borne pathogenic bacteria such as *Staphylococcus aureus*, *Listeria* spp., *Bacillus cereus*, *Clostridium perfringens* (Dinçer et al., 2010). It is known that D-allulose promotes the growth of lactic acid bacteria in foods (Kimoto-Nira et al., 2017). Therefore, in this study, it is thought that the possible reason why the total mesophilic and total thermophilic bacteria counts related to shelf life were lower in the cake groups containing D-allulose compared to the control group. Alshehry (2019) investigated the anti-microbial effects of cupcakes with natural antioxidant supplementation during storage (days 0, 7, 15 and 21) and compared the total bacteria and yeast-mildew supplement in beet cupcakes during storage compared to control cupcakes made from 72% extracted wheat flour. showed

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that the number was inhibited. D-allulose is thought to be a natural sweetener that increases antioxidant capacity and inhibits yeast and mold formation in the product it is used in. In the microbiological analysis of Goranova et al., (2020) sponge cake cakes, coliform group, yeast-mold and total mesophilic aerobic group were not found on the first day of production. Gill et al. (2020) investigated microbiological analyzes in bakery products and found the total number of mesophilic aerobic bacteria in cake samples to be $5.7/10^4$ cfu/L. Coliform bacteria and yeast-mold were not detected. In another study, it was determined that the total aerobic bacteria counts in cakes produced from buckwheat flour were $2.9*10^2$, $4.7*10^3$ and $9.4*10^5$ cfu/g at 0, 7 and 10 days, respectively. In addition, total coliform bacteria and yeast-mold results were shown to be within normal values (Farzana et al., 2021). The effect of shelf life was not investigated in this study. According to the Turkish Food Codex Microbiological Criteria Regulation (2013) of the Ministry of Agriculture and Forestry, plain cakes, plain biscuits, plain crackers, etc. The upper limit for coliform bacteria in coated, filled and/or flavored biscuits, cakes and crackers and wafers is 10^2 cfu. It is seen that the results of coliform bacteria obtained from this project are suitable for Turkish Food Codex Microbiological criteria.

Determination of the total amount of dietary fiber

Dietary fibers are defined as the edible parts of plants that cannot be digested in the small intestine, but are fully or partially digested in the large intestine, according to the AACC International (International Society of American Cereal Chemists) (Anonymous, 2001). Soluble and insoluble dietary fibers in the diet are important in terms of many nutrition-related errors such as obesity, diabetes, hypertension, cardiovascular diseases, especially the gastrointestinal system, and should be included in the diet in sufficient quantities. It is reported that healthy adult individuals should take at least 25-35 g/day or 10-13 g/1000 kcal dietary fiber per day to prevent diseases. Legumes, whole grain foods, oilseeds, vegetables and fruits are rich sources of dietary fiber. In addition, foods with increased functionality are also used as a source of dietary fiber. In this study, in which we developed 3 different cake samples using D-Allulose and sugar, the dietary fiber values of the groups are shown in Table 3. When the experimental cake groups consumed 100 g (approximately 2 portion), the control group cakes met 23.7% of the total daily recommended dietary fiber (25-35 g/day), while the cakes produced with 100% D-allulose provided 42.8% of the total dietary fiber. meets. No studies have been found in the literature showing the effect of D-allulose on dietary fiber in bakery products. Rugji et al. (2022) reported that D-allulose has a prebiotic effect in whey beverages. We demonstrated the prebiotic effects of D-allulose in a previous traditional review (Özgür et al., 2022). The contribution of D-allulose to the total dietary fiber in the diet has been clearly shown in this study. This project is the first study to show the effect of D-alllulose on dietary fiber. In the literature, there are studies showing the effects of different components on the physico-chemical properties of cakes. In a study by Goswami et al. (2015) on muffin muffins, it was determined that muffin muffins with millet added more dietary fiber compared to the control group. In a study in which the total dietary fiber of the cakes with blueberry was determined, it was reported that the cakes produced with blueberry added at different rates (8%, 16% and 24%) could meet 13.3%, 17.1 or 19.6%, respectively (Işık and Urgancı, 2017). In a study in which dietary fiber was determined in cakes produced using different fiber sources, it was reported that the highest dietary fiber was found in the sample containing 10% palm kernel flour (Hamzaçebi, 2017). Ataman and Gül (2020) investigated the effect of cracked chickpea flour on muffin quality in a study where the total dietary fiber content, which was determined as 4.65% in the control sample, was 5.05%, 5%, respectively in muffin cakes with 10%, 20% and 30% cracked chickpea flour added. 65 and 6.43%. In line with these results, it is thought that the cakes produced from D-allulose have the

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potential to be a healthy snack especially for obese and diabetic individuals with their high dietary fiber contribution that does not contain sugar.

As a result, it is believed that cakes with D-allulose contained and controls are produced in hygienic conditions and protected against microbial deteriorating while in storage. Depending on the shelf life, D-allulose can inhibit both total thermophilic and total mesophilic bacteria. D-allulose is therefore expected to also have antibacterial properties. D-Allulose contributes to dietary fiber because digestive enzymes do not alter most of it before it reaches the large intestine. Because of its prebiotic impact, it is believed that it can be an important alternative sweetener for diseases related to nutrition. Future research should support these effects with human studies. Additionaly, even though cakes with D-allulose are a healthy snack with low energy, high dietary fiber and antibacterial effects, they should be consumed by paying attention to portion control in both healthy adults and individuals with diabetes.

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Research Article / Araştırma Makalesi

Molecular Prevalence of Canine Leishmaniasis in Burdur, Türkiye

Burudur'da Kanin Leishmaniazisin Moleküler Prevalansı

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Abstract: Canine leishmaniasis (CanL) is among the most important vector-borne parasitic diseases in dogs worldwide, except for Oceania, and can be fatal if left untreated. The present study aimed to investigate the molecular prevalence of *L. infantum*, the causative agent of CanL, in Burdur province of Türkiye. Blood samples collected from 120 dogs were first examined for *Leishmania* spp. by genus-specific PCR and then positives were examined for *L. infantum/donovani* complex by species-specific PCR. As a result, *Leishmania* DNA was detected in five out of the 120 samples in the first-stage PCR, resulting in a molecular positivity rate of 4.16% at the genus level. Among these five samples, four were positive for *L. infantum* in the second-stage PCR, leading to a prevalence of 3.33% for *L. infantum*. No statistically significant differences were found in terms of gender and age concerning *L. infantum* positivity. Additionally, blood smears were examined under a microscope, but no *Leishmania* amastigotes were observed in any of the samples. With this obtained data, the presence of *Leishmania* spp. and *L. infantum* in dogs in Burdur is reported for the first time using a molecular method. The importance of vector fly control has been remembered once again in order to protect from the disease both animals that can be host of parasites, especially dogs, and human society, since it is a zoonotic disease.

Keywords: Burdur, Canine leishmaniasis, *Leishmania infartum*, PCR, Prevalence.

Öz: Kanin leishmaniasis (CanL); Okyanusya haricindeki tüm kıtalarda görülen ve tedavi edilmediği durumlarda ölümcül olabilen, köpeklerin en önemli vektör kaynaklı paraziter hastalıkları arasında yer almaktadır. Bu çalışma ile Burdur ilinde kanin leishmaniasis etkeni olan *L. infantum*'un moleküler yaygınlığının araştırılması amaçlanmıştır. Bu amaçla 120 köpekten toplanan kan örnekleri önce cins düzeyinde *Leishmania* spp. sonra da tür düzeyinde *L. infantum* primerleri kullanılarak Polimeraz Zincir Reaksiyonu (PCR) yöntemi ile incelenmiştir. Sonuç olarak birinci aşama PCR ile incelenen 120 köpek kan örneğinin beş tanesinde *Leishmania* DNA'sı saptanmış, böylece *Leishmania* cins düzeyinde moleküler pozitiflik oranı %4,16 olarak bulunmuştur. Cins düzeyinde pozitiflik saptanan bu beş örneğin *L. infantum* için yapılan ikinci basamak PCR sonucunda ise dört örnek pozitif sonuç vermiş ve *L. infantum* prevalansı %3,33 şeklinde bulunmuştur. Cinsiyet, yaş ve ırk ile *L. infantum* pozitifliği arasında istatistiksel olarak anlamlı bir farklılık bulunmamıştır. Aynı zamanda kan frotileri de mikroskobik bakı ile incelenmiş ancak örneklerin hiçbirinde *Leishmania* amastigotlarına rastlanmamıştır. Elde edilen bu veriler ile Burdur'da ilk defa moleküler tabanlı bir yöntem olan PCR ile köpeklerde *Leishmania* spp. ve *L. infantum* pozitifliği bildirilmektedir. Hastalıktan hem köpekler başta olmak üzere parazite konaklık yapabilen hayvanları hem de zoonoz bir hastalık olması nedeniyle insan toplumunu koruyabilmek için vektör sinek mücadelesinin önemini bir kez daha hatırlanmıştır.

Anahtar Kelimeler: Burdur, Kanin leishmaniazis, *Leishmania infartum*, PCR, Prevalans.

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Introduction

Leishmaniasis is a vector-borne, zoonotic protozoan disease mostly seen in tropical and subtropical climates (Lamotte et al., 2017; Steverding, 2017). *Leishmania* species, the causative agents of the disease, are flagellated protozoans in the Trypanosomatidae family; use some carnivores, rodents, lizards, insectivores, especially humans and dogs as definitive, *Phlebotomus* (in

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Europe, Asia and Africa) and *Lutzomyia* sandflies (in America) as intermediate hosts and transmitted by infected female sandflies sucking blood (Ayele and Seyoum, 2016; Alemayehu and Alemayehu, 2017). *Leishmania* species are heteroxene parasites with a life cycle between sandflies where they multiply as free promastigotes in the intestinal lumen and mammalian hosts where they multiply as obligate intracellular amastigotes in mononuclear phagocytic cells (Handman, 1999; Lamotte et al., 2017). The disease may be observed in different clinical forms of varying characters and severities, based on the parasitized species and immune response against the infection. According to this, the disease may have various names such as; visceral leishmaniasis (VL or Kala Azar), cutaneous leishmaniasis (CL), mucocutaneous leishmaniasis (MCL) and diffuse cutaneous leishmaniasis (DCL) (Akhoundi et al., 2016; Steverding, 2017; Galluzzi et al., 2018).

Dogs are in a critical position for public health, because of they are reservoir hosts of zoonotic *Leishmania* species, in addition canine leishmaniasis (CanL) is among the most important vector-borne parasitic diseases of all ages and breeds of dogs in both cutaneus and visceral forms (Slappendel and Greene, 1990; Ribeiro et al., 2018). Leishmaniasis may occur in all continents except Oceania and can be fatal when untreated (Baneth et al., 2008; Dantas-Torres et al., 2012; Travi et al., 2018; Olias-Molero et al., 2019). The primary responsible pathogen of canine leishmaniasis is *L. infantum* in the *L. donovani* group, in almost every region of the world, especially in Mediterranean countries such as Turkey, Italy, Spain and Portugal (Ready, 2014; Ribeiro et al., 2018; Teimouri et al., 2018; Olias-Molero et al., 2019). In addition, canine leishmaniasis cases have been reported caused by *L. tropica* and *L. major* (Bamorovat et al., 2015; Baneth et al., 2014; 2017; Hakkour et al., 2019). In the Americas (especially Central and South parts), the most important agent responsible for canine leishmaniasis is *L. chagasi*, which is synonymous (identical) of *L. infantum* (Akhoundi et al., 2016; Marcondes and Day, 2019).

Various prevalence studies have been carried out for leishmaniasis in Turkey and the most commonly reported species were *L. tropica* and *L. infantum* (Özbel et al., 2000; Ertabaklar et al., 2005; İça et al., 2008; Özensoy Töz et al., 2009; Aydenizöz et al., 2010; Bakırcı et al., 2016; Ünlü et al., 2019). There are also reports for *L. major* and *L. donovani* (Koltaş et al., 2014; Zeyrek et al., 2014). According to the results of the previous studies; the prevalence of canine leishmaniasis (regardless of diagnostic method) were reported between 37.4% from different geographical regions of Turkey.

Studies on obtaining epidemiological data, especially prevalence of pathogens, have priority for the design and implementation of effective prevention and control strategies against diseases. Therefore, the aim of the present study was to investigate the prevalence of *L. infantum*, mainly causative agent of CanL, in dog blood samples in Burdur province by using Polymerase Chain Reaction (PCR).

Materials and Methods

Ethical Approval

The present study was approved by the Burdur Mehmet Akif Ersoy University Animal Experiments Local Ethics Committee (date: 17.02.2021 and decision number: 2021/722).

Blood samples and Deoxyribonucleic acid (DNA) extraction

The blood specimens were obtained from dogs visiting Animal Hospital of Burdur Mehmet Akif Ersoy University Faculty of Veterinary Medicine and private veterinary clinics with various complaints and municipal animal shelter in Burdur city. A total of 120 dogs with different age, sex and breed were sampled and 5-6 ml of blood specimens were taken from cephalic vein (*vena cephalica antebrachii*) into EDTA-containing blood tubes. All samples were collected between June and October 2021. DNA extraction was performed from 300 µL of each blood sample via

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Promega Wizard Genomic DNA Extraction Kit (Madison, WI, USA) by manufactural instructions.

Microscopical examination

In case of detecting amastigote forms of the parasite, blood smears were prepared for each sample, fixed in methanol, stained with 5% Giemsa solution and then examined under 100x objective of light microscope.

Polymerase Chain Reaction (PCR)

Examination of the samples for *Leishmania* sp. DNA was performed using a two-stage PCR protocol. In the first step; RV1 (5'- CTTTTCTGGTCCCGCGGGTAGG - 3') and RV2 (5'- CCACCTGGCCTATTTTACAC - 3') primer pairs were used to amplify the 145 base pair (bp) region of kinetoplast DNA (kDNA) minicircles of the *Leishmania* genus, which includes *L. donovani*, *L. infantum*, *L. tropica* *L. major* and *L. braziliensis* (le Fichoux et al., 1999; Gao et al., 2015). In the second step, *Leishmania* spp. positive isolates were examined using MC1 (5' - GTTAGC CGATGGTGGTCTTG - 3') and MC2 (5' - CACCCATTTTTCCGATTTTG - 3') primer pairs amplifying a 447 bp region of *L. infantum/donovani* complex kDNA minicircles (Cortes et al., 2004). Total final volume for each PCR mix was 50 µL. PCR mixes consisted of; 1 × PCR buffer, 1.5 mM MgCl2, 200 mM of each dNTP, 2.5 U of hotstart Taq DNA polymerase, 25 pmol of forward and reverse primers and 2 µL of template DNA. Reaction conditions were as follows: an initial denaturation at 94˚C for 10 min was followed by 45 cycles of denaturation (95˚C for1 min), annealing (62˚C for 1.5 min for RV1/RV2 primers, 50˚C for 1.5 min for MC1/MC2 primers) and extension (72˚C for 30 sec), then followed by 72˚C for 10 min as a final extension. Following PCR amplification, 9 µL of each sample were electrophoresed in a 1.5% agarose gel prepared with 1 x TAE (Tris-asetik asit-EDTA) buffer at 100 Volt about 45 minutes and visualized by using ultraviolet (UV) light.

Statistical analysis

The data obtained from the present study was analysed using Minitab 16 Statistical Software. The Chi-Square test was performed to analyse the association among age, sex, breed parameters and PCR positivity. Differences for which the P value was less than 0.05 were considered statistically significant during comparisons for each parameter within itself.

Results

Due to microscopic examination, none of the samples were found positive in terms of amastigote forms of *Leishmania* spp. even positive ones in PCR, however, *Ehrlichia* spp. gamonts were detected in one sample (Figure 1).

In the first step PCR, *Leishmania* spp. DNA was detected in five of the 120 dog blood samples, therefore the molecular positivity rate of *Leishmania* spp. was was found 4.16%. As a result of the second step PCR performed for *L. infantum*, four samples were positive, therefore, the prevalence of *L. infantum* was found to be 3.33% (Table 1). The agarose gel electrophoresis images of the PCR results for both primer pairs are shown in Figure 2.

According to data analysis, no statistical significance was determined in the *L. infantum* positivity rates among age groups (Table 2) and sex (Table 3).

Considering the relationship among breeds and *L. infantum* positivity, crossbred and kangal breed dogs were found positive as 8.57% (3/35) and 12.5% (1/8), respectively, while positivity was not found in other 22 different breeds. Besides, P value could not be obtained by chi-square test, since the number of dogs found positive by breed was insufficient for statistical analysis.

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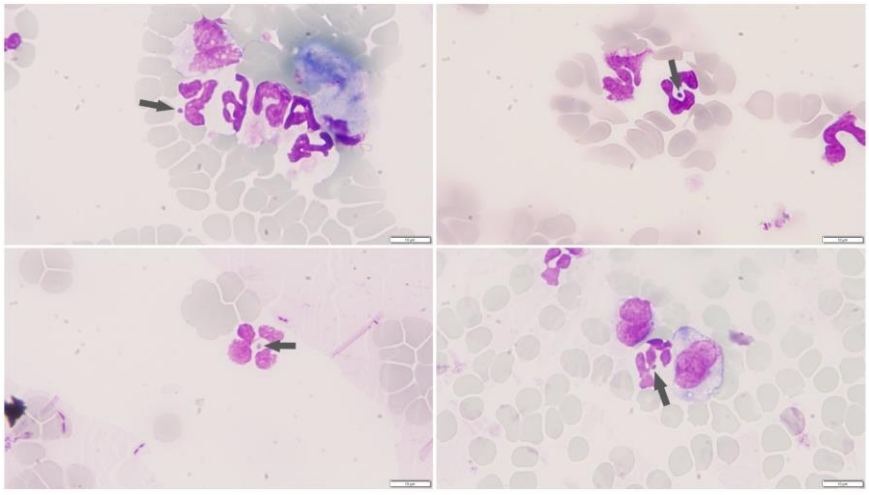

Figure 1. *Ehrlichia* spp. gamonts (arrows), 100x objective, scalebar: 10 µm

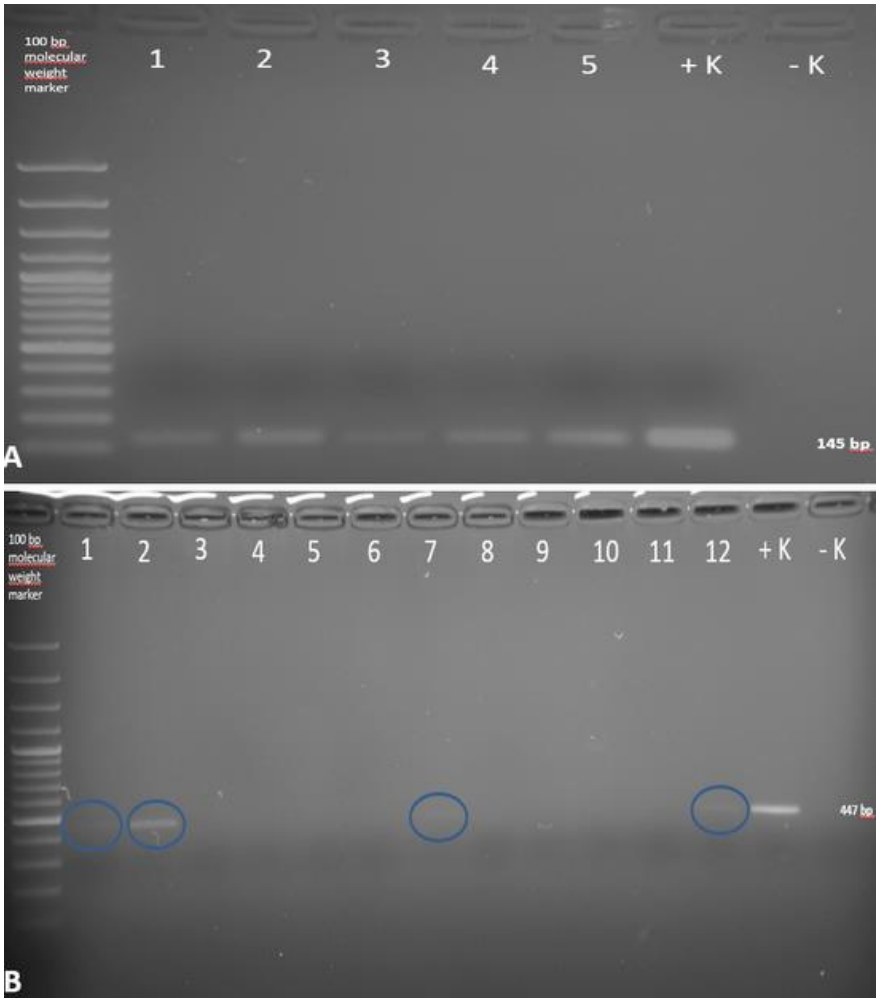

Figure 2. Agarose gel electrophoresis images of amplified DNA using; RV1-RV2 (A) and MC1-MC2 (B) primer pairs. A: 1-5 positive samples, +K positive control, -K negative control. B: 1,2,7,12 positive samples (1, 7 and 12 were very faint), +K positive control, -K negative control.

Table 1. Number and percentages of *Leishmania* spp. and *L. infantum* positive dogs

Number of examined dog	Number and percentage of *Leishmania* spp. positive dogs	Number and percentage of *L. infantum* positive dogs
120	5 (4.16%)	4 (3.33%)

Table 2. Distribution of *L. infantum* positive dogs according to age groups

Age groups (year)	Number of examined dogs	Number and percentage of *L. infantum* positive dogs
0-3	57	2 (3.5%)
3-6	29	1 (3.44%)
6-12	27	1 (3.7%)
>12	7	0 (0%)
P		> 0.05

Table 3. Distribution of *L. infantum* positive dogs according to sex

Sex	Number of examined dogs	Number and percentage of *L. infantum* positive dogs
Female	63	3 (4.76%)
Male	57	1 (1.75%)
P		0.359

Discussion

According to literature review; microscopic examination, in vitro cultivation, serological and molecular based methods are commonly used for studies on the epidemiology of canine leishmaniasis. Sensitivity of traditional methods such as microscopic examination are low especially in reservoir hosts and are insufficient in distinguishing different *Leishmania* species (İça et al., 2008; Özerdem et al., 2009; Aydenizöz et al., 2010; Töz et al., 2013). Various serological methods, such as Indirect Fluorescent Antibody Test (IFAT), Direct Antiglobulin Test (DAT), Enzyme Linked Immunosorbent Assay (ELISA), C-ELISA and Dot-ELISA have been used to determine the seroprevalence of canine leishmaniasis (Schalling and Oskam, 2002; İça et al., 2008; Özerdem et al., 2009). However, these methods may not always detect current infections, and the commonly used IFAT method can produce cross-reactivity between different species (Solano-Gallego et al., 2009; Bourdeau et al., 2014). Molecular based diagnosis methods such as PCR, multiplex and nested PCR, Restriction Fragment Length Polymorphism (RFLP) have been used for more reliable results and discrimination of *Leishmania* species in the recent decades (Schallig and Oskam, 2002; Andrade et al., 2006; Töz et al., 2013; Ayele and Seyoum, 2016). These techniques allow researchers to determine *Leishmania* species by targeting various regions of the nuclear and kinetoplast DNA (le Fichoux et al., 1999; Cortes et al., 2004). The mini-circle regions of kinetoplast DNA are particularly important in detecting *Leishmania* species (Noyes et al., 1998; Lachaud et al., 2002; Bensoussan et al., 2006). Therefore, in the present study, the PCR method

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was used to amplify the regions of kinetoplast DNA (kDNA) minicircles of the *Leishmania* genus and the *L. infantum/donovani* complex. Additionally, microscopic examinations were performed; however, no amastigotes were found in any of the samples. Similarly, Ozerdem et al. (2009) reported that microscopic examination of peripheral blood for amastigotes had a sensitivity of 76% and 34.5% compared to the rK39 dipstick test and PCR methods, respectively, whereas the PCR method showed a sensitivity of 100%. These results indicate that amastigotes cannot always be detected in peripheral blood, even when the animal is infected with parasites. Hence, it is concluded that examining different materials using multiple methods would be beneficial for obtaining the most accurate results.

In the present research, three out of the five samples that were found positive for *Leishmania* at the genus level were also *Leishmania* positive in the rapid test kits applied in the veterinary clinics where the samples were taken. However, interestingly, among the 120 samples analyzed, five other samples found positive with the rapid test kit but yielded negative results in the PCR analysis in this study. Rapid test kits are commonly used in clinics for the diagnosis of diseases due to their practical application and ability to provide quick results. Whereas, as demonstrated in this study, it is important to note that these test kits may not always yield accurate results. On the other hand, it should be noted that PCR testing may not always give accurate results for *Leishmania* species, especially in peripheral blood samples. Therefore, if possible, it is essential to examine biopsy samples from spleen, liver, skin, bone marrow or lymph node aspirates or blood serum in addition to blood for confirmation (Schallig and Oskam, 2002). Besides, using multiple diagnostic methods (molecular, serological, etc.) together ensures the reliability and accuracy of the results.

According to the results of the previous studies; the prevalence of canine leishmaniasis (regardless of diagnostic method) were reported between 0-37.4% from different geographical regions of Turkey (Aslan Çelik et al., 2019) and the most prevalent is Aegean region with a rate of 37.4% (Bakırcı and Topçuoğlu, 2021). Then followed by Mediterranean (12.96%), Central Anatolia (5.82%), Black Sea (5.38%), Eastern Anatolia (4.38%), Marmara (2.40%), and Southeastern Anatolia regions (0%) (Aslan Çelik et al., 2019). According to literature review, a serological prevalence study from Burdur province was encountered where 49 dogs were examined, and all were found to be negative (Beyhan et al., 2016). In the present research, molecular positivity rates were found to be 4.16% for *Leishmania* spp. and 3.33% for *L. infantum* in dog blood samples. With this result, molecular-based PCR method was used for the first time to report *Leishmania* spp. and *L. infantum* positivity from Burdur.

In the present study, a higher percentage of positivity was found in female dogs (4.76%) compared to males (1.75%). However, the difference was not statistically significant among genders. Similarly, there was no statistically significant difference among age groups. In addition, the number of positive dogs by breed was insufficient for statistical analysis. Therefore, inadequate sample size and the number of positive samples hinders a comprehensive evaluation of the impact of gender, age, and breed on canine leishmaniasis positivity. To better understand whether these parameters are predisposing factors for leishmaniasis, further studies with a larger number of samples are needed. However, it is crucial to remember that vector fly control remains a much more prioritized issue in preventing the spread of the disease.

Regionally, differences in climate conditions, significantly influencing vector populations and the presence of animal reservoir hosts, as well as environmental and ecological changes and migration movements are other important factors affecting the spread of leishmaniasis (Parker et al., 2021). Therefore, rehabilitating the habitats of vector flies holds great importance in disease prevention. Additionally, controlling human and animal movements, conducting regular screening

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studies to obtain epidemiological data on the prevalence of the pathogen and vectors, treating carrier humans and animals, are all extremely important for both animal and human health.

Another crucial aspect in terms of leishmaniasis epidemiology is determining the habitats and distributions of sand fly species that act as vectors for *Leishmania* species. In Burdur, where this study was conducted, the presence of species such as *Phlebotomus kandelakii* s.l., *Adlerius* spp., *P. tobbi*, *P. major* s.l., *Larroussius* spp., *P. papatasi*, *Transphlebotomus* spp., *P. sergenti* s.l., *Paraphlebotomus* spp., *P. perfiliewi*, *P. halepensis* and *S. dentata* has been reported (Kaynaş, 2019). Among them, *P. kandelakii*, *P. perfiliewi* and *P. tobbi* are proven vectors for *L. infantum*, while *P. halepensis* is a suspected vector (Cunze et al., 2019). The presence of these three proven and one suspected vector species in the region supports the presence of *L. infantum* (3.33%) detected in dog blood samples in the present study. In addition, in the same study (Kaynaş, 2019), sand fly species identified, such as *P. sergenti* and *P. papatasi* are proven vectors for *L. tropica* and *L. major* respectively (Cunze et al., 2019). Some reports indicating the presence of both *L. tropica* and *L. major* exists in Turkey (Akman et al., 2000; Töz et al., 2013; Zeyrek et al., 2014). In the present study, although the isolate showed *Leishmania* spp. positivity at the genus level, its species-specific PCR result indicated it was not positive for *L. infantum*. Due to the presence of its vectors in this region, the isolate could potentially be *L. tropica* or *L. major*. Therefore, due to the rapid changes in climate conditions caused by global warming, which lead to alterations and expansions in vector habitats, it becomes evident that further studies are needed concerning both the epidemiology of vectors and *Leishmania* species in Burdur, as well as in other regions of Turkey and the rest of the world.

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Research Article / Araştırma Makalesi

Evaluation of the Gender Determining Features of Upper and Lower Extremity Morphometric Measurements in the Newborn

Yenidoğanda Üst ve Alt Ekstremite Morfometrik Ölçümlerinin Cinsiyet Belirleyici Özelliklerinin Değerlendirilmesi

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Abstract: Somatometric measurements are frequently used for identification in forensic investigations. In particular, data on the extremities may be needed to determine the age and gender of the dismembered corpse remains as a result of mass disasters. In the literature, the limitations of studies on sex determination in the prepubertal period are frequently mentioned. Therefore, in our study, it was aimed to determine the morphometric measurements of the upper and lower extremities in the neonatal period and to evaluate them in terms of gender determination. The study was conducted on 399 newborns (196 females, 203 males; 326 Turkish, 73 Syrian) newborns with no external anomaly and pathology. Initially, length and width parameters were measured for the upper and lower extremities of the newborns. Subsequently, index values were determined to examine the proportional relationship between the upper and lower extremities The obtained data were evaluated statistically and compared according to gender and groups. According to the results, in the comparison of all parameter and index values obtained from the upper and lower extremities, it was observed that there was no statistically significant difference between the genders, except for the leg/thigh length index. It is thought that the newborn extremity parametric values obtained in the study will contribute to fields such as forensic sciences, fetopathology, anatomy, obstetrics and pediatrics in terms of growth-development and gender determination.

Keywords: Newborn, Gender determination, Morphometry, Upper extremity, Lower extremity.

Öz: Somatometrik ölçümler adli soruşturmalarda kimlik tespitinde sıklıkla kullanılmaktadır. Özellikle kitlesel felaketler sonucu parçalanan ceset kalıntılarının yaşını ve cinsiyetini belirlemek için ekstremitelere ilişkin verilere ihtiyaç duyulabilir. Literatürde ergenlik öncesi dönemde cinsiyet belirlemeye yönelik çalışmaların sınırlılıklarından sıklıkla bahsedilmektedir. Bu nedenle çalışmamızda yenidoğan döneminde üst ve alt ekstremite morfometrik ölçümlerinin belirlenmesi ve cinsiyet tespiti açısından değerlendirilmesi amaçlandı. Çalışma herhangi bir dış anomali ve patolojisi olmayan 399 (196 kız, 203 erkek; 326 Türk, 73 Suriyeli) yenidoğan üzerinde gerçekleştirildi. Bu kapsamda öncelikle yenidoğanın üst ve alt ekstremite uzunluk ve genişlik parametreleri ölçüldü, daha sonra üst ve alt ekstremite arasındaki orantısal ilişkinin incelenmesi amacıyla indeks değerleri belirlendi. Elde edilen veriler istatistiksel olarak değerlendirilerek cinsiyet ve gruplara göre karşılaştırıldı. Sonuçlara göre üst ve alt ekstremiteden elde edilen tüm parametre ve indeks değerleri karşılaştırıldığında bacak/uyluk uzunluğu indeksi dışında cinsiyetler arasında istatistiksel olarak anlamlı bir fark olmadığı görüldü. Çalışmada elde edilen yenidoğan ekstremite parametrik değerlerinin büyüme-gelişme ve cinsiyet tespiti açısından adli bilimler, fetopatoloji, anatomi, kadın doğum ve pediatri gibi alanlara katkı sağlayacağı düşünülmektedir.

Anahtar Kelimeler: Yenidoğan, Cinsiyet tespiti, Morfometri, Üst ektremite, Alt ektremite.

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Introduction

Identification of human remains with impaired body integrity or reduced to skeletal remains is an important part of forensic investigations (Krogman and İscan, 1986; Ubelaker, 2006). The process of determining the gender, age, height, and ethnic origin of a living or deceased individual is referred to as identification. In this identification process, gender, age, and height play a crucial role

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in accurate determination (Scheuer, 2002; Sing et al., 2017). In forensic investigations, the identification of intact human bodies can be easily achieved. However, difficulties arise when attempting to identify fragmented bodies or skeletal parts in mass fatalities. In such cases, anthropometric and somatometric methods are employed (Krogman and İscan, 1986; Scheuer, 2002; Sing et al., 2017).

Gender determination often relies on morphological and metric findings from the skull, pelvis, and extremity bones (Çöloğlu and İscan, 1998; Loth and Henneberg, 2001). Especially in cases of dismembered bodies, the evaluation of structural features and detailed morphometric measurements of the extremities becomes essential for identifying a limb belonging to a specific corpse (Scheuer, 2002; Sing et al., 2017). Additionally, to determine the gender and height of a dismembered body part, somatometric and anthropometric measurements obtained from the extremities of living individuals are needed.

Anatomically, the structure of extremities displays a complex organization. This complexity allows for the formation of individual-specific extremities. The individual-specific shaping of extremities is influenced by various factors, including genetic factors, climate, dietary habits, environmental factors, physical activity, and more. As a result, these factors can lead to differences among societies or between genders within the same society. Metrics measurements play a crucial role in identifying these differences and serve as a guide, especially in forensic investigations and gender-specific anatomical evaluations (Giles and Vallandigham, 1991; Gordon and Buikstra, 1992; Fessler et al., 2005; ; Özden et al., 2005; Kumar et al., 2015).

There are studies that evaluate the parameters of upper and lower extremities at different stages of human development. However, these studies mostly focus on post-pubertal periods, and research on the prepubertal period is limited. The importance of increasing data on prepubertal periods is emphasized in the literature. Therefore, our study aims to assess morphometric measurements of upper and lower extremities in newborns and evaluate their potential for gender determination. Specifically, we have measured general parameters, as well as width and length measurements of upper and lower extremities externally in newborns (0-1 month) and compared them based on gender. The results obtained from this study are expected to be beneficial in various fields, including forensic sciences, anatomy, anthropology, obstetrics, and pediatrics.

Materials and Methods

The research was conducted on 399 (196 female (F), 203 male (M)) newborns (aged between 0 and 1 month) born at the TC Ministry of Health Istanbul Sancaktepe Şehit Prof. Dr. İlhan Varank Education and Research Hospital, with permission obtained from their families. Prior to the research, ethical committee approval was obtained. The study included infants with no external pathology or anomalies. Newborn measurements were taken considering bone reference points that can be externally detected.

In this study, morphometric parameters of the upper and lower extremities of the newborns were measured. Subsequently, the index values mentioned below were calculated to examine the proportional relationship between the upper and lower extremities. The measurements were taken using a tape measure, plastic ruler, anthropometric set, and a digital caliper sensitive to 0.01 mm. The study involved measuring various parameters of the upper and lower extremities in newborns. The measurements and index values used in the study are as follows:

Upper Extremity Parameters:

Arm length: Distance between the shoulder (acromion) and elbow (olecranon midpoint).

Forearm length: Distance between the elbow (olecranon midpoint) and wrist (styloid process of the radius).

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Hand length: Vertical distance between the outer edges of the distal radius and ulna (styloid processes) and the tip of the middle finger.

Hand width: Transverse distance between the outer edges of the hand at the level of the second and fifth metacarpophalangeal joints.

Hand digit lengths: Distances between the midpoints of the metacarpophalangeal joints and the distal tips of 1st, 2nd, 3rd, 4th and 5th fingers.

Upper Extremity Index Values:

Forearm/Arm Index: Forearm Length/Arm Length x 100

Hand/Arm Index: Hand Length/Arm Length x 100

Hand Index: Hand Width/Hand Length x 100

Lower Extremity Parameters:

Femur length: Vertical distance between the greater trochanter and the midpoint of the knee joint.

Leg length: Distance between the midpoint of the knee joint and the lower end of the lateral malleolus of the fibula.

Foot width: Transverse distance between the outer edges of the foot at the level of the first and fifth metatarsophalangeal joints.

Plantar length: Distance between the midpoint of the digitopalmar crease and the furthest point on the back of the heel.

Foot digit lengths: Distances between the midpoints of the metatarsophalangeal joints and the distal tips of 1st, 2nd, 3rd, 4th and 5th toes.

Lower Extremity Index Values:

Leg/Femur Index: Leg Length/Femur Length x 100

Foot/Leg Index: Plantar Length/Leg Length x 100

Foot Index: Foot Width/Plantar Length x 100

Additionally, the 2D:4D, 2D:5D, 3D:4D, 3D:5D, and 4D:5D ratios were calculated for both the hands and feet. These ratios involve comparing the lengths of the second, third, fourth, and fifth fingers or toes in relation to each other.

Statistical Analysis

The data analysis was performed using the statistical package program SPSS 20.0 in the Windows environment. Mean and standard deviation of all parameters were determined for each gender. In the statistical evaluation, a significance level of $p<0.05$ was considered. For parametric data, gender-based comparisons were conducted using the Student's t-test.

Results

In the study, measurements were conducted on a total of 399 newborns, including 196 females and 203 males, who had no external pathologies or anomalies. Among the female newborns, 167 were Turkish and 30 were Syrian refugees, while among the male newborns, 159 were Turkish and 43 were Syrian refugees.

The upper and lower extremity parameter measurements were statistically evaluated based on groups and gender, and their mean and standard deviations were determined ($p>0.05$; Table 1, Table 2, Table 3). Subsequently, the mean and standard deviations of the index values related to the upper and lower extremities were determined ($p>0.05$; Table 1, 2, 3, 4 Figure 1, 2, 3, 4).

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Table 1. Mean and standard deviation of upper extremity parameters from newborns of Turkish and Syrian according to gender (mm).

Gender	Case (N)	Arm length	Forearm length	Hand length	Hand width	Forearm/Arm ındex	Hand/Arm ındex	Hand ındex
Turkish F (Mean±SD)	167	79,66±1,17	67,29±1,29	63,65±1,12	30,84±1,28	84,49±1,98	79,92±1,80	48,47±2,13
Syrian F (Mean±SD)	30	79,73±0,94	66,87±1,28	63,40±1,13	31,07±1,05	83,87±1,82	79,52±1,57	49,02±1,87
Total F (Mean±SD)	196	79,66±1,14	67,25±1,26	63,61±1,12	30,88±1,25	84,43±1,91	79,87±1,77	48,55±2,10
Turkish M (Mean±SD)	159	79,78±1,14	67,22±1,32	63,74±1,08	30,94±1,16	84,27±2,03	79,90±1,66	48,56±2,08
Syrian M (Mean±SD)	43	79,74±1,14	67,26±1,16	63,51±1,14	31,02±1,14	84,36±1,89	79,66±1,75	48,86±1,95
Total M (Mean±SD)	203	79,78±1,14	67,21±1,32	63,69±1,09	30,96±1,15	84,26±2,04	79,85±1,68	48,62±1,04
Total F+M (Mean±SD)	399	79,72±1,14	67,23±1,29	63,65±1,11	30,92±1,20	84,35±1,98	79,86±1,72	48,59±2,07

There were no significant difference between genders in totaly and within groups (p>0.05). F: female, M: male.

Table 2. Mean and standard deviation of lower extremity parameters from newborns of Turkish and Syrian according to gender (mm).

Gender	Case (N)	Femur length	Leg length	Plantar length	Foot width	Leg/Femur ındex	Foot/Leg ındex	Foot ındex
Turkish F (Mean±SD)	167	99,30±1,57	94,31±1,27	59,95±1,33	30,13±1,03	95,00±1,90	60,39±162	50,28±1,95
Syrian F (Mean±SD)	30	99,47±1,53	94,73±0,91	60,17±1,12	29,87±0,86	95,26±1,55	60,50±1,32	49,65±1,54
Total F (Mean±SD)	196	99,30±1,52	94,39±1,22	59,99±1,30	30,10±1,01	95,07±1,80	60,43±156	50,18±1,90
Turkish M (Mean±SD)	159	99,01±1,40	94,60±1,22	59,97±1,29	30,12±1,25	95,57±1,82	60,58±1,53	50,24±2,24
Syrian M (Mean±SD)	43	99,19±1,01	94,53±1,12	60,26±1,31	30,02±1,44	95,32±1,43	60,76±156	49,86±2,79
Total M (Mean±SD)	203	99,07±1,37	94,58±1,21	60,02±1,30	30,09±1,29	95,48±1,81	60,60±1,55	50,17±2,14
Total F+M (Mean±SD)	399	99,18±1,45	94,48±1,22	60,01±1,30	30,10±1,16	95,28±1,81	60,51±1,56	50,17±2,15

There were no significant difference between genders in totaly and within groups, (p>0.05) (except for Leg/Femur ındex, differences between genders in totally, p=0,24) F: female, M: male.

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Table 3. Mean and standard deviation of finger and toe length parameters from newborns of Turkish and Syrian according to gender (mm).

Gender	Case (N)	1st digit length	2nd digit length	3rd digit length	4th digit length	5th digit length
Hand						
Turkish F (Mean±SD)	167	27,65±1,14	29,38±1,14	30,13±1,03	31,98±1,34	28,10±1,44
Syrian F (Mean±SD)	30	27,53±1,17	29,10±1,12	29,87±0,86	31,80±0,61	28,03±1,94
Total F (Mean±SD)	196	27,64±1,14	29,33±1,14	30,10±1,01	31,95±1,26	28,09±1,52
Turkish M (Mean±SD)	159	27,56±1,14	29,28±1,20	30,12±1,25	31,97±1,07	28,02±1,37
Syrian M (Mean±SD)	43	27,51±1,20	29,09±1,11	30,02±1,44	31,70±1,12	27,72±0,83
Total M (Mean±SD)	203	27,54±1,15	29,25±1,18	30,09±1,29	31,91±1,08	27,96±1,27
Total F+M (Mean±SD)	399	27,59±1,15	29,29±1,16	30,10±1,16	31,93±1,17	28,02±1,40
Foot						
Turkish F (Mean±SD)	167	17,44±0,61	14,57±0,58	14,40±0,59	13,76±0,38	13,70±0,32
Syrian F (Mean±SD)	30	17,56±0,67	14,57±0,57	14,61±0,51	13,80±0,37	13,65±0,26
Total F (Mean±SD)	196	17,46±0,62	14,57±0,58	14,44±0,58	13,77±0,38	13,69±0,31
Turkish M (Mean±SD)	159	17,46±0,59	14,55±0,57	14,36±0,59	13,77±0,37	13,66±0,32
Syrian M (Mean±SD)	43	17,35±0,57	14,52±0,57	14,38±0,56	13,79±0,32	13,65±0,30
Total M (Mean±SD)	203	17,43±0,59	14,54±0,57	14,36±0,59	13,77±0,36	13,65±0,31
Total F+M (Mean±SD)	399	17,45±0,61	14,55±0,57	14,40±0,58	13,77±0,37	13,67±0,31

There were no significat difference between genders in totaly and within groups (p>0,05). F: female, M: male.

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Table 4. Mean and standard deviation of finger and toe ratios from newborns of Turkish and Syrian according to gender.

Gender	Case (N)	2D:4D ratio	2D:5D ratio	3D:4D ratio	3D:5D ratio	4D:5D ratio
Hand						
Turkish F (Mean±SD)	167	0,92±0,05	1,05±0,07	0,95±0,05	1,07±0,06	1,14±0,07
Syrian F (Mean±SD)	30	0,92±0,04	1,04±0,07	0,94±0,03	1,07±0,07	1,14±0,06
Total F (Mean±SD)	196	0,92±0,04	1,04±0,04	0,94±0,04	1,07±0,06	1,14±0,05
Turkish M (Mean±SD)	159	0,92±0,04	1,05±0,06	0,94±0,05	1,08±0,06	1,14±0,06
Syrian M (Mean±SD)	43	0,92±0,05	1,05±0,05	0,95±0,06	1,08±0,06	1,14±0,05
Total M (Mean±SD)	203	0,91±0,04	1,04±0,05	0,94±0,05	1,07±0,06	1,14±0,05
Total F+M (Mean±SD)	399	0,92±0,05	1,05±0,06	0,95±0,05	1,08±0,06	1,14±0,06
Foot						
Turkish F (Mean±SD)	167	1,06±0,05	1,06±0,05	1,05±0,05	1,05±0,05	1,01±0,04
Syrian F (Mean±SD)	30	1,06±0,05	1,07±0,05	1,06±0,05	1,07±0,04	1,01±0,03
Total F (Mean±SD)	196	1,06±0,04	1,06±0,04	1,04±0,05	1,05±0,05	1,00±0,03
Turkish M (Mean±SD)	159	1,06±0,05	1,07±0,05	1,04±0,05	1,05±0,05	1,01±0,04
Syrian M (Mean±SD)	43	1,05±0,05	1,06±0,05	1,04±0,05	1,05±0,05	1,01±0,03
Total M (Mean±SD)	203	1,05±0,05	1,06±0,04	1,04±0,04	1,05±0,04	1,00±0,03
Total F+M (Mean±SD)	399	1,06±0,05	1,07±0,05	1,05±0,05	1,05±0,05	1,01±0,04

There were no significant difference between in totally and within the groups with respect to genders ($p>0,05$). F: female, M: male.

Discussion

The structure of the extremities is a complex organization formed by the ideal alignment of bones, muscles, and joints from an anatomical perspective. This structure enables the formation of unique hand and foot shapes specific to individuals. Anthropometric or somatometric measurements are taken to reveal these differences and determine the intersexual variations within the community. Moreover, these measurements are essential for obtaining population-specific data and identifying differences between populations. This is particularly significant for forensic sciences in guiding forensic identification (Giles and Vallandigham, 1991; Fessler et al., 2005; Özden et al., 2005; Kumar et al., 2015).

The most critical elements in identity determination are age, gender, race, and height (Krogman and İscan, 1986; Ubelaker, 2006). Gender determination is a crucial and foremost criterion in identifying an individual. In forensic applications, rapid identification of the probable gender of a decomposed body or body part is expected (Mall et al., 2001). Gender determination is often considered one of the simplest tasks in forensic analysis because external and internal genital organs can directly determine an individual's gender. However, in intersex cases, highly decomposed bodies, mutilated, fragmented, and skeletal remains, gender differentiation becomes complex (Kanchan and Krishan, 2011). Therefore, in cases where gender cannot be determined from primary anatomical structures, anthropometric or somatometric methods are employed (Krogman and İscan, 1986; Gupta et al., 2017).

The emergence of DNA technology in forensic investigations has significantly simplified gender determination. However, this technology may not be a reasonable option for identifying the identity of bodies that have been fragmented or whose body integrity has been compromised in mass disaster incidents like natural disasters, transportation accidents, wars, terrorism, and bombings. DNA technology, especially in developing countries and cases where DNA analysis is not feasible, may have limitations concerning qualified human resources, time, and financial resources. Therefore, the development of alternative methods is essential (Scheuer, 2002; Kanchan and Krishan, 2011; Sing et al., 2017).

In forensic investigations, gender determination is sometimes made using extremity bones if necessary (Çöloğlu and İscan, 1998; Loth and Henneberg, 2001). Additionally, alternative studies have been conducted to determine the gender of a fragmented body based on the available parts in cases of mass deaths. These studies utilize hand measurements, arm length, leg length, and foot measurement data obtained from living individuals to identify the gender of the body (İris and Celbiş, 2003; Jasuja and Singh, 2004; Sanlı et al., 2005; Krishan and Sharma, 2007; Sahni et al., 2010; Agnihorti et al., 2011).

There are studies evaluating upper and lower extremity parameters at different stages of human development. However, studies specifically focusing on morphometric parameters of both upper and lower extremities in the newborn period are relatively rare. In this study, unlike previous research, a wide range of newborns was included, and both upper and lower extremity parameters were evaluated morphometrically together. Additionally, the fact that the measurement series consisted of newborns from both Turkish and Syrian backgrounds allowed for the comparison of different ethnic groups.

In previous studies, it has been suggested that the development of the urogenital system and external genitalia, as well as extremity development, may reflect differences in prenatal androgen exposure (Kondo et al., 1997; Ernsten et al., 2021; Goodman, 2002; McIntyre, 2006). Sex hormones such as androgens and estrogens at a genetic level support the phenotypic differentiation of males and females secondarily. However, it is worth noting that these studies have mainly focused on either the prenatal period or the postnatal period after puberty. Studies covering the prenatal period emphasize the significance of prenatal androgen exposure in determining gender-specific outcomes, while studies covering the postnatal

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period after puberty highlight the role of activated gonadal hormones in gender differentiation (Ernsten et al., 2021). It is particularly emphasized that reliable observations of gender differences can be made during adolescence when puberty starts. However, there are very few studies focused on the prepubertal period.

One of the phenotypic differences between males and females is related to the hands. Generally, males have longer fingers and broader and longer hands compared to females (Amirsheybani et al., 2001). Therefore, hands and fingers tend to show sexual dimorphism as a reflection of prenatal androgen exposure (Ernsten et al., 2021). There are studies covering the prenatal period related to this assumption (Malas et al., 2006, 2008). However, no significant gender differences in hand length, hand width, and hand index values have been observed during the fetal period (Malas et al., 2006, 2008). Even in prepubertal children, no differences in hand size between genders have been observed (Raziye et al., 2016; Ernsten et al., 2021).

However, it is noted that most of the studies in the literature related to hand and finger measurements are conducted on adult samples, and there are very few studies that include preadolescent individuals (Cohen-Bendahan et al., 2005; Arnold, 2009; Kanchan and Rastogi, 2009; Hönekopp and Watson, 2010; Krishan et al., 2011; Jowaheer and Agnihotri, 2011; Manning, 2012).

Kanchan and Rastogi, (2006) conducted evaluations on hand length and width measurements in an adult population and reported gender classification accuracy rates ranging from 81.7% to 91.9%. They especially emphasized that hand width is generally the strongest dimorphic measurement (Kanchan and Rastogi, 2009). Jowaheer and Agnihotri, (2011) used multiple regression models to investigate the gender identification potential of hand length and found that 91.2% of adult individuals were correctly classified by gender for both their right and left hands (Jowaheer and Agnihotri, 2011). Krishan et al., (2011) determined gender biases at rates of 79.5% for hand width and 86.0% for hand length using hand width and length measurements, and they mentioned that hand width is more dimorphic than hand length (Krishan et al., 2011).

One of the phenotypic differences between genders is the feet. A literature review indicates that fetal feet have a characteristic normal growth pattern and show gradual length increase relative to the embryo's length, which can be used to estimate gestational age (Kumar and Kumar, 1993). Besides determining gestational age, foot length parameter also appears as an alternative parameter in detecting fetal pathologies. Kumar and colleagues have shown that fetal hands and feet also have a characteristic normal growth model, and they propose that fetal hand and foot length can be used to estimate gestational age (Kumar and Kumar, 1993).

In our study, the mean and standard deviations of length and width parameters of upper and lower extremities of newborns were determined according to gender (Table 1, 2). According to the results obtained in our study, there were no significant differences between genders in the length and width parameters of all newborn upper extremities ($p>0.05$; Table 1). Similarly, when evaluated according to gender within the same nationality (male-female), no significant gender differences were found, and when comparing the same genders in both nationalities (female-female, male-male), there were no significant differences in terms of nationality ($p>0.05$; Table 1).

Regarding the evaluation of lower extremity length and width parameters, there was only a significant gender difference in foot length ($p=0.04$) among all newborns, while no other lower extremity parameters showed significant differences ($p>0.05$; Table 2). In the comparison of Turkish and Syrian newborns' lower extremity parameters (female-female, male-male), no significant differences were found between Turkish female and male newborns, except for leg length ($p=0.03$), and no differences were found in any

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lower extremity parameter between Syrian female and male newborns ($p>0.05$; Table 2). Moreover, there were no statistically significant differences in any lower extremity parameter between male and female newborns of different nationalities ($p>0.05$; Table 2).

In the literature, hand and foot index values are frequently mentioned as potential indicators for determining gender. Studies conducted on postpubertal individuals in different age groups suggest that the index values obtained from hand and foot length and width measurements can be effective in determining gender. Gupta et al., (2017) conducted a study on 300 adults (150 females, 150 males) and performed hand and foot measurements and index calculations. They found that hand length and width were larger in males compared to females. Similarly, they noted that hand index values were also greater in males than in females. Moreover, they reported that foot length and width were greater in males than in females, and the foot index was also larger in males. Therefore, it was emphasized that both hand and foot length, width, and index values can be used as gender determinants (Gupta et al., 2017; Ernsten et al., 2021).

In studies conducted during the prepubertal period, similar results were not observed. Ernstein et al., (2021) measured hand width, length, and index in 6-month-old infants (364 females, 399 males). According to the results obtained, average hand width and length values were reported to be larger in males. Additionally, hand index values were compared between genders, and no significant difference was found between the hand indices of boys and girls (Ernsten et al., 2021). Malas et al., (2008) conducted a study on newborns, including 60 infants (30 females, 30 males), and reported that average hand length and width did not show statistically significant differences between boys and girls (Malas et al., 2008).

In the current study, the mean values of extremity parameters were obtained from newborns. When comparing these values, a significant gender difference was found in the leg/thigh index in all newborns ($p=0.24$) and in Turkish newborns ($p=0.06$). However, no statistically significant differences were observed in other index values between the same genders within the same nationality and between the same genders of different nationalities (female-female, male-male) ($p>0.05$; Table 1, 2; Figure 1, 2). The results of the hand index in our study are consistent with other studies that cover the prepubertal period. According to the obtained results, hand and foot length, width, and index values were not determinative for gender identification in the newborn period. However, it is suggested that leg/thigh index values may be decisive for gender determination during the prepubertal period and should be supported by new studies with larger sample sizes.

The second to fourth digit ratio (2D:4D) is one of the most debated indicators in the literature, known to differ between males and females. In these studies, it has been proposed that males have a lower 2D:4D ratio compared to females (Gordon et al., 1991; Patriquin et al., 2005; Krishan et al., 2011). It has been emphasized that this gender difference in the ratio can be detected from the 14th week of pregnancy, but may vary over time (Knickmeyer et al., 2011; Wong and Hines, 2016). Besides the 2D:4D ratio, the proportional evaluation of the fingers, including the fifth finger, has also been suggested to be an alternative for gender determination (McIntyre et al., 2006; Dressler and Voracek, 2011; Kumar et al., 2017). On the other hand, a study on children between the ages of 2 and 18 has stated that evaluations other than the 2D:4D ratio may not be reliable indicators (Manning, 2012). However, it is crucial to consider the variations in study designs and measurement techniques in these studies while comparing and interpreting the results (Aboul-Hagag et al., 2011; Kanchan and Krishan, 2011). Research on sexual dimorphism in finger lengths has revealed that males tend to have longer fourth and fifth fingers, while females have longer second and third fingers (Loehlin et al., 2009; Stenstrom

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et al., 2011). As evident from the literature, there are numerous studies supporting the assumption that hand and finger measurements reflect sexual dimorphism. The literature contains limited studies specifically focusing on hand and finger lengths as well as foot and toe lengths in prepubertal individuals (Malas et al., 2008; Ernsten et al., 2021). Hence, comparing the data obtained in our study poses some challenges.

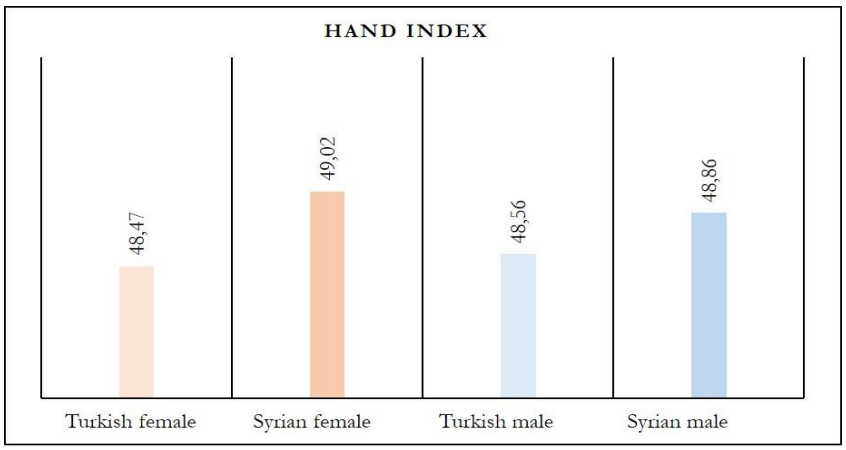

Figure 1. Hand index values in Turkish female and male newborns and Syrian female and male newborns.

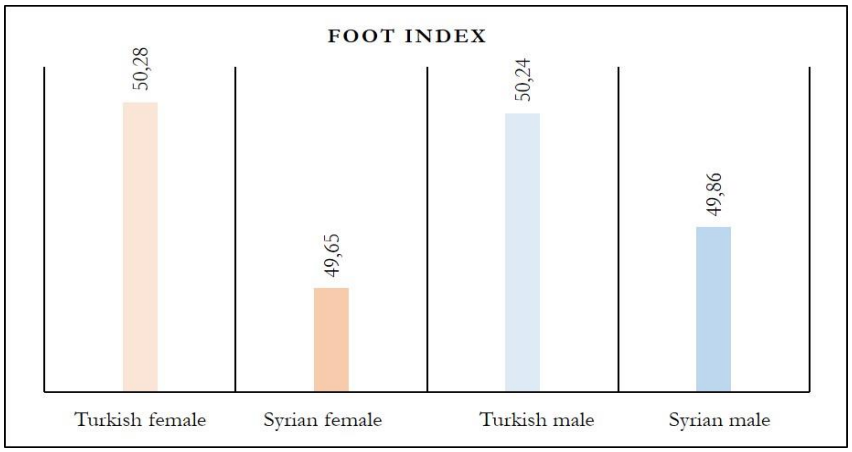

Figure 2. Foot index values in Turkish female and male newborns and Syrian female and male newborns.

In the present study, hand and foot finger lengths were evaluated. A thorough review of the literature indicates that finger lengths, and even the proportional relationships between fingers, have been frequently emphasized as indicators for gender determination. Studies suggest that phalangeal lengths grow faster in boys (Butovskaya et al., 2021). Particularly in prepubertal children, finger lengths tend to be longer in girls compared to boys, while the 2D:4D ratio shows the expected sexual dimorphism (men < women) (Manning and Fink, 2018). However, after the age of 13, sexual dimorphism in finger lengths becomes more pronounced, with boys having longer finger lengths than girls. In another study, it was observed that initially girls tend to have longer phalanges than boys, but around the age of 13, both sexes reach approximately equal phalangeal lengths (Butovskaya et al., 2021).

In the literature, it has been indicated that finger growth in males continues even after the age of 18.

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Therefore, finger length values between the ages of 20 to 30 are considered to be important in terms of gender differences. However, despite the periodic variations in phalangeal lengths in terms of gender, the ratio of phalangeal lengths to each other remains relatively constant. Studies suggest that gender differences in 2D:4D are not related to finger lengths, and the sexual dimorphism in 2D:4D remains stable (Butovskaya et al., 2021). In the study conducted by Malas et al., (2008), it was reported that 2nd and 4th finger length measurements did not show gender differences, but the 2D:4D finger ratio exhibited sexual dimorphism (men < women) (Malas et al., 2008).

In our study, first, finger lengths of hands and feet were statistically evaluated, and no significant differences were found between male and female genders in all newborns ($p>0.05$; Table 3,4). Comparisons were made between same-gender newborns within the same ethnicity and between different genders within different ethnic groups, and similarly, no statistically significant differences were found ($p>0.05$; Table 3,4).

Studies in the literature have not only focused on the 2D:4D finger ratio but also explored the ratios of other fingers. Particularly, the relationship of the 5th finger with other fingers has been suggested to be effective in gender determination. In the study conducted by Ernsten et al. (2021), the ratios of all fingers except the thumb were examined, and they found that males had larger finger ratios compared to females. Additionally, besides the 2D:4D ratio, significant differences were observed in other finger ratios, such as 2D:3D, 2D:5D, 3D:4D, 3D:5D, and 4D:5D, between male and female infants. In the same study, relative finger lengths were emphasized as potential gender determinants (Ernsten et al., 2021). In our study, we measured finger ratios and for both hands and feet as defined in the literature. However, no statistically significant differences were found in finger ratios between male and female newborns and between Turkish and Syrian groups ($p>0.05$; Table 3,4; Figure 3, 4).

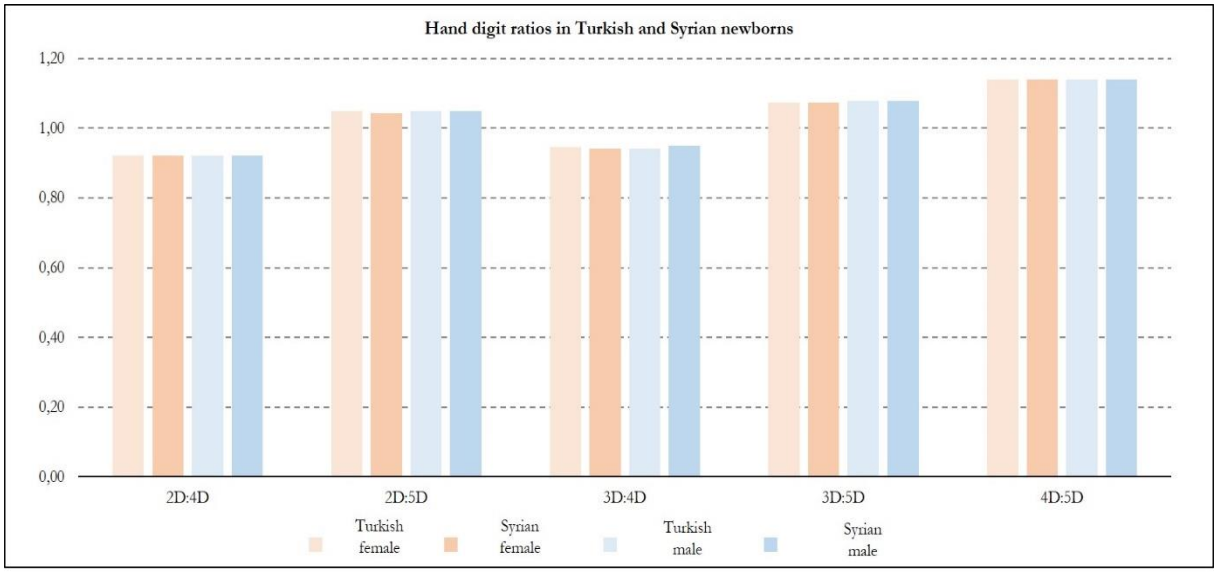

Figure 3. Hand digit ratios in Turkish female and male newborns and Syrian female and male newborns.

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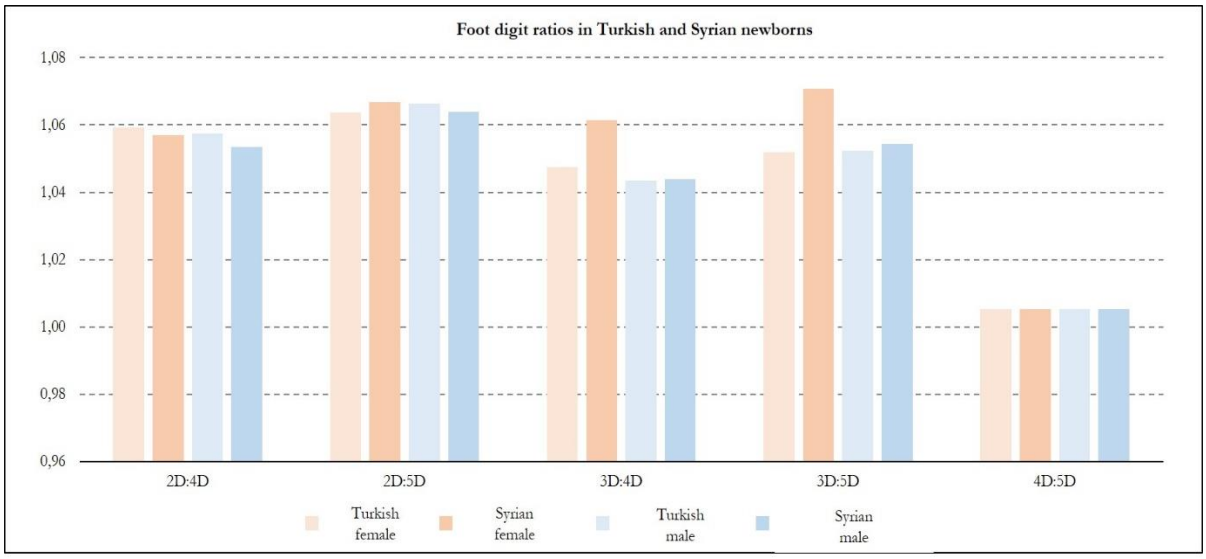

Figure 4. Foot digit ratios in Turkish female and male newborns and Syrian female and male newborns.

As a result, there are several studies evaluating upper and lower extremity parameters at different stages of human development. In these studies, somatometric or radiological methods were used to examine the upper and lower extremities of adults. In addition, finger, hand and foot measurements were evaluated in different age groups according to gender. However, studies focusing on newborns (0-1 months) in this developmental period are insufficient. İn our study upper and lower extremities, especially hand and foot region measurements on 399 newborns were examined according to gender. In particular, the fact that the sample group consisted of both Turkish and Syrian newborns enabled the comparison of ethnic groups. Although our findings partially overlap with previous studies, they also reveal some differences. These differences can be attributed to variation in sample size or differences in measurement techniques. We believe that the results obtained in our study will contribute to forensic sciences as well as anatomy, gynecology and pediatric sciences and will be a guiding reference for future research. Further work in this area is necessary to improve our understanding of limb development in newborns and its implications in different fields of medicine and science.

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Research Article / Araştırma Makalesi

Effect of Single or Combined Homo- and Heterofermentative Silage Additives on the Quality, Nutritive Value, and In Vitro Digestibility of Ensiled Wheat Harvested at Early Dough Stage of Maturity

Tekli veya Kombine Homo ve Heterofermentatif Silaj Katkı Maddelerinin Erken Hamur Olgunluk Aşamasında Hasat Edilen Silaj Buğdayının Kalitesi, Besin Değeri ve İn Vitro Sindirilebilirliği Üzerine Etkisi

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Abstract: The present study was conducted to evaluate the effect of single or combined homo- and heterofermentative silage additives on silage quality, nutritional composition, feed value, and *in vitro* digestibility of ensiled wheat harvested at early dough stage of maturity. The study was carried out as a completely randomized design with a 2 × 2 factorial arrangement of two levels of homofermentative silage inoculant (0 or 0.8 mg/kg) and two levels of heterofermentative silage inoculant (0 or 500 mg/kg) consisting of 4 groups with 4 replicates in each group. Control group received no silage additive. Remaining groups received either HMF, HTF, or a combination of both silage additives (HMF + HTF). pH and Flieg point of silage prepared with HMF and HTF alone or in combination were respectively lower and greater compared to control group. Nutritional composition, feed value, and *in vitro* true dry matter and organic matter digestibilities were unaffected among the treatments. In conclusion, the study shows that the application of single or combined HMF and HTF inoculants yields well-preserved wheat silage whereas the nutritional composition and *in vitro* digestibility may remain unaffected.

Keywords: Digestibilty, Inoculant, Nutritive value, Silage quality, Wheat..

Öz: Bu çalışma, erken hamur olum döneminde hasat edilen buğdayın tekli ya da kombine homofermentatif (HMF) veya heterofermentatif (HTF) mikrobiyal inokulantlar ile silolanmasının silaj kalitesi, yem değeri ve in vitro sindirilebilirlik üzerine etkilerini araştırmak amacıyla yapılmıştır. Denemede iki faklı düzeyde homofermentatif (0 veya 0,8 mg/kg) ve/veya heterofermentatif (0 ve 500 mg/kg) silaj inokulantı kullanılarak 2 × 2 deneme deseni uygulanmıştır. Her deneme grubu dört tekrar grubundan oluşturulmuştur. Kontrol grubuna herhangi bir silaj katkısı uygulanmazken; diğer gruplara HMF, HTF ya da bu ikisinin kombinasyonu (HMF + HTF) uygulanmıştır. Silaj inokulantı uygulanan grupların kontrol grubuna kıyasla daha düşük pH ve daha yüksek Flieg puanına sahip olduğu belirlenmiştir. Besin madde bileşimi, yem değeri ve in vitro kuru madde ve organik madde sindirilebilirliği açısından gruplar arasında fark oluşmamıştır. Sonuç olarak tekli ya da kombine HMF ve HTF inokulantların buğday silajının daha iyi korunabilmesini sağladığı ancak besin madde bileşimi ve in vitro sindirilebilirliğini etkilemediği belirlenmiştir.

Anahtar Kelimeler: Buğday, İnokulant, Beslenme değeri, Silaj kalitesi, Sindirilebilirlik.

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Introduction

Feeding practices of farmers in Turkey giving preference to cereals and straws instead of forages creates the shortage of forages and roughages (Arslan & Erdurmuş, 2012). It has resulted in an increase in the cultivation area of cereal crops compared to the forage crops (TÜİK, 2022). Wheat continues to be a major crop in Turkey especially for cereals and silage due to double cropping system in which wheat sown in winter is harvested in early or late spring to clear the fields

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for summer crops (Başkavak et al., 2008). Depending on the stage of maturity, wheat provides considerable dry matter (DM) with reasonable nutritive value for animal production (Filya, 2003a). Being a less expensive crop for ensiling, wheat has been considered an alternative for traditional silage crops i.e., grass and corn. However, the ensiling of wheat is an arduous task since it contains comparatively less water-soluble carbohydrates and starch. Additionally, the whole crop ensiling often ends in poor fermentation and aerobic stability resulting in higher butyrate concentration in the silage (Kaiser et al., 2003).

There are different strategies to improve the ensiling and to reduce the losses occurring at storage and feedout phases. Ensiling process of wheat can be improved using various silage additives of chemical or biological origin. Biological additives are easy-to-use and are not corrosive, therefore, the application of biological or bacterial additives is useful compared to their chemical counterparts. Studies have reported that the bacterial silage inoculants help in the fermentation, silage preservation, protection against pathogenic bacterial, and improvement of aerobic stability of silage that would other undergo spoilage thereby causing a loss in the nutritive value of the silage (Başkavak et al., 2008; Sucu and Filya, 2016). In general, bacterial silage additives are based on lactic acid bacteria (LAB) that may be homo- or heterofermentative. In practice, homofermentative (HMF) and heterofermentative (HTF) LAB silage additives are used to improve the fermentation and aerobic stability (Filya, 2003b), respectively. Although there are studies describing the use of HMF or HTF silage additives on the microbial composition and nutrient digestibility of ensiled bread wheat, there is still room for further study to evaluate the silage quality, nutritive value, and nutrient digestibility of wheat silage. In addition, a limited number of studies are available describing the wheat silage preserved using HMF and HTF silage additives. Therefore, the present study investigated the effect of single or combined application of HMF and HTF silage additives on the quality, nutritional composition, and nutrient digestibility of ensile durum wheat.

Materials and Methods

Location of the study

The study was conducted at the agricultural land of the Center for Agriculture, Livestock and Food Research, Burdur Mehmet Akif Ersoy University, Turkey in the western Mediterranean region of Turkey located 1280 m above the sea level.

Study design and experimental groups

The study was carried out as a completely randomized design with a 2 × 2 factorial arrangement of two levels of homofermentative silage inoculant (0 or 0.8 mg/kg) and two levels of heterofermentative silage inoculant (0 or 500 mg/kg) consisting of 4 groups with 4 replicates in each group. Control group received no silage additive. Remaining groups received either HMF, HTF, or a combination of both silage additives (HMF + HTF). The HMF silage additive consisted of *Lactobacillus plantarum* and *Enterococcus faecium* (Pioneer® brand 1188; Corteva Agriscience, Inc., IN, US) whereas HTF silage additive was comprised of *Lactobacillus buchneri* (Pioneer® brand 11A44; Corteva Agriscience, Inc., IN, US).

Wheat was sown by broadcasting in the mid of November 2021 (230 kg seed/hectare). Diammonium phosphate was used to fertilize the land (100 kg/hectare). Wheat was cultivated under dryland condition without irrigation. Wheat was harvested at the end of June 2022 at early dough stage of maturity. Fresh weight per m^2 and average plant height were measured by harvesting the forage using a quadrant at three different sites in the field. Additionally, three samples were collected to assess the nutritional composition of the forage.

Homofermentative silage additive consisted of 4 strains of *Lactobacillus plantarum* (2.5×10^{10} cfu/g *Lactobacillus plantarum* LP286 DSM 4784 ATCC 53187, 2.5×10^{10} cfu/g *Lactobacillus plantarum*

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LP318 DSM 4785, 2.5 × 10^{10} cfu/g *Lactobacillus plantarum* LP319 DSM 4786, and 2.5 × 10^{10} cfu/g *Lactobacillus plantarum* LP346 DSM 4787 ATCC 55943) and 2 strains of *Enterococcus faecium* (1.25 × 10^{10} cfu/g *Enterococcus faecium* SF301 DSM 4789 ATCC 55593 and 1.25 × 10^{10} cfu/g *Enterococcus faecium* SF202 DSM 4788 ATCC 53519) (Pioneer® brand 1188; Corteva Agriscience, Inc., IN, US).

Heterofermentative silage additive was comprised of 1.0 × 10^{11} cfu/g *Lactobacillus buchneri* LN4637 ATCC PTA-2494 (Pioneer® brand 11A44; Corteva Agriscience, Inc., IN, US).

Silage preparation, ensiling, opening, and physical quality assessment of ensiled wheat

Harvested wheat forage was cut to a particle size of 1.5-2.5 cm and vacuum packed in plastic bags (250 g in each bag) for ensiling after respective applications of silage additives. Control group was ensiled without the application of any additive whereas, prior to vacuum packing, silage additives were applied to the respective groups in accordance with the manufacturer's recommendations. Wheat forage was allowed to ensile for 120 days. The ensiled wheat forages were opened, and physical characteristics of each silage was assessed in terms of color (three-point scoring; 0 to 2), structure (four-point scoring; 0 to 4), and odor (15-point scoring; 0 to 14) following the DLG scoring method developed by German Agricultural Society (Deutsche Landwirtschafts Gesellschaft). A panel of three experts was employed to assess the physical quality of ensile wheat forage. The scores were summed up and categorized as follows according to the average score of the panel: bad (0 to 4 points), moderate (5 to 9 points), good (10 to 15 points), and excellent (16 to 20 points).

Fermentation characteristics and acidity of ensiled wheat forage

Following the assessment of physical quality, fermentation characteristics of ensiled wheat forages were evaluated in terms of Flieg point, pH, and ammonia nitrogen (NH_3-N). pH and dry matter (DM) of ensiled wheat forages were measured to calculate the Flieg point of each silage according to the method described by Dong et al. (2017) using equation below:

Flieg point = 220 + (2 × DM% – 15) – (40 × pH)

To measure the pH, 100 g of ensiled wheat forage was blended in 100 mL distilled water for 5 minutes, filtered through 4 layers of cheese cloth, and glass electrode of pH meter (Apera Instruments, LLC., Columbus, OH, US) was immersed into the filtrate to measure the pH of wheat silages.

The NH_3-N was measured according to the method previously described by Meeske et al. (2002). Briefly, 50 g silage was homogenized in 250 ml 0.1 N sulphuric acid followed by filtration of the homogenate through a four-layer cheesecloth. Finally, the filtrate was subjected to distillation and titration according to Kjeldahl method described by AOAC (2000).

Nutritive value of fresh and ensiled wheat forage

The DM of freshly harvested and ensile wheat forages were dried in hot air oven at 105ºC for 8 hours. Crude protein (CP), ether extract (EE), and crude ash were analyzed using AOAC (2000) methods. Crude fiber (CF), neutral detergent fiber (aNDFom), acid detergent fiber (ADFom), and acid detergent lignin (ADL) were analyzed using automatic fiber analyzer (ANKOM A2000 Fiber Analyzer, ANKOM Technology, NY, US). Other fractions were calculated according to the equations reported by Horrocks and Vallentine (1999) as follows:

Non-structural carbohydrates (%, DM basis) = 100 – (aNDFom + CP + Ash + EE)

Hemicellulose (%, DM basis) = aNDFom% – ADFom%

Digestible DM (%, DM basis) = 88.9 – (0.779 × ADFom%)

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DM intake (%, DM basis) = 120 ÷ aNDFom%

Relative feed value (%, DM basis) = DDM% × DMI% × 0.775

Net energy for lactation = [1.044 – (0.0119 × ADFom%)] × 2.205

Total digestible nutrients = (–1.291 × ADFom%) + 101.35

Total carbohydrates (%, DM basis) = DM% – (CP% + Ash% + EE%)

Cellulose (%, DM basis) = ADFom% – ADL%

In vitro rumen digestibility of fresh and ensiled wheat forage

Fresh and ensiled wheat forages were subjected to incubation in ANKOM Daisy^{II} incubator to investigate the *in vitro* true DM and OM, digestibilities. For this purpose, the samples in duplicates were placed in bottles of Daisy^{II} incubator containing ruminal fluid as inoculum from a slaughtered cow. The samples, packed in ANKOM F57 filter bags, were incubated for 24 and 48 hours. All the procedures were conducted under anaerobic conditions using carbon dioxide gas to ensure the anaerobic environment at each stage. *In vitro* true digestibilities were calculated for DM and OM.

Statistical analysis

The data were tested for normality followed by logarithmic or square root transformation of non-normalized traits. The data were subjected to two-way analysis of variance applying the general linear model procedures using a statistical software package SPSS (IBM Corp., Armonk, NY, US) according to the following model:

$$Y_{ijk} = \mu + s_i + l_j + e_{ijk}$$

Where:

Y_{ijk} = phenotypic value of the trait for the k^{th} group of silages belonging to i^{th} HMF and j^{th} HTF silage additives;

μ = mean value of the trait for a given population;

s_i = effect of i^{th} HMF additive (i = 1, 2);

l_j = effect of l^{th} HTF additive (j = 1, 2);

e_{jkl} = effect of experimental error.

Confidence interval was assumed at 95% ($P < 0.05$) for significant different among the means. Tukey's test was applied as post-hoc test to separate the significantly different means in case of significant interactions. Results were presented as mean ± pooled standard error of the mean.

Results

All the silages were of excellent quality (Table 1). No difference was noted in the quality traits of the silages. There was a significant interaction between HMF and HTF for silage pH ($P < 0.001$) and Flieg point ($P < 0.001$) of ensiled wheat. Application of HTF reduced the pH of wheat silage compared to control group that further declined with the inclusion of HMF + HTF silage additives. An opposite trend was seen for Flieg point of wheat silages. Besides these, nutritional composition (Table 2), feed value, and *in vitro* digestibilities (Table 3) remained unaffected across the groups.

Discussion

Silage quality is dependent on the rapid pH decline, temperature, and other factors related to the packing and plant material intended for ensiling process. In the present study, pH was lower in wheat ensiled with single or combined HMF and HTF silage additives compared to control group. Similarly, Flieg point was greater in wheat silage prepared with HMF and HTF silage additives applied alone or in combination. These findings are consistent with those of Filya (2003b) who reported a decrease in pH of ensiled wheat with HMF and HMF + HTF silage additives. Likewise, Zhang et al. (2009) reported a decrease in the pH of alfalfa silage prepared with single or combined HMF and HTF silage inoculants.

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Table 1. Physical characteristics of wheat ensiled with single or combined homo- and heterofermentative silage additives.

Item	pH	NH_3-N§	Odor	Structure	Color	DLG Score	Flieg point
Homofermentative inoculant							
Not added	4.42	0.137	12.83	4.00	1.92	18.75	113.10
Added	3.99	0.161	12.83	4.00	1.92	18.75	132.06
P-value	<0.001	0.298	0.999	0.999	0.999	0.999	<0.001
Heterofermentative inoculant							
Not added	4.32	0.125	12.67	4.00	1.92	18.58	117.20
Added	4.09	0.172	13.00	4.00	1.92	18.92	126.96
P-value	<0.001	0.239	0.282	0.999	0.999	0.397	<0.001
SEM	0.02	0.07	0.20	0.00	0.08	0.26	0.94
Interaction means							
Control	4.64[a]	0.112	13.00	4.00	2.00	19.00	104.74[c]
HMF[1]	3.99[c]	0.139	12.33	4.00	1.83	18.17	132.06[a]
HTF[2]	4.20[b]	0.162	12.67	4.00	1.83	18.50	121.46[b]
HMF + HTF	3.98[c]	0.183	13.33	4.00	2.00	19.33	132.46[a]
SEM	0.04	0.03	0.29	0.00	0.12	0.37	1.34
HMF × HTF	<0.001	0.158	0.050	0.999	0.195	0.056	<0.001

§ Relative of total nitrogen
[1] HMF = homofermentative
[2] HTF = heterofermentative

However, there was no effect on NH_3-N content of silages in the present study as opposed to Filya (2003b) who reported that the addition of HTF alone or in combination with HMF reduces the NH_3-N of ensiled wheat. Zhang et al. (2009) reported that the addition of HMF and HTF silage additives alone or in combination had no effect on the NH_3-N of alfalfa at d 2, 5, 9, 15, and 30, however, it significantly decreased at d 90 in silage prepared with a combination of HMF and HTF (HMF + HTF). Similarly, HMF or HTF silage inoculants reduced the pH and NH_3-N of potato hash silage (Nkosi et al., 2010). It seems that the increase in Flieg point of ensiled wheat in the HMF, HTF, and HMF + HTF groups was contributed by the pH of the silages since the DM was not different among the groups. The quality of all the silages was categorized as 'excellent' according to the DLG scoring method based on odor, structure, and color of the silage. This might be attributed to a rapid decrease in the pH of wheat silages under the action of HMF and HTF silage additives that helped in the preservation of silages via effective fermentation by producing acetic acid.

In our study, application of silage additives alone or in combination had no effect on the nutritional composition, feed value, and *in vitro* DM and OM digestibility of wheat silages. There are a limited number of studies describing the effect of HMF and HTF silage additives alone or in combination on wheat silage. Most studies have focused on the microbiological quality, fermentation characteristics, and of wheat silages while there is no study describing the nutritional composition and feed value of wheat silages. Consistent with our findings, Filya (2003b) reported that the *in situ* nutrient digestibility of wheat silage prepared with single or combined HMF and HTF silage additives remain unaffected. Similarly, Zhang et al. (2009) reported that the application of HMF and HTF silage inoculants alone or in combination had no effect on *in situ* DM, NDF, and ADF digestibility of alfalfa silages.

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Table 2. Nutrient composition Physical characteristics of wheat ensiled with single or combined homo- and heterofermentative silage additives (%, dry matter basis).

Item	Nutrients[1]											
	DM	CF	EE	CP	Ash	Total CHO	ADFom	ADL	aNDFom	NFC	HEC	CEL
Homofermentative inoculant												
Not added	42.45	23.83	4.26	9.77	7.84	20.58	26.79	4.31	47.05	31.08	20.26	22.48
Added	43.33	23.54	3.84	9.40	7.33	22.76	27.86	4.93	47.35	32.09	19.49	22.92
P-value	0.121	0.702	0.326	0.721	0.182	0.167	0.151	0.156	0.471	0.257	0.281	0.367
Heterofermentative inoculant												
Not added	42.50	23.62	4.24	9.22	7.56	21.48	27.02	4.56	46.59	32.39	19.58	22.46
Added	42.78	23.75	3.85	9.94	7.62	21.37	27.63	4.68	47.79	30.82	20.16	22.95
P-value	0.331	0.831	0.570	0.025	0.686	0.758	0.392	0.765	0.259	0.413	0.639	0.323
SEM	0.25	0.33	0.30	0.25	0.20	0.37	0.48	0.28	0.56	0.48	0.32	0.33
Interaction means												
Control	42.67	23.64	4.64	9.05	7.95	21.03	25.90	4.18	46.38	31.97	20.48	21.72
HMF[2]	43.33	23.60	3.85	9.39	7.16	22.93	28.14	4.94	46.81	32.79	18.67	23.20
HTF[3]	42.23	24.02	3.88	10.48	7.74	20.13	27.68	4.44	47.71	30.20	20.03	23.24
HMF + HTF	43.33	23.48	3.82	9.41	7.50	22.60	27.58	4.93	47.88	31.38	20.30	22.65
SEM	0.63	0.42	0.43	0.35	0.34	0.49	0.67	0.40	0.51	0.64	0.34	0.46
HMF × HTF	0.670	0.892	0.799	0.239	0.392	0.585	0.212	0.744	0.393	0.314	0.418	0.055

[1] DM = dry matter, CF = crude fiber, EE = ether extract, CP = crude protein, Total CHO = total carbohydrates, ADFom = ash-free acid detergent fiber, ADL = acid detergent lignin, aNDFom = ash-free neutral detergent fiber after amylase treatment, NFC = non-fibrous carbohydrates, HEC = hemicellulose, CEL = cellulose
[2] HMF = homofermentative
[3] HTF = heterofermentative

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Table 3. Feed value and *in vitro* digestibility of wheat ensiled with single or combined homo- and heterofermentative silage additives.

Item	Feed Value [1]			IVTDMD[2]		IVTOMD[3]			
	DDM	DMI	RFV	NE_L	TDN	24 h	48 h	24 h	48 h
Homofermentative additive									
Not added	68.03	2.55	134.44	1.60	66.77	60.88	63.91	61.23	65.64
Added	67.20	2.53	131.76	1.57	65.39	57.73	63.14	60.68	63.59
P-value	0.151	0.367	0.605	0.151	0.151	0.137	0.870	0.239	0.192
Heterofermentative additive									
Not added	67.85	2.58	135.66	1.59	66.47	60.22	63.16	61.10	65.83
Added	67.38	2.51	131.07	1.58	65.69	58.39	63.89	60.81	64.89
P-value	0.392	0.397	0.502	0.392	0.392	0.405	0.794	0.341	0.218
SEM	0.37	0.18	1.61	0.01	0.61	0.93	0.42	0.29	0.49
Interaction means									
Control	68.73	2.58	137.43	1.62	67.92	61.25	63.24	61.38	65.40
HMF[4]	66.98	2.56	132.88	1.56	65.03	59.18	63.08	60.82	63.26
HTF[5]	67.34	2.52	131.52	1.58	65.62	60.52	64.59	61.07	65.87
HMF + HTF	67.42	2.51	131.15	1.58	65.75	58.28	63.19	60.54	63.92
SEM	0.52	0.23	4.28	0.02	0.87	1.13	0.57	0.45	0.71
HMF × HTF	0.120	0.318	0.483	0.120	0.120	0.541	0.892	0.477	0.623

[1] DDM = digestible dry matter (%, DM basis), DMI = dry matter intake (% body weight), RFV = relative feed value, NE_L = net energy for lactation (Mcal/kg), TDN = total digestible nutrients (%, DM basis)
[2]IVTDMD = *in vitro* true dry matter digestibility
[3] IVTOMD = *in vitro* true organic matter digestibility
[4] HMF = homofermentative
[5] HTF = heterofermentative

Unlike our findings, the application of HMF or HTF silage additives reduced the DM, aNDFom, and ADF while increasing the CP of potato hash silage. However, the DM and OM digestibilities were not affected by the application of inoculants (Nkosi et al., 2010). Similar findings were reported by Zhang et al. (2021) in response to single or combined HMF and HTF silage additives.

Conclusions

Under the conditions of the present study, it is concluded that the application of homo- and heterofermentative silage inoculants alone or in combination yields well-preserved wheat silage harvested at early dough stage of maturity. Nutritional composition, feed values, and *in vitro* dry matter and organic matter digestibilities may remain unaffected. Further studies involving the *in situ* and *in vivo* nutrient digestibilities may present the true picture on the effect of wheat silage prepared with homo- and heterofermentative silage inoculants alone or in combination.

Conflict of Interest

No commercial funding was acquired for this study that may be construed as a potential conflict of interest.

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Case Report / Olgu Sunumu

The Case of Uterine Prolapse in Golden Retriver Bitch - Vulval SutureTechnique

Golden Retriver Irkı Dişi Köpkte Prolapsus Uteri Olgusu - Vulval Sütür Tekniği

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Abstract: This case report describes the treatment of uterine prolapse in a bitch with a vulval suture technique after the rejection of the uterine. A two-year-old Golden Retriever bitch was brought in with the complaint of protruded uterine after dystocia. The edema in the uterus was removed by applying pressure with the help of a bandage in the direction of blood circulation. The outer part was lubricated with paraffin liquid and put inside. After this process, a vulval suture procedure was applied to the upper third of the vulva. After the treatment, it was observed that the uterine prolapse did not recur and the patient recovered. It was learned that the bitch became pregnant in the next sexual period.

Keywords: Bitch, Dystocia, Uterine prolapse.

Öz: Bu vaka raporu, dişi bir köpekte uterusun reddedilmesinden sonra vulval ligatür tekniği ile prolapsus uterinin tedavisini anlatmaktadır. İki yaşında Golden Retriever ırkı köpek güç doğum sonrası prolapsus uteri şikayeti ile getirildi. Kan dolaşımı yönünde bir sargı bezi yardımıyla basınç uygulanarak uterustaki ödem alındı. Dışarıdaki kısım parafin likit ile kayganlaştırılarak içeriye reddedildi. Bu işlemin ardından vulvanın üst 1/3'lük kısmına vulval ligatür işlemi uygulandı. Tedavi sonrasında prolapsus uterinin tekrarlamadığı ve hastanın iyileştiği görüldü. Bir sonraki seksüel periyotta köpeğin gebe kaldığı öğrenildi.

Anahtar Kelimeler: Dişi köpek, Güç doğum, Prolapsus uteri.

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Introduction

Uterine prolapse is the protrusion of one or both uterine horns together with the corpus uteri, passing through the cervix uteri and out of the vagina (Wood, 1986). This disease, which is frequently observed as a complication of delivery in cows, sheep, and mares, has an incidence of 0.03% in bitches. Uterine inertia, dystocia, and oversized fetus are among the causes of this disease (Jackson, 2004; Miesner et al, 2008). The prognosis of the disease and the severity of the clinical findings generally depend on the time since the prolapse and the preservation of the integrity of the uterine arteries. Therefore, the disease may have no symptoms other than uterine prolapse; severe symptoms such as dehydration, hypothermia, and shock may also be observed. Ovariohysterectomy is usually performed for treatment purposes (Payan-Carreira et al, 2012).

The aim of this case report is to modify the vulva suture technique used in prolapsed vagina and uterine cases in ruminants to bitches and thus preserve fertility without the need for ovariohysterectomy.

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Materials and Methods

A two-year-old, 16 kg Golden Retriever bitch was brought to Burdur Mehmet Akif Ersoy University, Veterinary Faculty, Animal Hospital with the complaint of postpartum uterine prolapse after delivery of a stillborn puppy a day before (Figure 1A). According to the anamnesis, it was learned that the patient whelped for the first time and the delivery took longer than normal. In the physical examination, it was seen that there was no necrotic area in the uterus. Detailed information about the treatment procedure was given to the patient owner and an informed consent form was signed. Rectal body temperature, heart rate, and respiratory rate were within normal values. In the haematologic examination, it was observed that all parameters were within the reference range (Table 1). It was determined that the uterine corpus and right uterine horn were prolobsed. It was observed that the uterine was edematous and congested. There were no puppies in the uterus during the abdominal ultrasound examination.

Table 1. Haematologic and physical examination findings.

Parameter	Measured	Reference Value
WBC ($x10^9$/L)	9.35	6-17
RBC ($x10^{12}$/L)	7.32	5.5-8.5
HGB (g/dL)	16.2	12-18
HCT	50.10	37-55
MCV (fL)	62	60-77
PLT ($x10^9$/L)	425	200-500
Temperature (^{0}C)	38.5	37.5-39
Heart rate (bmp)	110	80-120
Respiratory rate (rpm)	26	20-40

White blood cell (WBC), red blood cell (RBC), hemoglobin (HGB), hematocrit (HCT), mean corpuscular volume (MCV), platelet count (PLT).

Treatment in uterine prolapse cases includes medical and surgical treatment options according to the patient's condition and preference (Hedlund, 2007; Agaoglu et al, 2007). In this case, because the owner did not want to lose its fertility and there was no permanent damage to the uterine, it was preferred to replace it after the edema in the uterine was removed. Anesthesia was induced with propofol (Propofol®, Polifarma, Turkey) at a dose of 5.5 mg/kg. Then, endotracheal intubation and standard anesthesia monitoring (ECG, blood pressure, and saturation) were performed. Anesthesia was maintained with sevoflurane (Sevorane®, Abbvie, USA). Initially, the uterine was cleaned with 10% diluted povidone-iodine and wrapped with a bandage after making sure that there was no other abdominal organ in it. Massage was performed with 20% mannitol (Mannitol, Polifarma, Turkey) to reduce edema (Figure 1B). After the edema was reduced, massage was performed with paraffin liquid, and the uterine was replaced (Figure 1C). A horizontal mattress ligature was applied to the upper third of the vulva by passing absorbable sutures through the infusion set rubber cut about 3 cm in length (Figure 1D). Thus, it was aimed to reduce the suture pressure applied to the vulva. In addition, this method provided less irritation of the vulva.

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Postoperatively, 20 mg/kg amoxicillin-clavulanic acid (Synulox®, Zoetis, Germany) was administered for 5 days and 0.2 mg/kg meloxicam (Meloxicam®, Bavet, Turkey) was administered for 2 days. In the clinical and ultrasound examination performed 1 week after the treatment, it was observed that the uterus was in its normal anatomical position. The suture was removed. No pathological changes were found in the vulva. It was learned that the bitch mated in the next cycle, became pregnant, and whelped healthy puppies.

Figure 1. Uterine prolapse (A), right uterine horn (a), left uterine horn (b), corpus uteri (c), vagina (d). Use of hypertonic solution and pressure to remove uterine edema (B). Lubricating with paraffin liquid (C). Vulval suture procedure to the upper third of the vulva (D).

Discussion

Causes of uterine prolapse include prolonged labor, uterine inertia, dystocia and oversized fetus (Jackson, 2004). In this case, it is thought that the dystocia may have caused the uterine prolapse. Uterine prolapse is a disease that can result in the death of the patient, especially when the integrity of the uterine arteries is disrupted. Therefore, the uterine should be carefully examined for bleeding and rupture. Symptoms such as ischemia, hemorrhage, necrotic areas in the uterine, poor general condition of the patient, and shock are signs of rupture in the uterine arteries (Payan-Carreira et al, 2012). Jadhao et al. (2020) reported that a bitch with uterine prolapse had a general condition failure and some of the haematologic parameters were not at reference values (Jadhao et al, 2020). In this case, the general condition was normal and haematologic parameters were within reference ranges. This situation may be associated with the patient's young age and the fact that a short time has passed since the prolapse. In ruminants, edema due to uterine prolapse is reduced by massaging with hypertonic solutions. Moreover, the ligature applied to the vagina after prolapse vagina rejection is passed through a rubber and soft material so that it does not damage the vagina (Miesner et al, 2008). This technique was created by modifying the vulva suture technique used in prolapse vagina cases in the ruminants to bitches. A similar suture technique was used in a case of prolapsed uteri in a bithc (Jadjo et al (2020). In the presented case, the edema was reduced by massaging the uterine with 20% mannitol hypertonic solution and then it was lubricated with paraffin liquid and rejected. After the uterine was placed, the sutures were passed through the rubber infusion set, which was cut about 3 cm in length, so that the ligature material would not damage the vulva. It was thought that the vulval suture technique would not prevent prolapse uteri in bitches, but it could prevent

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injuries that may occur in the uterus in recurrent cases.

Conclusions

In conclusion, it was revealed that the vulval suture technique is a convenient method in the treatment of uterine prolapse in bitches. However, more cases are needed to determine whether the method is practicable or not.

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Conflict of Interest

There is no conflict of interest.

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