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The Journal of Pediatric Research

JPR

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Dergi Hakkında

The Journal of Pediatric Research, Ege Üniversitesi Tıp Fakültesi Çocuk Sağlığı ve Hastalıkları Anabilim Dalı'nın bir yayın organıdır. Dergi üç ayda bir, Mart, Haziran, Eylül, Aralık aylarında Türkçe ve İngilizce dillerinde yayınlanır.

The Journal of Pediatric Research hakemli, online erişilebilir bir dergidir ve pediatri alanında araştırma makaleleri, derlemeler, klinik çalışmalar ve olgu sunumları yayınlar.

Derginin amacı araştırmacılara ve sağlık alanında çalışanlara çocuk sağlığı ve hastalıklarının tanısı, izlemi, tedavisi, yeni araştırma alanlarının yaratılması ve klinik yaklaşım gibi konularda rehberlik yapmak ve yeni araştırma alanlarının yaratılmasına aracılık etmektir. Derginin web sayfasına http://www.jpedres.org/ yoluyla ulaşabilirsiniz.

Yayın politikası, "ICMJE Recommendations" (2013, http://www.icmje.org/) kuralları temel alınarak belirlenmiştir.

Araştırma bulgularının denetlenmesi istatistik editörü tarafından yapılır.

Dergi içeriği pediatri ile ilgili öncelikle genel pediatri konuları olmakla birlikte, pediatri yan dallarını (Acil Tıp, Allerji ve İmmünoloji, Endokrinoloji, Gastroenteroloji, Beslenme ve Hepatoloji, Genetik, Kardiyoloji, Hematoloji-Onkoloji, Enfeksiyon Hastalıkları, Metabolizma, Nefroloji, Nöroloji, Romatoloji, Solunum Yolları, Sosyal Pediatri, Yenidoğan, Yoğun Bakım, Etik ve Sağlık Servisleri), Çocuk Psikiyatrisi, Çocuk Cerrahisi, Çocukluk Yaş Grubunun Ağız ve Diş Sağlığı ve Çocuk Sağlığı ve Hastalıkları Hemşireliği ile ilgili genel konuları kapsamaktadır.

The Journal of Pediatric Research'e gönderilen çalışmalarınızın ortalama değerlendirme süresi 6 haftadır.

Makalenin kabulünü takiben bütün yazarların makalenin son halini görmesi ve içeriğini onaylaması beklenmektedir. Başka bir yayın organında basılmış ya da değerlendirme aşamasında olan makaleler kesinlikle değerlendirme sürecine giremezler. Araştırma makalelerinde etik onay belgesi (etik onay numarası ile birlikte), olgu sunumlarında bilgilendirilmiş onam formu istenir.

The Journal of Pediatric Research, **Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, Tübitak/ Ulakbim TR Dizini, TurkMedline ve Türkiye Atıf Dizini'nde** indekslenmektedir.

Açık Erişim Politikası

Dergide açık erişim politikası uygulanmaktadır. Açık erişim politikası Budapest Open Access Initiative (BOAI) http://www.budapestopenaccessinitiative.org/ kuralları esas alınarak uygulanmaktadır.

Açık Erişim, "[hakem değerlendirmesinden geçmiş bilimsel literatürün], İnternet aracılığıyla; finansal, yasal ve teknik engeller olmaksızın, serbestçe erişilebilir, okunabilir, indirilebilir, kopyalanabilir, dağıtılabilir, basılabilir, taranabilir, tam metinlere bağlantı verilebilir, dizinlenebilir, yazılıma veri olarak aktarılabilir ve her türlü yasal amaç için kullanılabilir olması"dır. Çoğaltma ve dağıtım üzerindeki tek kısıtlama yetkisi ve bu alandaki tek telif hakkı rolü; kendi çalışmalarının bütünlüğü üzerinde kontrol sahibi olabilmeleri, gerektiği gibi tanınmalarının ve alıntılanmalarının sağlanması için, yazarlara verilmelidir.

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Yazarlara Bilgi

Yazarlara bilgi dergi sayfasında ve www.jpedres.org web sayfasında yayınlanmaktadır.

Materyal Sorumluluk Reddi

The Journal of Pediatric Research, yayınlanan tüm yazılarda görüş ve raporlar yazar(lar)ın görüşüdür ve Editör, Editörler Kurulu ya da yayıncının görüşü değildir; Editör, Editörler Kurulu ve yayıncı bu yazılar için herhangi bir sorumluluk kabul etmemektedir.

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The Journal of Pediatric Research abonelik işlemleri için Galenos Yayınevi ile iletişime geçmeleri gerekmektedir. Ait olduğu dönemler içinde dergi eline ulaşmamış aboneler, Galenos Yayınevi'ne müracaat etmelidirler. Derginin tüm sayılarına ücretsiz olarak www.jpedres.org adresinden tam metin olarak ulaşılabilinir.

Kapak Fotoğrafı

Yazarlar dergimizde yayınlanmak üzere çocuk temalı ve düşündürücü özelliği olan fotoğraflar gönderebilirler. Uygun görülen fotoğraflar ilgili sayıda yayınlanacaktır. Fotoğraflarınızı ozgur.cogulu@ege.edu.tr adresine göndermenizi rica ederiz.

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About Journal

The Journal of Pediatric Research is the official publication of Ege University and Ege Children's Foundation. It is a peer-reviewed journal published quarterly in March, June, September and December in Turkish and English languages.

The Journal of Pediatric Research is a peer-reviewed, open access journal, which publishes original research articles, review articles, clinical reports and case reports in all areas of pediatric research.

The journal publishes original research and review articles, which summarize recent developments about a particular subject based on standards of excellence and expert review and case reports. Target audience includes specialists in general pediatrics and pediatric subspecialties (Emergency Medicine, Allegy and Immunology, Endocrinology, Gastroenterology, Hepatology and Nutrition, Genetics, Cardiology, Hematology-Oncology, Infectious Diseases, Metabolism, Nephrology, Neurology, Rheumatology, Pulmonology, Social Pediatrics, Newborn, Critical Care Medicine, Ethics and Health Service Research), as well as relevant specialties such as Pediatric Surgery, Child and Adolescent Psychiatry, Pedodontics, Pediatric Nursing and Family Physicians. The web page of The Journal of Pediatric Research is http://www.jpedres.org/.

The journal's editorial policies are based on "ICMJE Recommendations" (2013, http://www.icmje.org/) rules.

Statistics Editor evaluates research articles and systematic reviews/meta-analyses for appropriateness of data presentation and correctness of statistical analyses.

All submitted manuscripts are peer reviewed which take on average 6 weeks. Following acceptance of the submission, all authors are required to see and approve the final version of the manuscript and be willing to take responsibility for the entire manuscript. It is strictly expected that submitted manuscripts have not been published elsewhere or even being submitted by another publication. Studies performed on human require ethics committee certificate including approval number. For the manuscripts involving cases, a written informed consent should be obtained from the parents or the responsible persons.

The Journal of Pediatric Research is indexed in **Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, Tübitak/Ulakbim TR Index, TurkMedline and Turkiye Citation Index.**

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This journal provides immediate open access to its content on the principle that making research freely available to the public supports a greater global exchange of knowledge.

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Yazarlara Bilgi

The Journal of Pediatric Research, Ege Üniversitesi Tıp Fakültesi Çocuk Sağlığı ve Hastalıkları Anabilim Dalı ve Ege Sağlık Vakfı'nın yayın organı olup, çocuk sağlığı ve hastalıkları ile doğrudan ya da dolaylı olarak ilgili konularda özgün klinik ve laboratuvar araştırmaları, olgu sunumları, derleme yazıları yayınlar. Derginin yayın dili Türkçe ve İngilizce'dir.

Türkçe yazılarda Türk Dil Kurumu'nun Türkçe Sözlüğü ve Yazım Kılavuzu temel alınmalıdır. Kullanılan terimlerin Türkçe kullanılmasına özen gösterilmelidir.

The Journal of Pediatric Research Dergisi makale başvuru ücreti veya makale işlem ücreti uygulamamaktadır.

The Journal of Pediatric Research'nin kısaltması JPR'dir, ancak kaynaklarda kullanılırken J Pediatr Res şeklinde belirtilmelidir. Uluslararası indekslerde ve veritabanında, derginin adı The Journal of Pediatric Research, İngilizce kısaltması J Pediatr Res olarak kaydedilmiştir.

Dergiye kabul edilen eserlerin özgün ve daha önceden başka ortamlarda yayınlanmamış olması esas alınır. Yayın için dergiye yollanan her yazı hakem değerlendirmesine gönderilir. Yazarlar 6 hafta içinde süreçle ilgili haberdar edilir. Değerlendirme sonucunda basılması kabul edilen yazılar dergide basılır ve dergi web sayfası olan http://www.jpedres.org adresinde yayınlanır.

Yazıların bilimsel ve etik sorumlulukları yazarlara, telif hakkı ise JPR'ye aittir. Yazıların içeriğinden ve kaynakların doğruluğundan yazarlar sorumludur. Yazarlar, yayın haklarının devredildiğini belirten onay belgesini (Yayın Hakları Devir Formu) yazıları ile birlikte göndermelidirler. Bu belgenin tüm yazarlar tarafından imzalanarak dergiye gönderilmesi ile birlikte yazarlar, gönderdikleri çalışmanın başka bir dergide yayınlanmadığı ve/veya yayınlanmak üzere incelemede olmadığı konusunda garanti vermiş, bilimsel katkı ve sorumluluklarını beyan etmiş sayılırlar.

Dergiye yayımlanmak üzere gönderilen tüm yazılar 'iThenticate' programı ile taranarak intihal kontrolünden geçmektedir. İntihal taraması sonucuna göre yazılar red ya da iade edilebilir.

The Journal of Pediatric Research'te yayınlanmak amacıyla gönderilen ve etik kurul onayı alınması zorunluluğu olan deneysel, klinik ve ilaç araştırmaları için uluslararası anlaşmalara ve 2013'de gözden geçirilmiş Helsinki Bildirisi'ne uygun etik kurul onay raporu gereklidir (http://www.wma.net/en/30publications/10policies/b3/). Etik kurul onayı ve "bilgilendirilmiş gönüllü onam formu" alındığı araştırmanın "Gereç ve Yöntem" bölümünde belirtilmelidir. Deneysel hayvan çalışmalarında ise yazarlar, "Guide for the care and use of laboratory animals" (http://oacu.od.nih.gov/regs/guide/guide.pdf) doğrultusunda hayvan haklarını koruduklarını belirtmeli ve kurumlarından etik kurul onay raporu almalıdır.

Yayın, direkt ya da indirekt ticari bağlantı içeriyorsa veya çalışmaya materyal desteği veren bir kuruluş varsa, yazarlar kullanılan ticari ürün, ilaç, firma vs. ile ticari hiçbir ilişkisinin olmadığını ya da var ise nasıl bir ilişkisinin olduğunu (konsültan, diğer anlaşmalar), editöre sunum sayfasında belirtmek zorundadır.

Araştırmalara yapılan her türlü yardım ve diğer desteklerin alındığı kişi ve kuruluşlar beyan edilmeli ve çıkar çatışmasıyla ilgili durumları açıklamak amacıyla Çıkar Çatışmaları Bildirim Formu doldurulmalıdır.

Tüm yazılar, editör ve editör yardımcıları ile danışman hakemler tarafından incelenir.

The Journal of Pediatric Research bağımsız, önyargısız ve çift-kör hakemlik ilkeleri çerçevesinde yayın yapan süreli bir yayın organıdır. Hakemler, yazının konusuyla ilgili uluslararası literatürde yayınları ve atıfları olan bağımsız uzmanlar arasından seçilmektedir. Makale baş editöre ulaşınca değerlendirmeye alınır ve bölüm editörüne gönderilir. Bölüm editörü ilk değerlendirmeyi takiben makaleyi hakemlere gönderir. Hakemler 21 gün içinde kararlarını bildirmelidirler. Bölüm editörü hakem kararlarına kendi değerlendirme ve önerisini ekleyerek baş editöre gönderir ve son kararı hakemlerin görüşleri doğrultusunda bölüm editörü verir. Hakemlerin kararları birbirleriyle çelişkili ise dergi editörü gerektiğinde yeni hakem atayabilir.

Dergide yayınlanacak yazıları değerlendiren hakemler dergide belirtilen danışmanlar ve gerekirse yurt içi/dışı konu ile ilgili uzmanlar arasından seçilir. Yazarlar, yayına kabul edilen yazılarda, metinde temel değişiklik yapmamak kaydı ile editör, editör yardımcıları, biyoistatistik uzmanı ve İngilizce dil uzmanının düzeltme yapmalarını kabul etmiş sayılır.

İncelemeye sunulan araştırmada olası bir bilimsel hata, etik ihlal şüphesi veya iddiasıyla karşılaşılırsa, bu dergi verilen yazıyı destek kuruluşların veya diğer yetkililerin soruşturmasına sunma hakkını saklı tutar. Bu dergi sorunun düzgün biçimde takip edilmesi sorumluluğunu kabul eder ancak gerçek soruşturmayı veya hatalar hakkında karar verme yetkisini üstlenmez.

Yayın Politikası ve Makale Yazım Kuralları aşağıda belirtilen maddeler "Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals (ICMJE Recommendations)" (2013, http://www.icmje.org/) temel alınarak hazırlanmıştır.

Araştırma makalelerinin hazırlığı, sistematik derleme, meta-analizleri ve sunumu ise uluslararası kılavuzlara uygun olmalıdır:

Araştırma makalelerinin hazırlığında sistematik derlemeler ve meta analizler için aşağıdaki tasarım klavuzları:Randomize çalışmalar için; CONSORT (Moher D, Schultz KF, Altman D, for the CONSORT Group. The CONSORT statement revised recommendations for improving the quality of reports of parallel group randomized trials. JAMA 2001; 285:1987-91) (http://www.consort-statement.org/).

Sistematik derleme ve meta-analizlerin raporlamaları için; PRISMA (Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group. Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 2009; 6(7): e1000097) (http://www.prisma-statement.org/).

Tanısal değerli çalışmalar için; STARD (Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig LM, et al, for the STARD Group. Towards complete and accurate reporting of studies of diagnostic accuracy: the STARD initiative. Ann Intern Med 2003;138:40-4) (http://www.stard-statement.org/).

Gözlemsel çalışmalar için; STROBE (http://www.strobe-statement.org/).

Meta-analizleri ve gözlemsel çalışmaların sistematik derlemeleri için; MOOSE (Stroup DF, Berlin JA, Morton SC, et al. Meta-analysis of observational studies in epidemiology: a proposal for reporting "Meta-analysis of observational Studies in Epidemiology" (MOOSE) group. JAMA 2000; 283: 2008-12).

GENEL KURALLAR

Yazılar sadece çevrim-içi olarak kabul edilmektedir. Yazarların makale gönderebilmesi için Journal Agent web sayfasına (http://www.journalagent.com/jpr/) kayıt olup, şifre almaları gerekmektedir. Bu sistem

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çevrim-içi yazı gönderilmesine ve değerlendirilmesine olanak tanımaktadır. Bu sistem ile toplanan makaleler Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, Tübitak/Ulakbim TR Dizini, TurkMedline ve Türkiye Atıf Dizini kurallarına uygun olarak sisteme alınmakta ve arşivlenmektedir.

Sayfa düzeni: Özgün Araştırmalar, klinik araştırma, klinik gözlem, yeni teknikler, deneysel ve laboratuvar çalışmalarını kapsar. Özgün araştırmalar, başlık, özet, yazının ana konusu ile ilgili anahtar kelimeler, giriş, gereç ve yöntem, bulgular, tartışma, teşekkür, kaynaklar, tablolar, resimler bölümlerini içermelidir. Özet bölümü, "Öz" başlığı ile yazılmalıdır. Metin "Times New Roman" yazı stili, 12 punto, 1,5 satır aralığı ile yazılmalıdır. Metnin tümü 2500 kelimeyi geçmemelidir. Olgu Sunumları, nadir görülen ya da tanı ve tedavide farklılık gösteren, mevcut bilgilerimize katkıda bulunan, eğitici olguyu/olguları içermeli, giriş, olgu sunumu, tartışma bölümlerini içermelidir. Metnin tümü 1500 kelimeyi geçmemelidir. Derlemeler güncel bir konuyu, bağımsız, literature bilgisini de içerecek şekilde derinlemesine inceleyen yazılarıdır. Metnin tümü 18 adet A4 sayfasını geçmemelidir. Editöre Mektuplar yayınlanmış makaleler hakkında ya da güncel pediatrideki gelişmeleri içeren 1000 kelimeyi geçmeyen ve kaynak belirten yazılar olmalıdır. Özet bölümlerini içermez. Kaynak sayısı 5 ile sınırlıdır.

Kısaltmalar: Kelimenin ilk geçtiği yerde parantez içinde ve tüm metin boyunca kullanılır. Uluslararası kullanılan kısaltmalar için "Bilimsel Yazım Kuralları" kaynağına başvurulabilir.

Editöre sunum sayfası: Gönderilen makalenin kategorisi, daha önce başka bir dergiye gönderilmemiş olduğu, var ise çalışmayı maddi olarak destekleyen kişi ve kuruluşlar ve bu kuruluşların yazarlarla ilişkileri, makale İngilizce ise; İngilizce yönünden kontrolünün ve araştırma makalesi ise biyoistatistiksel kontrolünün yapıldığı belirtilmelidir.

KAYNAKLAR

Kaynakların gerçekliğinden yazarlar sorumludur.

Metin içinde: Kullanılan kaynaklar, ilgili cümlenin içinde veya sonunda parantez içinde belirtilmelidir. Eğer, kullanılan kaynağın yazar/yazarları cümle başında belirtiliyorsa, kaynak isimden hemen sonra gelecek şekilde parantez içi olarak yazılmalıdır. Tüm yazılarda Türk yazarlarca yapılmış yerli veya yabancı yayınların kullanılmasına özellikle dikkat edilmeli ve Türkçe dil kurallarına uyulmasına özen gösterilmelidir.

Kongrelerde sunulan bildiriler, basılmamış yayınlar, tezler, internet kaynaklı adresler, kişisel görüşme ya da deneyimler kaynak olarak belirtilmemelidir. Adı geçen kaynaklardan bahsedilmek isteniyorsa, yazıda geçtiği cümlenin sonunda kaynak numarası belirtilmeden, açık yazı ile parantez içine alınarak kaynağın niteliği belirtilmelidir.

Kaynaklar bölümünde: Kaynaklar metin içerisinde geçiş sırasına göre Arap rakamları ile numaralandırılmalıdır. Kaynaklarda tüm yazarlar belirtilmelidir. Ancak yazar sayısı 6'dan fazla olan çalışmalarda ilk 3 yazarın adı yazılmalı, daha sonra Türkçe makalede (ve ark.), İngilizce makalede (et al.) eki yapılmalıdır. Dergilerin isimleri Index Medicus'ta kullanılan stillere göre kısaltılmalıdır.

Kaynak yazılımı için örnekler:

Dergi: Yazar(lar)ın soy isim(ler)i ve yazar isim(ler)inin ilk harfi, makale başlığı, dergi adı (dergide belirtilen orijinal kısaltması), yıl, cilt ve sayfa numaraları.

Örnek: Koening JQ. Air pollution and asthma. J Allergy Clin Immunol 1999;104:717-22.

Kitap: Yazar(lar)ın soy isim(ler)i ve yazar isim(ler)inin ilk harfi, bölüm başlığı, editörün(lerin) ismi, kitap ismi, kaçıncı baskı olduğu, basıldığı şehir, basım yeri, yayınevi, basım yılı ve sayfa numaraları.

Örnek: Fletcher CDM, Unni KK, Mertens F. Genetics of Tumours of Soft Tissue and Bone. Lyon, France, IARC Press, 2002. p. 225-419.

Kitap bölümü: Yazar(lar)ın soy isim(ler)i ve yazar isim(ler)inin ilk harfi, bölüm ve kısım, editörün(lerin) ismi, kitap adı, basım yeri, yayınevi adı, basım yılı, sayfa numaraları.

Örnek: Whitsett JA, Pryhuber GS, Rice WR. Acute respiratory disorders. In: Avery GB, Mac- Donald MG (eds). Neonatology: Pathophysiology and Management of the Newborn, 5th ed. Philadelphia, Lippincott Williams&Wilkins, 1999;505-15.

RESİM, TABLO, GRAFİK VE ŞEKİLLER

Tüm görsel materyaller metnin sonunda ayrı birer sayfa olarak hazırlanmalıdır. Şekil, resim, tablo ve grafiklerin açıklamaları makale sonuna eklenmelidir. Orijinal filmler, EKG kayıtları gibi belgeler kesinlikle yollanmamalıdır. Renkli resimlerin masrafları yazarlar tarafından bizzat karşılanacaktır.

Resimler: Resimlere metindeki geçiş sırasına göre numara verilmeli ve kısa birer başlık yazılmalıdır. Başka bir yayından alıntı yapılıyorsa yazılı baskı izni birlikte yollanmalıdır. Fotoğrafların ayrıntıları seçilmeli, JPEG formatında ve en az 300 dpi (çözünürlük) olarak kaydedilmelidir.

Tablolar, Grafikler, Şekiller: Tüm tablolara, grafiklere ve şekillere metinde geçiş sırasına göre numara verilmeli ve kısa birer başlık yazılmalıdır. Tablolar yazıda geçiş sıralamasına göre Romen rakamlarıyla (I, II) sıralandırılmalı ve başlık taşımalıdır. Şekiller geçiş sıralamasına göre Arap harfleri (1,2) ile sıralanmalıdır. Kullanılan kısaltmalar alt kısımda mutlaka açıklanmalıdır. Özellikle tablolar metni açıklayıcı ve kolay anlaşılır hale getirme amacı ile hazırlanmalı ve metnin tekrarı olmamalıdır. Olgu sunumlarında en çok 2 şekil veya resim kullanılmalıdır.

BİYOİSTATİSTİK

Araştırma bulgularının denetlenebilirliğini sağlamak için, araştırma düzeni, örneklem, yöntem, bilimsel yaklaşımlar ve uygulamalar tanımlanarak kaynakları sunulmalıdır.

Anlamlılık sınırı olarak seçilen "p" değeri ile birlikte uygun hata ve belirsizlik payları (güven aralıkları, vs) belirtilmelidir. Kullanılan istatistiksel terimler, kısaltmalar ve semboller tanımlanmalı, kullanılan yazılım (software) belirtilmelidir. İstatistik terminolojisi (random, signifikant, korelasyon, vs.) istatistik dışı anlamlarda kullanılmamalıdır.

Verilerin ve analizin tüm sonuçları tablo, şekil veya grafik olarak "Bulgular" bölümünde, kullanılan biyoistatistiksel yöntemler ve uygulama ayrıntıları yazının "Gereç ve Yöntem" bölümünde veya ayrı bir başlık altında sunulmalıdır.

YAZI ÇEŞİTLERİ

Özgün Araştırmalar

Klinik araştırma, klinik gözlem, yeni teknikler, deneysel ve laboratuvar çalışmalarını kapsar. Özgün araştırmalar; başlık, özet, yazının ana konusu

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Yazarlara Bilgi

ile ilgili anahtar kelimeler, giriş, gereç ve yöntem, bulgular, tartışma, çalışmanın kısıtlılıkları, sonuç, teşekkür, kaynaklar, tablolar, grafikler, resimler bölümlerini içermelidir. Başlık, özet ve anahtar kelimeler Türkçe ve İngilizce olarak yazılmalıdır. Makale yukarıda belirtilen yazım kuralları ile yazılı 16 A4 sayfasını aşmamalıdır.

Başlık sayfası: Makalenin başlığı, kısa başlık, yazar isimleri ve yazar bilgilerini kapsayan sayfadır. Sırasıyla şu tanımlar yapılmalıdır;

1. Makalenin başlığı (Türkçe ve İngilizce) mümkün olduğunca kısa ve açıklayıcı olmalı, boşluklar dahil 135 karakteri geçmemeli, kısaltma içermemelidir.

2. Kısa başlık (Türkçe ve İngilizce), en fazla 60 karakterden oluşmalıdır.

3. Yazar isimleri (yazarların isimleri tam olarak kısaltılmadan yazılmalıdır, yazarın akademik görevi yazılmamalıdır) ve bağlı bulunduğu kurumlar.

4. İletişim kurulacak yazarın ismi, adresi, telefon ve faks numarası ile e-posta bilgileri.

5. Bilimsel toplantılarda sunulan ve özeti kongre kitabında yer almış eserlerin toplantı yeri ve tarihi.

Öz: Yazının ana hatlarını içeren, en fazla 200 kelimeden oluşan öz Türkçe ve İngilizce olarak hazırlanmalıdır. Öz bölümünde kaynak gösterilmemeli, kısaltmalardan mümkün olduğunca kaçınılmalıdır. Yapılacak kısaltmalar metindekilerden bağımsız olarak ele alınmalıdır.

Araştırma makalelerinde öz 5 alt başlık olarak hazırlanmalıdır:

Amaç: Çalışmanın amacı açıkça belirtilmelidir.

Gereç ve Yöntemler: Çalışma tanımlanmalı, standart kriterleri, randomize olup olmadığı, retrospektif veya prospektif olduğu ve varsa istatistiksel yöntem belirtilmelidir.

Bulgular: Çalışmanın detaylı sonucu verilmeli, istatistik anlamlılık derecesi belirtilmelidir.

Sonuç: Çalışmanın sonuçlarını yansıtmalı, klinik uygulanabilirliği tanımlamalı, olumlu ve olumsuz yönleri gösterilmelidir.

Anahtar Kelimeler: En az 3, en çok 5 anahtar kelime özetin sonunda yer almalıdır. İngilizce anahtar kelimeler "Medical Subject Headings'e (MESH) uygun olarak verilmelidir (www.nlm.nih.gov/mesh/MBrowser.html). Türkçe anahtar kelimeler ise MESH terimlerinin aynen çevirisi olmalıdır. Anahtar kelimeler uygun nitelikte ve standart terminolojide yazılmalıdır. Türkçe anahtar kelimeler "Türkiye Bilim Terimleri" arasından seçilmelidir. Yazarlar bilgilendirme için http://www.bilimterimleri.com adresini kullanabilir.

Araştırma makalelerinde ana metin aşağıdaki başlıkları içermelidir;

Giriş: Konu hakkında kısa ve öz bilgi verilmeli, çalışmanın amacı belirtilmeli, bunlar literatür bilgisi ile desteklenmelidir.

Gereç ve Yöntem: Çalışma planı verilmeli, randomize olup olmadığı, retrospektif veya prospektif olduğu, denek sayısı, özellikleri, çalışmaya dahil edilme ve dışlanma kriterleri, kullanılan istatistiksel yöntem belirtilmelidir.

Bulgular: Elde edilen sonuçlar belirtilmeli, tablo ve resimler numara sırasıyla verilmeli, sonuçlar uygulanan istatistiki analiz yöntemine göre değerlendirilmelidir. Görsel materyallerin yazım kuralları hakkında gerekli bilgi "Genel Kurallar" başlığı altında bulunan "Resim, Tablo, Grafik ve Şekiller" bölümünde bulunmaktadır.

Tartışma: Elde edilen değerler olumlu ve olumsuz yönleriyle tartışılmalı, literatür ile karşılaştırılmalı, çalışmadan elde edilen sonuç vurgulanmalıdır.

Sonuç: Çalışmadan elde edilen sonuç vurgulanmalıdır.

Teşekkür: Her türlü çıkar çatışması, finansal destek, bağış ve diğer editöryal (istatistik analiz, İngilizce/Türkçe değerlendirme) ve/veya teknik yardım var ise metnin sonunda sunulmalıdır.

Kaynaklar: Kaynakların gerçekliğinden yazarlar sorumludur. Kaynakların yazım kuralları hakkında gerekli bilgi "Genel Kurallar" başlığı altında bulunan "Kaynaklar" bölümünde bulunmaktadır.

Olgu Sunumları

Nadir görülen, tanı ve tedavide farklılık gösteren, mevcut bilgilerimize yenilerini ekleyip, katkı sağlayan olguları içermelidir. Türkçe ve İngilizce başlık, 50 kelimeyi aşmayan, yapılandırılmamış özet ve anahtar kelimeler ilk sayfada yer almalıdır. Sunum metni, giriş, olgu sunumu, tartışma ve kaynaklardan oluşmalıdır. Metnin tümü yukarıda bahsedilen yazım kuralları çerçevesinde 1500 kelimeyi geçmemelidir. Olgu sunumları için en fazla 10 kaynak kullanılmalıdır.

Derlemeler

Bir bilgi ya da konunun klinikte kullanılması için vardığı son düzeyi anlatan, tartışan, değerlendiren ve gelecekte yapılacak olan çalışmalara yön veren bir formatta hazırlanmalıdır. **Dergi yalnızca davetli derleme kabul eder ve yayınlar.** Derleme başvurusu yapılmadan önce konunun editör ile görüşülmesi önerilir.

Güncel bir konuyu, bağımsız, hiçbir farklı görüşü öne çıkarmadan derinlemesine inceleyen yazılardır. Yazının ilk bölümünde Türkçe ve İngilizce başlık, özet, anahtar kelimeler bulunmalıdır. Özet bölümü yapılandırılmamalıdır. Konuda geçen her alıntının kaynağı mutlak gösterilmelidir. Tümü 18 A4 sayfayı geçmemelidir.

Editöre Mektuplar

Yayınlanmış makaleler hakkında ya da güncel pediatrideki, bilimsel ve sosyal yönden irdeleyen veya özellikle The Journal of Pediatric Research Dergisi'nde yayınlanan yazılarla ilgili soru veya katkı içerikli 1000 kelimeyi geçmeyen ve kaynak belirten yazılar olmalıdır. Başlık ve özet bölümleri içermez. Kaynak sayısı 5 ile sınırlıdır.

REKLAMLAR

Reklamlar için Editör ile görüşülmelidir. Derginin dış ve iç kapakları ile iç sayfalarında renkli reklam yayınlanması mümkündür.

YAYIN HAKKI

The Journal of Pediatric Research, Ege Çocuk Vakfı (EÇV) tarafından desteklenen, Ege Üniversitesi Tıp Fakültesi, Çocuk Sağlığı ve Hastalıkları Anabilim Dalı'nın yayın organıdır.

YAZIŞMA

Prof. Dr. Özgür Çoğulu

The Journal of Pediatric Research

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Intructions to Authors

The Journal of Pediatric Research is an official peer-reviewed publication of the Ege University, Faculty of Medicine, Department of Pediatrics and Ege Children's Foundation. This publication organ is responsible for the issue of unique clinical and laboratory research papers, case reports, reviews directly or indirectly related to children's health and diseases. The publication languages of the journal are Turkish and English.

Manuscripts written in Turkish should be in accordance with the Turkish Dictionary and Writing Guide ("Türkçe Sözlüğü ve Yazım Kılavuzu") of the Turkish Language Association.

The Journal of Pediatric Research does not charge any article submission or processing charges.

The abbreviation of The Journal of Pediatric Research is JPR, however, it should be denoted as J Pediatr Res when referenced. In the international index and database, the name of the journal has been registered as The Journal of Pediatric Research and abbreviated as J Pediatr Res.

A manuscript will be considered only with the understanding that it is an original contribution that has not been published elsewhere. All manuscripts submitted to the journal for publication are peer-reviewed. Authors shall be informed within a period of 6 weeks about the process. Upon review, those manuscripts, which are accepted, shall be published in the journal and issued on the http://www.jpedres.org official internet address.

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All manuscripts submitted to the The Journal of Pediatric Research are screened for plagiarism using the 'iThenticate' software. Results indicating plagiarism may result in manuscripts being returned or rejected.

Experimental, clinical and drug studies requiring approval by an ethics committee must be submitted to The Journal of Pediatric Research with an ethics committee approval report confirming that the study was conducted in accordance with international agreements and the Declaration of Helsinki (revised 2013) (http://www.wma.net/en/30publications/10policies/b3/). The approval of the ethics committee and the fact that informed consent was given by the patients should be indicated in the Materials and Methods section. In experimental animal studies, the authors should indicate that the procedures followed were in accordance with animal rights as per the Guide for the Care and Use of Laboratory Animals (http://oacu.od.nih.gov/regs/guide/guide.pdf) and they should obtain animal ethics committee approval.

Authors must provide disclosure/acknowledgment of financial or material support, if any was received, for the current study.

If the article includes any direct or indirect commercial links or if any institution provided material support to the study, authors must state in the cover letter that they have no relationship with the commercial product, drug, pharmaceutical company, etc. concerned; or specify the type of relationship (consultant, other agreements), if any.

Authors must provide a statement on the absence of conflicts of interest among the authors and provide authorship contributions.

The Journal of Pediatric Research is an independent international journal based on double-blind peer-review principles. The manuscript is assigned to the Editor-in-Chief, who reviews the manuscript and makes an initial decision based on manuscript quality and editorial priorities. Manuscripts that pass initial evaluation are sent for external peer review, and the Editor-in-Chief assigns an Associate Editor. The Associate Editor sends the manuscript to reviewers (internal and/or external reviewers). The reviewers must review the manuscript within 21 days. The Associate Editor recommends a decision based on the reviewers' recommendations and returns the manuscript to the Editor-in-Chief. The Editor-in-Chief makes a final decision based on editorial priorities, manuscript quality, and reviewer recommendations. If there are any conflicting recommendations from reviewers, the Editor-in-Chief can assign a new reviewer.

The scientific board guiding the selection of the papers to be published in the Journal consists of elected experts of the Journal and if necessary, selected from national and international authorities. The Editor-in-Chief, Associate Editors, biostatistics expert and English language consultant may make minor corrections to accepted manuscripts that do not change the main text of the paper.

In case of any suspicion or claim regarding scientific shortcomings or ethical infringement, the Journal reserves the right to submit the manuscript to the supporting institutions or other authorities for investigation. The Journal accepts the responsibility of initiating action but does not undertake any responsibility for an actual investigation or any power of decision.

The Editorial Policies and General Guidelines for manuscript preparation specified below are based on "Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals (ICMJE Recommendations)" by the International Committee of Medical Journal Editors (2013, archived at http://www.icmje.org/).

Preparation of research articles, systematic reviews and meta-analyses must comply with study design guidelines:

CONSORT statement for randomized controlled trials (Moher D, Schultz KF, Altman D, for the CONSORT Group. The CONSORT statement revised recommendations for improving the quality of reports of parallel group randomized trials. JAMA 2001; 285:1987-91) (http://www.consort-statement.org/);

PRISMA statement of preferred reporting items for systematic reviews and meta-analyses (Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group. Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 2009; 6(7): e1000097.) (http://www.prisma-statement.org/);

STARD checklist for the reporting of studies of diagnostic accuracy (Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig LM, et al., for the STARD Group. Towards complete and accurate reporting of studies of diagnostic accuracy: the STARD initiative. Ann Intern Med 2003;138:40-4.) (http://www.stard-statement.org/);

STROBE statement, a checklist of items that should be included in reports of observational studies (http://www.strobe-statement.org/);

MOOSE guidelines for meta-analysis and systemic reviews of observational studies (Stroup DF, Berlin JA, Morton SC, et al. Meta-analysis of observational studies in epidemiology: a proposal for reporting Meta-analysis of observational Studies in Epidemiology (MOOSE) group. JAMA 2000; 283: 2008-12).

GENERAL GUIDELINES

Manuscripts can only be submitted electronically through the Journal Agent website (http://journalagent.com/tjo/) after creating an account. This system allows online submission and review.

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The manuscripts are archived according to Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, Tübitak/Ulakbim TR Index, TurkMedline and Turkiye Citation Index.

Format: Unique research papers cover clinical research, observational studies, novel techniques, and experimental and laboratory studies. Unique research papers shall consist of a heading, abstract, keywords related to the main topic of the paper, introduction, materials and methods, results, discussion, acknowledgements, bibliography, tables and figures. Manuscripts should be prepared using 12 pt "Times New Roman" and 1.5 line spacing. The text shall not exceed 2500 words. Case reports must contain rare cases or those that are unique in diagnosis and treatment, those which contribute to current knowledge and provide educational information, along with the introduction, case reporting and discussion sections. The whole text must not exceed 1500 words. Reviews are texts in which a current subject is examined independently, with reference to scientific literature. The whole text must not exceed 18 A4 paper sheets. Letters to the Editor must be manuscripts, which do not exceed 1000 words, with reference to scientific literature, and those written in response to issued literature or those, which include development in the field of pediatrics. These manuscripts do not contain an abstract. The number of references is limited to 5.

Abbreviations: Abbreviations should be defined at first mention and used consistently thereafter. Internationally accepted abbreviations should be used; refer to scientific writing guides as necessary.

Cover letter: The cover letter should include statements about manuscript type, single-journal submission affirmation, conflict of interest statement, sources of outside funding, equipment (if applicable), approval of language for articles in English and approval of statistical analysis for original research articles.

REFERENCES

Authors are solely responsible for the accuracy of all references.

In-text citations: References shall be listed as the below formats on a separate page according to their sequence within the text and referred to within the text in parentheses.

Presentations presented in congresses, unpublished manuscripts, theses, Internet addresses, and personal interviews or experiences should not be indicated as references. If such references are used, they should be indicated in parentheses at the end of the relevant sentence in the text, without reference number and written in full, in order to clarify their nature. In all manuscripts, care must be given to the inclusion of references related to domestic and international studies performed by Turkish authors and compliance to Turkish written language rules.

References section: All author names shall be stated within all references. References shall be listed as the below formats on a separate page according to their sequence within the text and referred to within the text in parentheses. However, in studies where author numbers exceed 6, names of the first 3 authors shall be stated; "ve ark." and "et al." additions shall be made to the list of authors in Turkish and English references, respectively. The titles of journals should be abbreviated according to the style used in the Index Medicus.

Reference Format

Journal: Last name(s) of the author(s) and initials, article title, publication title and its original abbreviation, publication date, volume, the inclusive page numbers.

Example: Koening JQ. Airpollutionandasthma. J Allergy Clin Immunol 1999; 104:717-22.

Book: Last name(s) of the author(s) and initials, chapter title, book editors, book title, edition, place of publication, date of publication and inclusive page numbers of the extract cited.

Example: Fletcher CDM, Unni KK, Mertens F. Genetics of Tumours of Soft Tissue and Bone. Lyon, France, IARC Press, 2002. p. 225-419.

Book Chapter: Last name(s) of the author(s) and initials, chapter title, book editors, book title, edition, place of publication, date of publication and inclusive page numbers of the cited piece.

Example: Whitsett JA, Pryhuber GS, Rice WR. Acuteres piratory disorders. In: Avery GB, Mac- Donald MG (eds). Neonatology: Pathophysiology and Management of the Newborn, 5th ed. Philadelphia, Lippincott Williams & Wilkins, 1999;505-15.

TABLES, GRAPHICS, FIGURES, AND IMAGES

All visual materials together with their legends should be located on separate pages that follow the main text. Original documents such as films, ECG records must not be delivered. All cost related to colored printouts shall be covered by the authors' own expenses.

Images: Images (pictures) should be numbered and include a brief title. Permission to reproduce pictures that were published elsewhere must be included. All pictures should be of the highest quality possible, in JPEG format, and at a minimum resolution of 300 dpi.

Tables, Graphics, Figures: All tables, graphics or figures should be enumerated according to their sequence within the text and a brief descriptive caption should be written. Tables shall be numbered by Roman numerals (I, II) according to their sequence, and shall include a heading. Figures shall be numbered by Arabic numerals (1,2) according to their sequence. Any abbreviations used should be defined in the accompanying legend. Tables in particular should be explanatory and facilitate readers' understanding of the manuscript, and should not repeat data presented in the main text. A maximum of 2 figures or photographs shall be added to case reports.

BIOSTATISTICS

To ensure controllability of the research findings, the study design, study sample, and the methodological approaches and applications should be explained and their sources should be presented.

The "p" value defined as the limit of significance along with appropriate indicators of measurement error and uncertainty (confidence interval, etc.) should be specified. Statistical terms, abbreviations and symbols used in the article should be described and the software used should be defined. Statistical terminology (random, significant, correlation, etc.) should not be used in non-statistical contexts.

All results of data and analysis should be presented in the Results section as tables, figures and graphics; biostatistical methods used and application details should be presented in the Materials and Methods section or under a separate title.

MANUSCRIPT TYPES

Original Articles

Unique research papers cover clinical research, observational studies, novel techniques, and experimental and laboratory studies. Unique research papers shall consist of a heading, abstract, keywords related to the main topic of the paper, introduction, materials and methods, results, discussion, acknowledgements, bibliography, tables and figures. The text shall not exceed 2500 words. Case reports must contain rare cases or those that are unique in diagnosis and treatment, those which contribute to current knowledge and

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provide educational information, along with the introduction, case reporting and discussion sections. The whole text must not exceed 1500 words. Reviews are texts in which a current subject is examined independently, with reference to scientific literature. The whole text must not exceed 18 A4 paper sheets. Letters to the Editor must be manuscripts, which do not exceed 1000 words, with reference to scientific literature, and those written in response to issued literature or those, which include development in the field of pediatrics. These manuscripts do not contain an abstract. The number of references is limited to 5.

Title Page: This page should include the title of the manuscript, short title, name(s) of the authors and author information. The following descriptions should be stated in the given order:

1. Title of the manuscript (Turkish and English), as concise and explanatory as possible, including no abbreviations, up to 135 characters

2. Short title (Turkish and English), up to 60 characters

3. Name(s) and surname(s) of the author(s) (without abbreviations and academic titles) and affiliations

4. Name, address, e-mail, phone and fax number of the corresponding author

5. The place and date of scientific meeting in which the manuscript was presented and its abstract published in the abstract book, if applicable

Abstract: A summary of the manuscript should be written in both Turkish and English. References should not be cited in the abstract. Use of abbreviations should be avoided as much as possible; if any abbreviations are used, they must be taken into consideration independently of the abbreviations used in the text.

For original articles, the structured abstract should include the following sub-headings:

Aim: The aim of the study should be clearly stated.

Materials and Methods: The study and standard criteria used should be defined; it should also be indicated whether the study is randomized or not, whether it is retrospective or prospective, and the statistical methods applied should be indicated, if applicable.

Results: The detailed results of the study should be given and the statistical significance level should be indicated.

Conclusion: Should summarize the results of the study, the clinical applicability of the results should be defined, and the favorable and unfavorable aspects should be declared.

Keywords: A list of minimum 3, but no more than 5 key words must follow the abstract. Key words in English should be consistent with "Medical Subject Headings (MESH)" (www.nlm.nih.gov/mesh/MBrowser.html). Turkish key words should be direct translations of the terms in MESH.

Original research articles should have the following sections:

Introduction: Should consist of a brief explanation of the topic and indicate the objective of the study, supported by information from the literature.

Materials and Methods: The study plan should be clearly described, indicating whether the study is randomized or not, whether it is retrospective or prospective, the number of trials, the characteristics, and the statistical methods used.

Results: The results of the study should be stated, with tables/figures given in numerical order; the results should be evaluated according to the statistical analysis methods applied. See General Guidelines for details about the preparation of visual material.

Discussion: The study results should be discussed in terms of their favorable and unfavorable aspects and they should be compared with the literature. The conclusion of the study should be highlighted.

Study Limitations: Limitations of the study should be discussed. In addition, an evaluation of the implications of the obtained findings/results for future research should be outlined.

Conclusion: The conclusion of the study should be highlighted.

Acknowledgements: Any technical or financial support or editorial contributions (statistical analysis, English/Turkish evaluation) towards the study should appear at the end of the article.

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Case Reports

Case reports should present cases which are rarely seen, feature novelty in diagnosis and treatment, and contribute to our current knowledge. The first page should include the title in Turkish and English, an unstructured summary not exceeding 50 words, and key words. The main text should consist of introduction, case report, discussion and references. The entire text should not exceed 1500 words (A4, formatted as specified above). A maximum of 10 references shall be used in case reports.

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Review articles can address any aspect of clinical or laboratory pediatry. Review articles must provide critical analyses of contemporary evidence and provide directions of or future research. **The journal only accepts and publishes invited reviews.** Before sending a review, discussion with the editor is recommended.

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Editorial / Editörden

Dear Readers,

Welcome to the first issue of "The Journal of Pediatric Research" of 2017. We all hope 2017 may bring peace and justice to all children of the world. All children deserve to be happy, and we want to see them in full health on playgrounds and schools, not on battlefields.

Another feature of this issue is that all articles are written in English. We are delighted to have two valued authors from India and Russia who are sharing their research with us.

After the renovation of "The Journal of Pediatric Research" on 2014, we welcomed a rapidly increasing number of articles nationally and from abroad to be evaluated for publication. We proudly announce that our journal is now listed in Ulakbim-Tübitak database in addition to ESCI, EBSCOhost and "Türkiye Atıf Dizini." This is a great opportunity for us to spread our enthusiasm for improving our journal to the best.

Children are surrounded by many hazards like the excessive internet channels, environmental threats like endocrine disrupting agents and drug abuse in this era. As physicians and nurses in the pediatric care, we feel that it is our utmost duty to raise awareness of these threats and protect them. In this issue, we have two interesting original research articles covering these: "Substance Use Among Adolescents, and Influencing Factors in Şanlıurfa" by Karataş et al. from Şanlıurfa, Turkey, and "Endocrine Disrupting Chemicals: A Challenge to Child Health" by Mani et al. from India.

Research on child nutrition is never outdated in pediatrics. There are two original research articles covering different aspects of hypercholesterolemia and vitamin D deficiency for your attention.

The field of pediatrics requires complete integration of all relevant disciplines. Thus, we are more than happy to have two articles on orthopedic surgery and child psychiatry in this issue.

Our journal will be published in English beginning from 2018. We, therefore, invite articles written in English from all over the world to have a mutual ground for sharing knowledge and discussing health and well-being of all children.

As the Editor of this issue, I would like to take the opportunity to thank the members of our editorial board, reviewers, authors, and Galenos Publishing House for their tremendous help in preparing this featured issue of 2017.

We look forward to accepting your future papers. I believe that present and future design of "The Journal of Pediatric Research" will fulfill an important role by publishing scientific studies that advance the care of children by honoring caregivers who not only diagnose and treat but also have insight and talent to present their experience to the scientific community.

Prof. Dr. Güzide Aksu

Section Editor

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Original Article / Orijinal Araştırma

Association Between Soluble CD40 Ligand and Hypercholesterolemia in Children and Adolescents

Çocuk ve Adölesanlarda Soluble CD40 Ligandı ile Yüksek Kolesterol Düzeylerinin İlişkisi

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ABSTRACT

Aim: Coronary heart disease is one of the most common causes of death around the world. The pathological process of coronary heart disease like atherosclerosis starts in childhood. During this period thrombosis constitutes a high-risk factor. In this study, we investigated the effect of soluble CD40 ligand (sCD40L) and clotting activation on children and adolescents with hypercholesterolemia.

Materials and Methods: Plasma levels of sCD40L, P-selectin, 8-hydroxy-2-deoxyguanosine (8-OHdG), and prothrombin fragment 1+2 [(F) 1+2] were determined in thirty-five hypercholesterolemic patients (20 girls and 15 boys; age, median: 13 years) and forty healthy normocholesterolemic subjects (28 girls and 12 boys; age, median: 13 years).

Results: No significant differences were observed between the patient group and controls in terms of age, high-density lipoprotein (HDL) cholesterol, 8-OHdG, F1+2 ($p>0.05$). However, there were significant differences between the two groups with respect to total cholesterol, low-density lipoprotein (LDL) cholesterol, very LDL cholesterol, triglycerides, sCD40L and P-selectin ($p<0.05$), which were higher in the patient group than the controls. A positive correlation was observed between sCD40L and P-selectin ($p<0.05$) in accordance with the Spearman correlation analysis. The correlation coefficients were 0.735 in the patient group and 0.647 in the control group. But there was no significant

ÖZ

Amaç: Koroner kalp hastalığı tüm dünyadaki ölüm nedenleri arasında önemli bir yere sahiptir. Ateroskleroz gibi koroner kalp hastalığının patolojik süreci çocukluk çağı dönemlerinde başlamakta olup tromboz bu süreçte önemli bir risk faktörüdür. Bu çalışmada hiperkolesterolemili çocuk ve adölesanlarda soluble-CD40 ligand (sCD40L) ile pıhtılaşmayı aktifleştiren faktörlerin etkisi araştırılmıştır.

Gereç ve Yöntemler: Plazma sCD40L, P-selektin, 8-hidroksi-2-deoksiguanozin (8-OHdG), protrombin fragmanı 1+2 [(F) 1+2] düzeyleri, 35 hiperkolesterolemili hasta (20 kız ve 15 erkek; yaş ortalaması 13 yıl) ile 40 sağlıklı kontrol (28 kız, 12 erkek; yaş ortalaması 13 yıl) grubunda değerlendirilmiştir.

Bulgular: Her iki grup arasında yaş, yüksek dansiteli lipoprotein (HDL) kolesterol, 8-OHdG, F1+2 düzeyleri açısından anlamlı farklılık saptanmamıştır ($p>0,05$). Ancak toplam kolesterol, düşük dansiteli lipoprotein (LDL) kolesterol, çok LDL kolesterol, trigliserit, sCD40L ve P-selektin değerleri gruplar arasında anlamlı olarak farklı olup tüm bu değerler hasta grupta daha yüksek olarak ölçülmüştür ($p<0,05$). Spearman korelasyon analizine göre, sCD40L and P-selektin arasında pozitif korelasyon saptanmıştır ($p<0,05$). Korelasyon katsayısı hasta grupta 0,735 ve kontrol grupta 0,647 olarak ölçülmüştür. sCD40L ile toplam kolesterol, LDL-kolesterol, HDL-kolesterol,

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correlation between sCD40L and total cholesterol, LDL-cholesterol, HDL-cholesterol, very LDL-cholesterol, triglycerides, 8-OHdG, F1+2 ($p>0.05$).
Conclusion: We believe that future prospective studies to determine the increase in the level of sCD40L with a larger sample size of a pediatric population with dyslipidemias may be more helpful in predicting the risk of cardiovascular disease.
Keywords: Hypercholesterolemia, atherosclerosis, CD40 ligand

çok LDL-kolesterol, trigliserit, 8-OHdG, F1+2 arasında anlamlı korelasyon kurulamamıştır ($p>0,005$).
Sonuç: Daha geniş dislipidemili çocuk popülasyonunda yapılacak çalışmalarla sCD40L düzeyindeki artışların kardiyovasküler hastalık oluşum riskini belirlemede yardımcı olabileceğini düşünmekteyiz.
Anahtar Kelimeler: Hiperkolesterolemi, aterosklerozis, CD40 ligand

Introduction

Coronary heart disease (CHD) and atherosclerosis are the common causes of death around the world. The pathological process of CHD starts in childhood and its clinical effects are generally seen in adult patients. There are a lot of risk factors as underlying mechanisms of atherosclerosis, and one of these factors is hypercholesterolemia. Epidemiologic and clinical trials have provided clear evidence that cholesterol has a role in the occurrence of cardiovascular disorders (1,2). This may be related to enhanced thrombotic risk. Activated platelets adhere to the intact endothelium and induce inflammatory responses locally, which substantially contribute to the early phase of atherosclerosis. It has been seen that patients with hypercholesterolemia have increased urinary excretion of 11-dehydro-thromboxane B2 and elevated platelet response (3). Another study (4) states that the tendency to prothrombotic state is higher when the formation of monocyte tissue factor (TF) is increased, and prothrombin fragment [(F) 1+2] (a marker of thrombin generation *in vivo*) levels are elevated.

The expression of CD40 ligand (CD40L) is observed on CD4+ T cells, activated platelets, and vascular system cells (endothelial cells (ECs), smooth muscle cells (SMCs) and macrophages) (5,6). The interaction between the CD40L that is expressed on vascular cells, and soluble CD40L (sCD40L) causes inflammatory and prothrombotic responses, resulting in increased atherosclerotic progression (5,7). When CD40 and sCD40L are adhered, TF in SMCs, ECs, and macrophages are over expressed. Thus TF can provide the convertion of factor X to Xa on SMCs within the atherosclerotic plaque (8-10). Previous studies demonstrated that oxidative stress plays a major role in the monocyte expression of TF by promoting nuclear factor-kappa-B activation (11). The plasma level of P-selectin, an activation marker of platelets, can reflect the earlier defects of platelets.

To explore the effect of clotting factors in pediatric CHD pathogenesis, we investigated the behavior of sCD40L and clotting activation in children and adolescents with hypercholesterolemia.

Materials and Methods

Patients and Controls

Thirty-five hypercholesterolemic patients whose low-density lipoprotein (LDL)-cholesterol levels were >155 mg/dL (4 mmol/L) (20 girls and 15 boys; age, median: 13 years, minimum: 4 years, maximum: 18 years) and who had a history of familial hypercholesterolemia, and forty healthy normocholesterolemic subjects (28 girls and 12 boys; age, median: 13 years, minimum: 4 years, maximum: 18 years) were asked to participate in this study. Physical examination showed that none of the patients had any clinical evidence of peripheral vascular or inflammatory diseases. Potential patients with infection, tumor, diabetes, hypertension, hypothyroidism, hypertriglyceridemia, liver or kidney disease were excluded. Other exclusion criteria were, family history of thrombosis, metabolic syndrome, and medication, which might affect inflammatory and thrombotic markers.

The study was approved by the Gazi University Local Ethics Committee (approval number: 015, 21 January 2008) and all patients and their families provided written informed consent.

Blood Samplings

Peripheral venous blood samples were obtained after a 12-hour fast. The lipid profile (ß-quantification) was analyzed on fresh samples. The subjects whose total cholesterol and LDL-cholesterol were >95th percentile according to age and sex were accepted as hypercholesterolemic. Total cholesterol and high-density lipoprotein (HDL) levels were determined by colorimetric-spectrophotometric autoanalyzer (Aeroset®, Abbott®, Illinois, USA) at 500 nm according to the Trinder reaction. LDL levels were calculated according to the Friedewald formula [Total cholesterol - (HDL + very LDL [VLDL])]. Blood samples were immediately centrifuged at 2000 rpm for 20 min at 4 ˚C and the supernatant was collected and stored at-80 ˚C until measurement. Plasma levels of sCD40L, sP-selectin, 8-hydroxy-2-deoxyguanosine (8-OHdG), F1+2 were determined using commercial ELISA kits according to the manufacturers' instructions (human sCD40L ELISA, Bender Med, Austria; human sP-selectin ELISA, Bender Med, Austria; 8-hydroxy-2 deoxyguanosine ELISA, Northwest Life Science Specialties, LLC, USA; prothrombin F1+2 ELISA, Dade Behring Inc., USA).

Statistical Analysis

Statistical analysis was performed with SPSS 11.5 software for Windows (SPSS Inc., Chicago, Illinois). All

results are expressed as mean, standard deviation, median, minimum and maximum. Kolmogorov-Smirnov test was used to verify that the data were within normal distribution.

Mann-Whitney U test was used for the comparison of the groups. The association of measurements with other biochemical parameters was assessed by the Spearman rank correlation test. A p value of <0.05 was considered statistically significant.

Results

Age and laboratory parameters of the patient and control groups are shown in Table I. No significant differences were observed between the patient group and controls in terms of age, HDL-cholesterol, 8-OHdG, F1+2 (p>0.05). On the other hand, there were significant differences with respect to total cholesterol, LDL cholesterol, VLDL cholesterol, triglycerides, sCD40L and P-selectin (p<0.05). These values were higher in the patients than the controls. Serum levels of cholesterol and LDL-cholesterol were 308.7±142.4, 243.7±146.7 mg/dL in subjects with hypercholesterolemia, and 154.2±17.3 and 86.8±17.5 in those with normocholesterolemia. These differences between the two groups were significant (p<0.001). Compared to subjects with hypercholesterolemia, those with normocholesterolemia had higher values of sCD40L, as 3.3±2.8 and 1.9±2.3, respectively (p=0.004). In respect of P-selectin, the levels were 235.6±73.5 in hypercholesterolemic subjects and 209.3±96.1 in normocholesterolemic subjects (p=0.0049).

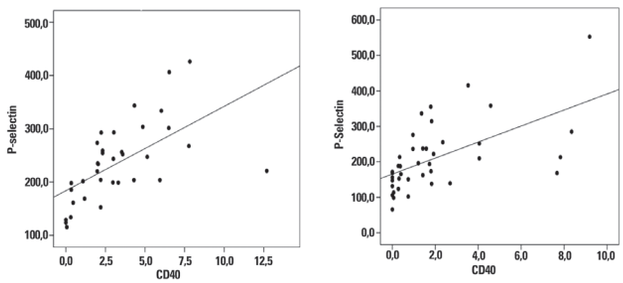
A positive correlation was observed between sCD40L and P-selectin (p<0.05) (Spearman correlation analysis) (Figure 1). Correlation coefficients were 0.735 in the patient group, and 0.647 in the control group. But there was no significant correlation between sCD40L and total cholesterol, LDL-cholesterol, HDL-cholesterol, VLDL-cholesterol, triglycerides, 8-OHdG, F1+2 (p>0.05) (Table II).

Discussion

This study provides evidence that, increased sCD40L and P-selectin levels, which are known as inflammatory molecules, are involved in the onset and progression of atherosclerotic disease in children with hypercholesterolemia.

Hypercholesterolemia, a risk factor for cardiovascular disease, is associated with inflammation and hypercoagulability. Thrombophilia is a frequent occurrence in patients with hypercholesterolemia, and can precipitate cardiovascular events in this setting.

Thrombophilia is basically characterized by platelet hyperactivation and up-regulation of TF with enhanced thrombin generation (3,4). CD40L may represent an important link between platelet activation and TF expression (5). It has been reported in a study (12) that, considerable increase in sCD40L would help the prediction of probable cardiovascular events problems in healty women. Another study (13) shows that, higher sCD40L would result in recurrent cardiovascular morbidity and mortality because of acute coronary syndromes. In patients with hypercholesterolemia, CD40L is up-regulated on the platelet surface and (14) may therefore represent an important stimulus for TF expression and clotting activation. It has been shown that, there is a significant correlation between sCD40L and prothrombin



Hypercholesterolemic patients Healthy subjects

Figure 1. Correlations between CD40 and P-selection

Table I. Markers in patients with familial hypercholesterolemia as compared with unaffected controls

Parameters	Hypercholesterolemic patients (n=35)			Healthy subjects (n=40)			p
	$\bar{X}$±SD	Med	(Min-Max)	$\bar{X}$±SD	Med	(Min-Max)	
Age (year)	12.5±3.5	13.0	(4.0-18.0)	12.4±3.5	13.0	(4.0-18.0)	0.969
Total cholesterol (mg/dL)	308.7±142.4	252.0	(220.4-704.8)	154.2±17.3	153.4	(118.0-188.0)	**<0.001**
LDL-cholesterol (mg/dL)	243.7±146.7	175.0	(160.0-657.0)	86.8±17.5	88.8	(50.0-122.6)	**<0.001**
HDL-cholesterol (mg/dL)	46.5±10.3	47.5	(25.0-71.0)	51.2±10.5	50.5	(36.0-85.0)	0.130
VLDL-cholesterol (mg/dL)	20.7±8.5	19.8	(9.4-49.0)	17.8±13.2	13.3	(6.6-89.8)	**0.013**
Triglycerides (mg/dL)	103.0±42.0	95.0	(47.0-245.0)	80.5±32.9	66.5	(33.0-175.0)	**0.009**
CD40	3.3±2.8	2.3	(0.0-12.7)	1.9±2.5	1.3	(0.0-9.2)	**0.004**
P-selectin	235.6±73.5	233.8	(115.4-426.3)	209.3±96.1	188.0	(65.8-552.6)	**0.049**
8-OHdG	8176.0±36160.7	2077.5	(1519.3-215992.0)	2058.4±212.3	2097.5	(915.5-2287.5)	0.675
F1+2	719.6±304.6	643.6	(286.4-1490.2)	838.5±377.4	714.3	(422.6-2159.9)	0.181

SD: Standard deviation, Med: Median, Min: Minimum, Max: Maximum, LDL: Low-density lipoprotein, HDL: High-density lipoprotein, VLDL: Very low-density lipoprotein, 8-OHdG: 8-hydroxy-2-deoxyguanosine, F: Fragment

F1+2 (15). Because CD40L exerts a prothrombotic effect through overexpression of TF, it would be reasonable to speculate that in hypercholesterolemia, clotting activation occurs as a consequence of CD40L overexpression.

This current study aimed to measure the plasma levels of sCD40L and other parameters in children with hypercholesterolemia. Although there are numerous similar studies of adults with hypercholesterolemia, measuring sCD40L concentrations in children with hypercholesterolemia is quite novel. According to our results, sCD40L was significantly higher in hypercholesterolemic patients than the normocholesterolemic control group in childhood. Of course it is important to follow this parameter and the clinical condition of all the subjects in the future for the possibility of coronary syndrome. So we can speculate that this sCD40L parameter can be an indicator to screen the CHD in childhood.

P-selectin is an adhesion molecule known to be involved in the pathogenesis of athero-thrombosis. Being the primary adhesion molecule in initiating cell activation and cell adhesion to platelets, leucocytes and ECs, it's believed that P-selectin may contribute to endothelial dysfunction, atherosclerosis and thrombosis (16). One research showed that, increased levels of soluble P-selectin with higher levels of cholesterol in obese children, might indicate EC damage and platelet activation (17). In our study, P-selectin level was higher in the hypercholesterolemic patients. Additionally a positive correlation between sCD40L and P-selectin was found. As well as being mediators of inflammation and having a significant effect on atherosclerosis, high levels of P-selectin can be a risk factor for CHD. 8-OHdG is a product of oxidative deoxyribonucleic acid (DNA) damage following specific enzymatic cleavage after 8-hydroxylation of the guanosine base. In recent years, 8-OHdG has been used widely in many studies not only as a biomarker for the measurement of endogenous oxidative DNA damage but also as a risk factor for many diseases including cancer and some degenerative diseases (18). According to our findings, there was no significant difference in the 8-OHdG levels of the hypercholesterolemic children compared to the control group. In a previous study, it was shown that the level of 8-OHdG was lower in hypercholesterolemic and normocholesterolemic children than in adults correspondingly (19). This result can be explained by the fact that age is associated with changes of mechanisms controlling oxidative stress and platelet activation. Therefore, our result can be related to the age of the participants. However, in that study (19) there was a significant correlation between the sCD40 and 8-OHdG levels in both hypercholesterolemic children and the adult group. In our study, we couldn't find a positive correlation between those parameters. We may explain this result with the limited number of patients in the study population.

Table II. Correlations between CD40 and other parameters

Parameters	Hypercholesterolemic patients (n=35)		Healthy subjects (n=40)	
	Spearman rho	p	Spearman rho	p
Total cholesterol (mg/dL)	-0.015	0.932	0.237	0.142
LDL-cholesterol (mg/dL)	-0.096	0.584	0.120	0.461
HDL-cholesterol (mg/dL)	-0.184	0.290	-0.073	0.657
VLDL-cholesterol (mg/dL)	0.126	0.472	0.267	0.096
Triglycerides (mg/dL)	0.059	0.735	0.202	0.210
P-selectin	0.735	**<0.001**	0.647	**<0.001**
8-OHdG	0.119	0.495	0.021	0.898
F1+2	-0.086	0.624	0.098	0.546

rho: Correlation coefficients, LDL: Low-density lipoprotein, HDL: High-density lipoprotein, VLDL: Very low-density lipoprotein, 8-OHdG: 8-hydroxy-2-deoxyguanosine, F: Fragment

F1+2, which comes from in vivo cleavage of prothrombin by factor Xa, is considered to be useful for the diagnosis of thrombosis (20). Increased F1+2 levels have been reported in venous thromboembolism, inflammation, cancer, sepsis, acute coronary syndromes, stroke, peripheral arterial disease, atrial fibrillation, and during the postoperative period. In our study, no relation was found in the F1+2 levels between the patient and the control groups. This result may relate to the differences of the coagulation characteristics in the pediatric population, as variability of these parameters in pediatric population are yet unknown.

In this study, we measured the levels of sCD40L in hypercholesterolemic patients as higher than the healthy control group, which is in accordance with the previous studies (13-15,21). P-selectin levels were higher in the hypercholesterolemic patients than the controls. The expression of CD40 is observed on various cell types such as: ECs, SMCs, macrophages and activated T lymphocytes. The engagement of CD40 and sCD40L triggers atherosclerosis. The expression of vascular cell adhesion molecule-1, intercellular adhesion molecule-1, and P-selectin on ECs facilitate the participation of monocytes and lymphocytes in the injury. Like sCD40L, interleukin (IL)-1, IL-6, and tumor necrosis factor-alpha may have a role in this process. The production of reactive oxygen species are elevated with regard to the CD40-sCD40L engagement, which induces the inhibition of endothelial nitric oxide, causing endothelial dysfunction and promoting atherogenesis (22,23). It is a well-known fact that the degree of the plasma level of cholesterol is directly related to the damage of vascular ECs. This study showed an elevated level of sCD40L in the hypercholesterolemic patients compared to the healthy controls. It was also shown that there was a positive correlation between the level of sCD40L and the level of P-selectin. Therefore, we can speculate that if one of these parameters is normal in range, the other facilitator factor

might be decreased. Another important aspect of this study is that the participitants were selected from childhood and the adolescent period. To our knowledge, there are very few studies in the literature that lead to the understanding of the mechanism of premature atherogenesis among children.

Study Limitations

The sample size for the patient group was small.

Conclusion

In conclusion, future prospective studies with a larger sample size might better detect the increased sCD40L level in the setting of a pediatric population with familial dyslipidemias, which may be more helpful in predicting the risk of cardiovascular events.

Ethics

Ethics Committee Approval: The study was approved by the Gazi University Local Ethics Committee. (Approval number: 015, 21 January 2008), Informed Consent: Consent form was filled out by all participants.

Peer-review: Internally peer-reviewed.

Authorship Contributions

Concept: Alev Hasanoğlu, Design: Leyla Tümer, Data Collection or Processing: Aynur Küçükçongar Yavaş, Analysis or Interpretation: Arzu Aral, Literature Search: İlyas Okur, Fatma Tuba Eminoğlu, Writing: Aynur Küçükçongar Yavaş.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: The authors declared that this study received no financial support.

References

1. Castelli WP, Anderson K. A population at risk. Prevalence of high cholesterol levels in hypertensive patients in the Framingham Study. Am J Med 1986;80:23-32.
2. Iso H, Jacobs DR Jr, Wentworth D, Neaton JD, Cohen JD. Serum cholesterol levels and six-year mortality from stroke in 350,977 men screened for the multiple risk factor intervention trial. N Engl J Med 1989;320:904-10.
3. Davi G, Averna M, Catalona I, et al. Increased thromboxane biosynthesis in type IIa hypercholesterolemia. Circulation 1992;85:1792-8.
4. Ferro D, Basili S, Alessandri C, Mantovani B, Cordova C, Violi F. Simvastatin reduces monocyte-tissue-factor expression type IIa hypercholesterolemia. Lancet 1997;350:1222.
5. Henn V, Slupsyk JR, Grafe M, et al. CD40 ligand on activated platelets triggers an inflammatory reaction of endothelial cells. Nature 1998;391:591-4.
6. Schönbeck U, Libby P. The CD40/CD154 receptor/ligand dyad. Cell Mol Life Sci 2001;58:4-43.
7. Lindmark E, Tenno T, Siegbahn A. Role of platelet P-selectin and CD40 ligand in the induction of monocytic tissue factor expression. Arterioscler Thromb Vasc Biol 2000;20:2322-8.
8. Hathcock J. Vascular biology-the role of tissue factor. Semin Hematol 2004;41(1 Suppl 1):30-4.
9. Schoenbeck U, Mach F, Sukhova GK, et al. CD40 ligation induces tissue factor expression in human vascular smooth muscle cells. Am J Pathol 2000;156:7-14.
10. Mach F, Schoenbeck U, Libby P. CD40 signaling in vascular cells: a key role in atherosclerosis? Atherosclerosis 1998;137 Suppl:S89-95.
11. Bowie A, O'Neill LA. Oxidative stress and nuclear factor-kappaB activation: a reassessment of the evidence in the light of recent discoveries. Biochem Pharmacol 2000;59:13-23.
12. Schonbeck U, Varo N, Libby P, Buring J, Ridker PM. Soluble CD40L and cardiovascular risk in women. Circulation 2001;104:2266-8.
13. Heeschen C, Dimmeler S, Ham CW, et al. Soluble CD40 ligand in acute coronary syndromes. N Engl J Med 2003;348:1104-11.
14. Garlics CD, John S, Schmeisser A, et al. Upregularion of CD40 and CD40 ligand (CD154) in patients with moderate hypercholesterolemia. Circulation 2001;104:2395-400.
15. Sanguigni V, Ferro D, Pignatelli P, et al. CD40 ligand enhances monocytes tissue factor expression and thrombin generation via oxidative stres in patients with hypercholesterolemia. J Am Coll Cardiol 2005;45:35-42.
16. Blann AD, Nadar SK, Lip GY. The adhesion molecule P-selectin and cardiovascular disease. Eur Heart J 2003;24:2166-79.
17. Gallistl S, Sudi KM, Borkenstein M, Weinhandl G, Zotter H, Muntean W. Correlation between cholesterol, soluble P-selectin, and D-dimer in obese children and adolescents. Blood Coagul Fibrinolysis 2000;11:755-60.
18. Valavanidis A, Vlachogianni T, Fiotakis C. 8-hydroxy-2' -deoxyguanosine (8-OHdG): A critical biomarker of oxidative stress and carcinogenesis. J Environ Sci Health C Environ Carcinog Ecotoxicol Rev 2009;27:120-39.
19. Martino F, Pignatelli P, Martino E, et al. Early increase of oxidative stress and soluble CD40L in children with hypercholesterolemia. J Am Coll Cardiol 2007;49:1974-81.
20. Páramo JA. Prothrombin fragments in cardiovascular disease. Adv Clin, Chem 2010;51:1-23.
21. Sanguigni V, Pignatelli P, Lenti L, et al. Short-Term Treatment With Atorvastatin Reduces Platelet CD40 Ligand and Thrombin Generation in Hypercholesterolemic Patients. Circulation 2005;111:412-9.
22. Watanebe M, Sangawa A, Sasaki Y, et al. Distrubution of inflammatory cells in adventia changed with advencing atherosclerosis of human coronary artery. J Atheroscler Thromb 2007;14:325-31.
23. Ueland T. Aukrust P, Yndestad A, et al. Soluble CD40 ligand in acute and chronic heart failure. Eur Heart J 2005;26:1101-7.

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Original Article / Orijinal Araştırma

Intestinal Metaplasia of Antral Superficial-foveolar Epithelium in Children with Atrophic Gastritis

Atrofik Gastritli Çocuklarda Antral Yüzeyel-foveolar Epitelin İntestinal Metaplazisi

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ABSTRACT

Aim: Chronic gastritis (CG), being one of the most common digestive diseases, is frequently underestimated both by patients and clinicians. However, CG developed as early as in childhood, and often accompanied by Helicobacter pylori contamination of antral mucus, has a persistent recurrent course in adult patients, complicated by mucosal transformations in the form of atrophy, metaplasia and dysplasia. CG can appear as an early stage in the morphogenesis of tumor growth. From this point of view, CG is worth noticing.

Materials and Methods: A complex morphological study of antral mucosa in school children with atrophic gastritis has been performed using histochemistry, immunohistochemistry and a morphometric methods.

Results: In atrophic gastritis we have revealed the decrease in the number of functionally mature cells responsible for the production of extracellular matrix and basal membrane components, the lack of which in paraepithelial localization results in the change of their special properties. In addition, sulphomucins, non-characteristic of stomach, overlapped on mucosa and in single foveolae have been found in atrophic gastritis.

Conclusion: Permanent impairment in the conjugation of cell and tissue components in mucosa accompanied by the change of an epithelial layer synthetic function with the impaired physicochemical properties of gastric mucin results in intestinal metaplasia as early as in childhood.

Keywords: Gastritis, intestinal metaplasia, regeneration, mucin

ÖZ

Amaç: Çocukluk çağının en sık görülen sindirim sistemi problemlerinden olan kronik gastrit (KG) çoğu kez hastalar ve klinisyenler tarafından küçümsenmektedir. Halbuki çocuklukta başlayıp sıklıkla antral mukusun *Helicobacter pylori* ile bulaşının eşlik ettiği KG, erişkin dönemde atrofi, metaplazi ve displazi gibi mukoza değişiklikleriyle komplike olan kalıcı, yineleyen bir seyir izlemektedir. KG tümör gelişimi morfogenezinin erken bir evresi olarak ortaya çıkabilir. Bu bakış açısıyla, KG dikkate alınması gereken bir patolojik durumdur.

Gereç ve Yöntemler: KG'li okul çocuklarında, histokimyasal, immünohistokimyasal ve morfometrik yöntemlerle antral mukozanın ayrıntılı morfolojik değerlendirilmesi gerçekleştirilmiştir.

Bulgular: Atrofik gastritte, paraepitelyal bölgedeki yoklukları, kendilerine özgül özelliklerin değişimiyle sonuçlanan ekstraselüler matriks ve bazal membran bileşenlerinin üretiminden sorumlu fonksiyonel yönden matür hücrelerin sayısında azalma saptadık. Buna ek olarak, yine atrofik gastrik örneklerinde midenin karakteristik bileşeni olmayan sülfomüsinlerin mukoza ve tekli foveolalarda birikimi saptandı.

Sonuç: Hücre ve doku bileşenlerinin bağlanımındaki kalıcı hasar ve buna eşlik eden epitelyal tabakanın sentetik fonsiyonunun değişimiyle birlikte gastrik müsinin bozulmuş fizikokimyasal özellikleri, çocukluk çağı gibi çok erken bir dönemde intestinal metaplaziye yol açmaktadır.

Anahtar Kelimeler: Gastrit, intestinal metaplazi, rejenerasyon, müsin

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Introduction

Chronic gastritis (CG), being one of the most common digestive diseases, is frequently underestimated both by patients and clinicians (1-3). However, CG developed as early as in childhood, and often accompanied by *Helicobacter pylori* contamination of antral mucus, has a persistent recurrent course in adult patients complicated by mucosa transformations in the form of atrophy, metaplasia and dysplasia. CG can appear as an early stage in the morphogenesis of tumor growth (4,5). From this point of view, CG is worth noticing (6,7).

According to the majority of experts in intestinal metaplasia (IM), which is able to act as precursor of neoplastic processes, the failure of epithelial cell differentiation occurs after epithelial layer damage (8,9). This has been supported by the studies carried out over the last years. According to their data, undifferentiated cells pre-existing in gastric epithelium can be differentiated by uncharacteristic intestinal type under certain unfavorable conditions. In addition, the disorders in cell differentiation are not due to drastic genome changes but determined by physico-chemical factors, which have an impact on them (10-12). And the formation of further transformations against the background of IM is due to its expansion and the character of mucins produced by epithelium (13-15).

However, in modern literature the causes and mechanisms of gastric epithelial IM formation (16-18) still remain under-investigated, and they are mainly represented by the studies of cell kinetics, the activity and intensity of inflammatory changes in adult population (19-21).

The present study aims to investigate the structural framework origin of the competence of the regeneration processes in antral mucosa (AM) with IM for the early optimization of the management and prevention of the formation of further neoplastic processes, as well as the implementation of the possibility of epithelium with metaplasia to regress and become normal. Therefore, the study of the biopsy material from children seems to be the most relevant objective (22-25).

The objective of this study was to determine the disorders of epithelial stromal relations contributing to the formation of a quite new epithelium in the form of IM foci in AM in children.

Materials and Methods

The present study is an observational, retrospective, analytical case-control. The materials were the antral biopsies of 7 to 16-year-old 1367 children with chronic gastroduodenitis, undergoing treatment at the clinic of Nizhny Novgorod Research Institute of Pediatric Gastroenterology over the period of 2001-2012. The patients underwent endoscopic and morphological examination in a strict accordance with the management algorithm of a gastrointestinal patient. All patients gave their informed consent to undergo medical procedures and satisfied the requirements of the Local Ethics Committee of Nizhny Novgorod Research Institute developed according to World Medical Association's Declaration of Helsinki (2000) (approval number: 2007/28). Morphologic and functional assessment of gastric mucosa biopsies, the character of *Helicobacter pylori* contamination were performed in accordance with Sydney system classification, while the severity degree was evaluated according to Classification and Grading of Gastritis, Houston (26-28).

The retrospective analysis of AM histological examinations over the period of 2001-2012 enabled to pick out children with gastroduodenitis from a group of children (n=42) who had undergone multiple (not less than 4) courses of treatment in the clinic, and over the last years were diagnosed with nonatrophic gastritis associated with *Helicobacter pylori* with +, ++ invasion degree, and in the last admissions they were found to have atrophic gastritis. Morphological study of AM in 42 patients with atrophic gastritis revealed IM in 11 cases.

The patients were classified into the following groups depending on the nature of changes in AM:

Group 1 (comparison group). *Helicobacter pylori* associated non-atropic gastritis patients with a contamination degree of +, ++ (n=30),

Group 2. Atrophic antrum-gastritis patients without IM (n=31),

Group 3. Atrophic antrum-gastritis patients with IM (n=11).

Biopsy material obtained by gastroesophageal endoscopy was embedded in 10% buffered formalin (pH: 7.2-7.4) and exposed to standard histological treatment (dehydration, de-embedding), and paraffin sections of 5 μm in thickness were received. The prepared microsections were hematoxylin and eosin stained. Histochemical identification of mucins produced by AM epithelial cells was performed using periodic acid-Schiff (PAS)-alcian blue stain in pH 2.5. The intensity of mucin staining was estimated by semiquantitative method in scores: (+)-weak; (++)-moderate; (+++)-high. Concurrently, we carried out an immunohistochemical reaction using Muc2, Muc5AC and Muc6 markers ("Novocastra"). Mast cells were defined using basic brown, and classified according to maturation, granulation degree, and location in mucosa. Immunohistochemistry was performed to assess the intensity of the renewal processes of gastric mucous epithelium using antibodies to Ki-67 antigen (MIB-1, "Novocastra") expressed in all phases of a cell cycle. The antigen was de-shielded in citrate buffer (pH 6.0) in boiling water bath within 1 h. NovoLink Polymer Detection System was used to reveal the expression of the markers (29,30). 3.3-diaminobenzidine tetrachloride was used as chromogen. Cell nuclei were counterstained by hemalum within 2 min. By means of 400-fold magnification of a microscope, proliferation index (nuclear label Ki-67) was defined as a percentage of positively stained nuclei of epithelial cells of AM in 10 randomly chosen visual fields. Histologic specimens were examined and photographed using Nikon Eclipse E400 microscope with Nikon DS-Fi2 camera and software NIS-

Elements Basic Research. For an objective study, the gastric mucosa structure was examined morphometrically. Cells were counted in 10 F. v., field lens 90 and ocular lens 10, and then the absolute number of cells per 1 mm^2 was calculated.

The results of the study were statistically processed. We determined the indices of descriptive statistics, the normalcy of distribution was checked using Kolmogorov-Smirnov test. Since the data distribution failed to match the normal distribution criteria, non-parametric Mann-Whitney U test was used in the statistical processing of the data. P=0.05 was taken as the critical level in statistical hypothesis testing.

Results

AM in non-atrophic antrum-gastritis (group 1) was characterized by structural alterations indicating the development of chronic catarrhal damage. Diffuse inflammatory infiltration was found in 67% of the cases, and surface infiltration in 33%. Superficial-foveolar epithelium over much of the mucosa was flattened, moderately infiltrated by inter-epithelial lymphocytes with insignificant content of eosinophils and neutrophils. Mucous cells showed a low degree of dystrophy with irregular mucoid production in cells. Neutral and acid mucopolysaccharide staining on epithelial surface revealed a mainly small and moderate amount of mucus located between the folds. PAS-positive staining of mucous cells covered just from 1/4 to 1/2 of the cell area versus 2/3 of the norm that indicated their low functional activity.

In the AM stromal compartment, the edema and full-blood vessels of microvasculature were associated with high cell density infiltration. Throughout the subepithelial zone and proper mucous plate (PMP) there was an increased vessel formation (microvasculature), most vessels had a dilated lumen, thickened wall with distended nuclei of endotheliocytes, some of which had sludge phenomenon of red blood cells.

Lymphocytes were dominant in inflammatory cell infiltrates in all cases, and among them there were plasma cells with different maturity degrees. A moderate number of eosinophils and neutrophils was determined. The number of fibroblastic cells had a tendency to decrease (Table I). Such a morphological picture with a long-time persistence of mononuclear infiltrate, with an inflammatory component prevailing, leads to the accumulation of toxins damaging epithelial cytoskeleton with intercellular structure weakening, resulting in the limitation of cell functional capabilities in the quality and quantity of mucus production.

In AM atrophic changes, 98% of the cases showed no *Helicobacter pylori* microbial invasion. Lymphoid follicles without cleared germinal centers were formed in 16% of observations. Focal proliferation of connective tissue in PMP was found in all cases, with collagen fibers locating predominantly between the glands without changing their contour. The epithelial surface had a small amount of mucus containing generally neutral mucopolysaccharides.

The inflammatory and dystrophic changes in groups of atrophic gastritis patients with and without intestinal metaplasia did not differ significantly. In contrast to the comparison group, lymphocytes predominated in the inflammatory infiltrates in these groups, while plasma cells were minimal, and neutrophils and eosinophils were represented by single cells. Alongside with that there was moderate dystrophy of both superficial-foveolar epitheliocytes and mucous cells with distended nuclei.

Detailed study of the AM biopsies of group 2 children (atrophic gastritis without IM) showed formed structural changes. Mucosa was characterized by the reduced height of surface epithelium, flattened epitheliocytes in both surface and foveolar epithelium with nuclei displacement in central parts of the cells, and preservation of clear cell boundaries. In addition, the functionalities of epithelium remained unchanged, which was confirmed by the sufficient synthesis of superficial-foveolar and glandular epithelium of corresponding mucins.

Basal membrane in some areas was loosened, and the intensity degree of its PAS-positive staining varied from slightly intense (+) to maximum intense (+++).

Against the general background of PAS-positive secretion produced by the epitheliocytes of superficial-foveolar epithelium, the appearance of alcyanophil secretion overlapping the mucosa in the gaps between single foveae in 3.3% of the histologic specimens of group 2 drew attention (Figure 1).

There was a total reduction in the number of gastric foveae, and tendency of the mucosa to preserve and maintain cytoarchitecture accompanied by the formation of deepened, tortuous and concrescent (two-chambered) foveae. Proliferative activity of the superficial-foveolar area was higher compared to that of the comparison group (p=0.04) (Figure 2), and there was no significant change of this parameter when compared to group 3 (p=0.091).

Infiltrate cell density in the interfoveolar space in antral PMP in group 2 children was significantly lower (p=0.013) than that in the comparison group (group 1). In PMP,

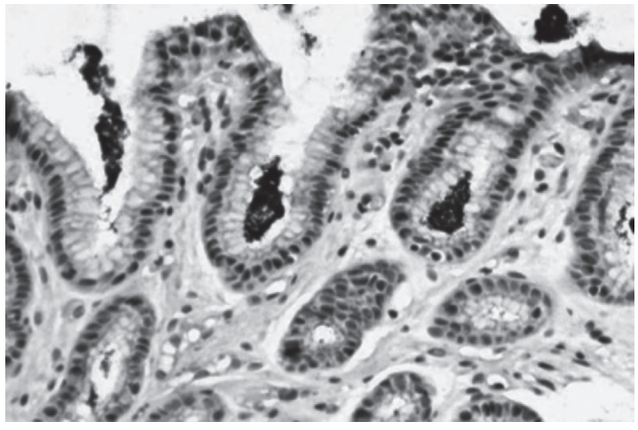

Figure 1. Immunohistochemical analysis: Muc2 expression in mucus overlapped in mucosa and in the gaps of single antral foveae in atrophic gastritis, 100x magnification

fibroblastic cells and lymphocytes dominated over all other cellular forms, programmed differentiation characterized by immature fibroblastic cells (p=0.018) with weak protein-synthesizing function (Table I).

The reduced total number of mast cells was significant as well (p=0.026), with the number of mature forms decreasing, which was observed in the near epithelial localisation, though a small amount of immature forms was also present around vascularly. Among mature mast cells mainly partially degranulated cells located in extracellular matrix were determined.

In the atrophic manifestations in lamina propria of AM, there was a reduced number of microcirculatory vessels compared to group 1 (p=0.027). The vessels were generally determined in the subepithelial zone and between foveae, and in the area of glands there were single vessels. The observed vessels were dilated, without stasis and changed vascular wall, and endotheliocyte nuclei.

Morphological examination of AM in group 3 children showed the abnormality of regeneration processes accompanied by the generative layer thinning-the reduction in the total number of gastric foveae and the appearance of intestinal type foveae (Figure 3).

In addition, intestinal foveae were determined in the form of single focal spots in 8 of the 11 cases of the 8-year-old children, in whom there were 1-3 intestinal foveae per 14 normal foveae. In 3 of the 11 cases (in children over 13 years) foveae of intestinal type occupied the most mucosa area, and there were 4-6 intestinal foveae per 10 gastric foveae that indicated multifocal IM. The epithelium of the foveae contained very few goblet cells of medium size, and single Paneth cells. The lumen of most of the cells was dilated, and the determined foveolar epithelium had dystrophic features and irregular mucoid production in the cells. Histochemical analysis of the histologic specimens of group 3 children showed a general reduced zone of PAS-positive staining of granulocytes and superficial-foveolar mucous cells with decreased intensity in comparison with group 1 (p=0.039), and with group 2 (p=0.048). Few goblet exocrinocytes of metaplastic epithelium contained mainly PAS-positive secretion, and in some of them acid mucins were determined. In the presence of such characteristics, more expressed structural changes were observed both in an epithelial layer itself, and in the PMP in the form of deformed foveae with flattened epithelium and apically located nuclei of epitheliocytes.

The observed PAS-positive uniformity in the cytoplasm sections of all types of epithelium, including metaplastic (PAS-positive secretion of goblet cells), is a positive sign since it indicated the preservation of the functional properties characteristic of intact superficial-foveolar epithelium.

The study of proliferative activity of the superficial-foveolar epithelium in IM foci showed the revealed proliferation level to be significantly higher than in the comparison group (p=0.019). The comparison of Ki-67-expression in gastric nonmetaplastic and metaplastic epithelium within one biopsy showed no significant changes (p=0.071).

Table I. Morphometric indices of atrial mucosa in children with atrophic gastritis

Parameters Median		Nonatrophic gastritis without IM		Atrophic gastritis with IM			
		Min-Max	Median	Min-Max	Median	Min-Max	
Proliferative activity of patching surface epithelium	Nonmetaplastic epithelium	22	17-28	28*	23-32	27.0*	23-32
	Metaplastic epithelium in focal manifestation	-	-	-	-	26.0*	23-30
	Metaplastic epithelium in multifocal manifestation	-	-	-	-	25.0	22-29
Height of surface epithelium, μm		30.0	26-33	28.75	25.5-33	28.75	24.0-31.5
Infiltrate cell density, mm^2		11155	9891-12426	8507*	7731-9445	8278*	6936-8932
Infiltrate lymphocytes mm^2		4304	3027-5003	2917*	2536-3365	2795*	2175-3104
Infiltrate plasma cells mm^2		2329	1754-3102	1228*	884-1361	783*,**	621-885
Stroma fibroblasts immature mm^2		435	336-609	771*	516-854	895*,**	641-1009
Stroma fibroblasts mature mm^2		1257	932-1385	1359	975-1463	1527*	1288-1641
Stroma fibrocytes, mm^2		1237	983-1381	1329*	987-1386	1508*	1354-1632
Stroma macrophages, mm^2		489	411-532	386*	284-442	362*	239-437
Stroma mast cells, mm^2		290	202-347	228*	143-285	143*,**	86-182
Infiltrate eosinophils, mm^2		471	321-518	189*	94-266	174*	83-263
Infiltrate neutrophils, mm^2		249	146-278	78*	31-108	73*	39-158

*Indices with revealed significant differences compared to the comparison group, when p<0.05
**Indices with revealed significant differences compared to the group of atrophic gastritis without intestinal metaplasia, when p<0.05
IM: Intestinal metaplasia, Min: Minimum, Max: Maximum

Morphometric study of AM biopsies of group 3 (Table I) patients revealed significantly reduced indices of infiltrate cell density compared to those of both group 1 and group 2 (p=0.016 and p=0.035, respectively). The feature of AM cell composition in the presence of IM areas was the increased number of fibroblastic cells, to a greater degree due to immature forms (p=0.014). The number of mature fibroblasts was significantly increased compared to the comparison group only (p=0.032). Mucosa interfoveolar space showed a growth in the number of fibrocytes compared to the comparison group (p=0.027) as well as group 2 (p=0.034). It should be noted that the fibroblasts and fibrocytes localized paraepithelially were singly determined, therefore, their influence on the preservation of the basal membrane structure of the epithelium of foveae and glands appeared to be minimal. Also observed was the absence of macrophage-fibroblast interaction, and significant reduction in the number of macrophages in AM in the atrophic gastritis patients with IM compared to the comparison group (p=0.019) and group 1 (p=0.028), which indicated their insufficient production of extracellular matrix components. Cells with lytic effect in the form of eosinophils and neutrophils were singly determined.

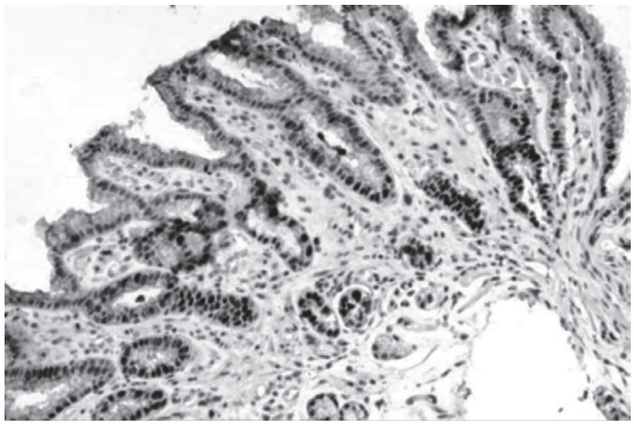

Figure 2. Immunohistochemical analysis: Ki-67 antigen exposure by foveolar epithelium of antral mucosa in atrophic gastritis, 100x magnification

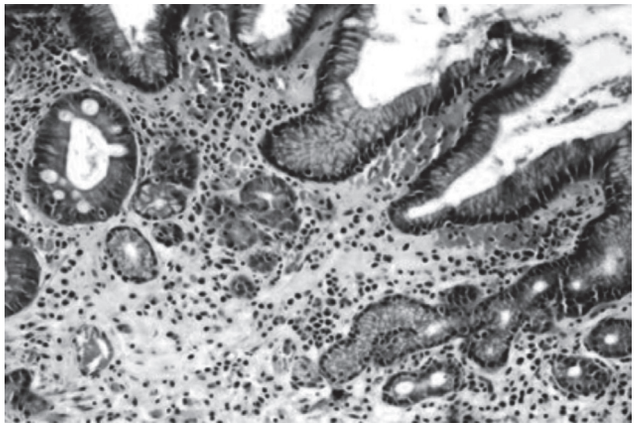

Figure 3. Chronic atrophic gastritis of antrum with intestinal metaplastic lesions of superficial-foveolar epithelium. Hematoxylin and eosin staining, 100x magnification

The analysis of mast cell population showed significant reduction in the total number of mast cells in the lamina propria of AM in group 2 children (p=0.038). Moreover, the number of maximally degranulated and degrading forms of mast cells was minimal (8.5%), and the absence of those paraepithelially located indicated their insufficient functional activity in producing the components of basal membrane, particularly, heparin sulfate proteoglycan.

Discussion

Atrophic antrum-gastritis in children was characterized not only by the alteration of mucosal cytoarchitecture (the reduced volume of foveae and glands), but also by the reduced number of functionally mature cell forms responsible for the production of extracellular matrix components and basal membranes, the absence of which in paraepithelial localization results in the change of specific properties of basal membranes and, subsequently, the reduction of environmental resistance of epithelium. Appearance of epithelial zones with uncharacteristic signs: synthesis of sulphomucins and the shift of the proliferation compartment to the zone of the bottom of glands in atrophic gastritis can indicate further possible appearance of metaplastic transformation foci just in these areas. In atrophic gastritis with IM, the progression of the disturbances of intercellular and tissue-like interactions in AM occurs. Other revealed conditions in the environment of epitheliocytes seem to impose further growth of changes in the algorithm of intercellular existence, switching a cell synthetic potential to the production of proteins uncharacteristic of this type of epithelium that eventually results in the formation of a phenotype of a cell differentiated in another direction, strengthening the formation of intestinal metaplasia and changing its focal nature into multifocal.

Conclusion

Morphological changes in AM traced over a period of several years in the same patients in childhood and adolescence make it possible to conclude that the presence of frequently recurrent inflammatory process can result in persistent structural changes until atrophy occurs. However, the increase in the total number of fibroblastic cells with immature forms prevailing, and adequate state of microvasculature suggest an irreversible atrophic process in childhood.

Ethics

Ethics Committee Approval: The study was approved by the Ethics Committee of our Research Institute (Approval number: 2007/28), Informed Consent: Consent form was filled out by all participants.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: Natalia Yurievna Shirokova, Design: Diana Anatolyevna Davydova, Data Collection or Processing: Natalia Yurievna Orlinskaya, Analysis or Interpretation: Natalia Yurievna Shirokova, Diana Anatolyevna Davydova, Literature Search: Natalia Yurievna Orlinskaya, Writing: Natalia Yurievna Shirokova.

Conflict of Interest: No conflict of interest was declared by the authors.

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References

1. Dohil R, Hassall E, Jevon G, Dimmick J. J Pediatr Gastroenterol Nutr 1999;29:378-94.
2. Brigic E, Hadzic D, Mladina N. Childhood and Coress model of carcinogenesis. Med Arch 2012;66:375-7.
3. Guarner J, Bartlett J, Whistler T, et al. Can pre-neoplastic lesions be detected in gastric biopsies of children with Helicobacter pylori infection? J Pediatr Gastroenterol Nutr 2003;37:309-14.
4. Correa P. The biological model of gastric carcinogenesis. IARC Sci Publ 2004;301-10.
5. Ricuarte O, Gutierrez O, Cardona H, Kim JG, Graham DY, El-Zimaity HM. Atrophic gastritis in young children and adolescents. J Clin Pathol 2005;58:1189-93.
6. Hassall E, Dimmick JE. Unique features of Helicobacter pylori disease in children. Dig Dis Sci 1991;36:417-23.
7. Satarkar RP, Sawant P, Nanivadekar S, Shroff C. Helicobacter pylori and intestinal metaplasia of gastric mucosa. Indian J Gastroenterol 1997;16:16-7.
8. Ierardi E, Francavilla R, Panella C. Effect of Helicobacter pylori eradication on intestinal metaplasia and gastric epithelium proliferation. Ital J Gastroenterol Hepatol 1997;29:470-5.
9. Ohkuma K, Okada M, Murayama H, et al. Association of Helicobacter pylori infection with atrophic gastritis and intestinal metaplasia. J Gastroenterol Hepatol 2000;15:1105-12.
10. Brittan M, Wright NA. Gastrointestinal stem cells. J Pathol 2002;197:492-509.
11. Ruiz B, Garay J, Correa P, et al. sMorphometric evaluation of gastric antral atrophy; improvement after cure of Helicobacter pylori infection. Am J Gastroenterol 2001;96:3281-7.
12. Yeh LY, Raj M, Hassan S, et al. Chronic atrophic antral gastritis and risk of metaplasia and dysplasia in an area with low prevalence of Helicobacter. Indian J Gastroenterol 2009;28:49-52.
13. Reis CA, David L, Correa P, et al. Intestinal metaplasia of human stomach displays distinct patterns of mucin (MUC1, MUC2, MUC5AC, and MUC6) expression. Cancer Res 1999;59:1003-7.
14. Jass JR, Filipe MI. The mucin profiles of normal gastric mucosa, intestinal metaplasia and its variants and gastric carcinoma. Histochem J 1981;13:931-9.
15. Smith JL, Dixon MF. Is subtyping of intestinal metaplasia in the upper gastrointestinal tract a worthwhile exercise? An evaluation of current mucin histochemical stains. Br J Biomed Sci 2003;60:180-6.
16. Dinis-Ribeiro M, Lopes C, da Costa-Pereira A, et al. A follow up model for patients with atrophic chronic gastritis and intestinal metaplasia. J Clin Pathol 2004;57:177-82.
17. Kapadia CR. Gastric atrophy, metaplasia and dysplasia: a clinical perspective. J Clin Gastroenterol 2003;36(5 Suppl):S29-36.
18. El-Zimaity H. Gastritis and gastric atrophy. Curr Opin Gastroenterol 2008;24:682-6.
19. Unger Z, Pronai L, Szaleczky E, Zagoni T, Molnar B, Tulassay Z. Effect of Helicobacter pylori infection and eradication on proliferative kinetics of the gastric mucosa. Orv Hetil 2000;141:2695-700.
20. Unger Z, Molnar B, Szaleczky E, et al. Effect of Helicobacter pylori infection and eradication on gastric epithelial cell proliferation and apoptosis. J Physiol Paris 2001;95:355-60.
21. Lerardi E, Francavilla A, Balzano T, et al. Effect of Helicobacter pylori eradication on gastric epithelial proliferation. Relationship with ras oncogene p21 expression. Ital J Gastroenterol Hepatol 1997;29:214-9.
22. Gillett P, Hassall E. Pediatric gastrointestinal mucosal biopsy. Special considerations in children. Gastrointest Endosc Clin N Am 2000;10:669-712, vi-vii.
23. Fichman S, Niv Y. Histological changes in the gastric mucosa after Helicobacter pylori eradication. Eur J Gastroenterol Hepatol 2004;16:1183-8.
24. Ozturk Y, Ozer E, Lebe B, Bekem O, Buyukgebiz B. Immunohistochemical evaluation of p53 expression and proliferative activity in children with Helicobacter pylori associated gastritis. J Pediatr Gastroenterol Nutr 2005;40:467-70.
25. Micu G, Staniceanu F, Zurac S, et al. Regression of precancerous epithelial alteration in patients with Helicobacter pylori chronic gastritis. Rom J Intern Med 2010;48:89-99.
26. Rugge M, de Boni M, Pennelli G, et al. Gastritis OLGA-staging and gastric cancer risk: a twelve year clinico-pathological follow-up. Aliment Pharmacol Ther 2010;31:1104-11.
27. Ramirez-Mendoza P, Gonzalez-Angulo J, Angeles-Garay U, Segovia-Cueva GA. Evaluation of Gastritis Atrophy. Comparison Between Sidney and OLGA Systems. Rev Med Inst Mex Seguro Soc 2008;46:135-9.
28. Rugge M, Kim JG, Mahachai V, et al. OLGA gastritis staging in Young Adults and country specific gastric cancer risk. Int J Surg Pathol 2008;16:150-4.
29. Naish SJ. Immunochemical staining methods. DAKO Corporation. 1989, 41.
30. Dabbls DJ. (Ed) Diagnostic immunohistochemistry. Edinburg: Churchill Livingstone, 2002, 673.

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The Effect of Vitamin D Deficiency on the Severity of Bronchiolitis in Infants

Süt Çocuklarında Vitamin D Eksikliğinin Bronşiolit Ciddiyeti Üzerine Etkisi

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ABSTRACT

Aim: This study was conducted to investigate the effect of vitamin D deficiency on the severity of bronchiolitis in infants who were hospitalized for treatment.

Materials and Methods: The infants who were followed up in our hospital and monitored due to acute bronchiolitis within a 0 to 24-month period were included in the study. The cases were evaluated by the clinical characteristics scoring system and divided into two groups (mild and moderate-severe). A questionnaire filled in by the mothers of the children to question the factors associated with vitamin D. The vitamin D levels of the cases, their clinical histories, and histories of taking vitamin D were examined.

Results: The number of patients with a vitamin D level <20 ng/mL in the moderate-severe disease group was higher than the number in the mild disease severity group ($p<0.05$). No difference was detected in the number of patients who received vitamin D prophylaxis between the moderate-severe and mild disease severity groups ($p>0.05$). The number of patients with vitamin D level <20 ng/mL was higher in the hospitalized group than in the non-hospitalized group ($p<0.05$). There was no difference in the number of patients that received vitamin D prophylaxis between the hospitalized and the non-hospitalized groups ($p>0.05$).

Conclusion: Vitamin D deficiency in infants is associated with increased severity of bronchiolitis and hospitalization.

Keywords: Bronchiolitis, vitamin D deficiency, infant

ÖZ

Amaç: Bu çalışma, bronşiyolit nedeniyle hastaneye yatırılarak tedavi edilen süt çocuklarında vitamin D eksikliğinin bronşiyolit ciddiyeti üzerine etkisinin araştırılması amacıyla yapılmıştır.

Gereç ve Yöntemler: Hastanemizde 0 ile 24 aylık dönem içinde akut bronşiyolit nedeniyle izlenen süt çocukları çalışmaya dahil edilmiştir. Olgular klinik özellikleri skorlama sistemi ile değerlendirilerek iki gruba (hafif ve orta-ağır) ayrılmış, olguların annelerine D vitamini ile ilişkili faktörleri sorgulayan anket formu uygulanmıştır. Olguların klinik öyküleri, D vitamini alma öyküleri ve D vitamini düzeyleri incelenmiştir.

Bulgular: Orta ve ağır derecedeki hastalık grubunda D vitamini düzeyi <20 ng/mL olan hastaların sayısı hafif şiddette hastalık grubundakilerin sayısından daha yüksekti ($p<0,05$). Hafif derecedeki hasta grubu ile orta-ağır derecedeki hasta grubunda D vitamini profilaksi alan hastaların sayısı arasında anlamlı farklılık saptanmadı ($p>0,05$). Hastaneye yatırılan hastalar arasında D vitamini düzeyi <20 ng/mL olan hastaların sayısı hastaneye yatırılmayan hastalara göre daha yüksekti ($p<0,05$). D vitamini profilaksisi alan hastaların sayısı açısından hastaneye yatırılan grup ile hastaneye yatırılmayan grup arasında anlamlı fark saptanmadı ($p>0,05$).

Sonuç: Süt çocuklarında D vitamini eksikliği, artmış bronşiyolit şiddeti ve hastaneye yatış ile ilişkilidir.

Anahtar Kelimeler: Bronşiyolit, D vitamini eksikliği, süt çocuğu

Introduction

Recently, the harmful effects of pathogens that cause bronchiolitis have strengthened, possibly due to increasing rates of vitamin D deficiency within infants. Vitamin D has been demonstrated to play roles in both immune system activation and the prevention of infections by microorganisms. An increased risk of respiratory system

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diseases has been demonstrated in the first three months in infants with low vitamin D levels in the cord blood at birth (1). Recent studies have defined vitamin D deficiency as a serum 25-hydroxyvitamin D [25(OH)D] level equal to or less than 20 ng/mL, whereas vitamin D insufficiency is defined as a serum level between 21 and 29 ng/mL. The lowest 25(OH) D level that will not activate parathormone is 30 ng/mL (75 nmol/L); therefore, a sufficient vitamin D level is accepted as >30 ng/mL (75 nmol/L) (2-4). Vitamin D effects susceptibility to infections that trigger wheezing at an early age and the response to this process. The relationship between serum vitamin D levels and respiratory tract infections was shown in the National Health and Nutrition Examination Survey (NHANES) III study, where this relationship was more evident in patients with asthma and chronic obstructive pulmonary disease (5). In another population-based study, a strong relationship was found between vitamin D serum levels and forced expiratory volumes in 1 second (6). Respiratory tract infections are frequent causes of asthma exacerbation among children and adults, and are associated with low vitamin D levels (7). The most frequently reported factors that result in exacerbation are rhinoviruses and corona viruses (8). In a recent study, vitamin D serum levels were shown to be inversely related to the total immunoglobulin E concentration and eosinophil numbers, and hospitalizations were rarer among patients with high vitamin D levels during the preceding one year period. The relationship between a high vitamin D level and a decrease in the need for anti-inflammatory treatment was shown in the same study (9).

Materials and Methods

Study Population

This single center prospective study was conducted at Bağcılar Training and Research Hospital, Clinic of Pediatrics in İstanbul, Turkey between January and April 2015.

Bronchiolitis was diagnosed in children aged 1-24 months in accordance with the study's inclusion criteria; a total of 102 children receiving treatment at the hospital were included. Children with known chronic lung disease, heart disease, recurrent lower respiratory tract infection, growth retardation, and a history of premature birth were excluded. The diagnosis of bronchiolitis was made during the medical examination of the patients, with regard to complaints of coughing, and respiratory distress alongside clinical findings such as rales and rhonchi.

The patient data were obtained from the history and file records. The history of bronchiolitis, history of hospitalization, use and duration of vitamin D prophylaxis in infancy were recorded according to the information obtained from the mother. The assessment of disease severity was determined in accordance with the Turkish Thoracic Society guidelines for acute bronchiolitis (10). The patients were divided into two groups (mild and moderate-severe) according to their clinical conditions. The classification was based on the general condition of the patient, breaths per minute, the presence of apnea, pulse rate, oxygen saturation and the existence of chest retraction (Table I).

The questionnaire forms were prepared. The form was filled in according to the definitions of the mothers during examination. The results of these questionnaire forms were given in Table II. Infants who had used vitamin D or vitamin preparations containing vitamin D for more than six months were accepted to have used vitamin D regularly.

Table I. Assessment of bronchiolitis severity

	Mild	Moderate	Severe
Apnea	-	-	+
Breaths/minute	<50	50-70	>70
Pulse/minute	<140	140-160	>160
Retraction	Mild	Moderate	Severe
SaO_2	>93%	86-92%	<85%
Cyanosis	-	-	+

SaO_2: Arterial blood oxygen saturation

Laboratory Measurements

Blood samples were taken from all the infants to study the serum vitamin D levels. The blood was centrifuged for 5 minutes at 3000 rpm, and the serum was separated. Each serum sample was covered with dark carbon paper to prevent light permeation and then frozen and stored at -30 degrees C prior to the analysis. The vitamin D levels were examined using the electro chemiluminescence enzyme immuno assay method (ADVIA Centaur, USADPC Co., USA).

The study protocol was approved by the Research Ethics Committee of Bağcılar Training and Research Hospital in accordance with the Declaration of Helsinki (approval number: 2014-300). Informed consent was obtained from the parents of all study participants.

Statistical Analysis

The mean, standard deviation, median lowest and highest values, frequency and ratio values were used as the descriptive statistics of the data. The distribution of the variables was measured using the Kolmogorov-Smirnov test. The Mann-Whitney U test was used in the analysis of the qualitative data together with the chi-square; when the chi-square test conditions were not met, Fisher's test was used. The Spearman correlation analysis was used to assess correlations.

Results

The demographic data from the infants included in the study are presented in Table II. No significant difference in disease severity was detected between the groups in terms of gender and age distribution. The vitamin D concentration

was significantly lower in the moderate-severe group. The number of patients with vitamin D concentrations <20 ng/mL in the moderate-severe disease group was higher than the number in the mild disease severity group. No difference was detected in the number of patients who received vitamin D prophylaxis between the moderate-severe and mild disease severity groups (Table III).

The number of patients between 0 and 2 years of age was higher in the hospitalized group compared to the non-hospitalized group. There was no difference in the gender distribution between the hospitalized and non-hospitalized groups, but the vitamin D concentration was lower in the hospitalized group. The number of patients with vitamin D concentrations <20 ng/mL in the hospitalized group was higher than that in the non-hospitalized group. No difference was detected between the rates of patients that received vitamin D prophylaxis in the hospitalized and non-hospitalized groups. There was no significant correlation between the vitamin D level and the hospitalization durations and patient age. The gender distribution and age distribution were not significantly different between the moderate-severe and mild disease severity groups. However, the vitamin D concentration was significantly lower in the moderate-severe disease severity group ($p<0.05$). The number of patients that received vitamin D prophylaxis was not significantly different between these two groups. The vitamin D concentration was significantly lower in the hospitalized group than in the non-hospitalized group ($p<0.05$).

Table II. Demographic data of infants with bronchiolitis

Generally characteristics			
Age (mean ± standard deviation)	5.8±4.76 (year)		
Age	0-3 months	47	27.7%
	4-6 months	12	33.0%
	6-24 months	43	32.1%
Vitamin D level (ng/mL)	23.24±13.6		
Vitamin D (ng/mL)	0-5	10	9.8%
	5-10	14	12.5%
	10-15	6	6.3%
	15-20	11	10.7%
	20-25	17	17.0%
	>30	44	43.8%
Duration of hospitalization (days)	4.15±3.29		
Gender	Girl	44	40.0%
	Boy	58	58.0%
Hospitalization	Yes	75	67.0%
	No	37	33.0%
Bronchiolitis severity	Mild	58	56.9
	Moderate-severe	44	43.1
Vitamin D prophylaxis	Received	88	84.8%
	No received	14	15.2%

Discussion

Vitamin D plays a role in immune system activation, it helps to prevent infections caused by microorganisms. Studies have shown a relationship between the severity of bronchiolitis, increased hospitalization, and vitamin D deficiency (11). In this study, we investigated the relationship between vitamin D deficiency and bronchiolitis severity, and found that vitamin D levels are very important for infants with bronchiolitis.

Many studies have proposed that vitamin D concentration decreases due to the decrease in sunlight during the winter months, resulting in a related decrease in immune functions and sensitivity to influenza infections (12,13).

The study by Mansbach and Camargo (12) showed that the wheezing rate was increased in infants of mothers who had vitamin D deficiency during pregnancy. An increased risk of respiratory system diseases was observed in the first three months in infants with low vitamin D concentration in the umbilical cord blood (14).

In the hospital-based case control study performed by Wayse et al. (15), the authors concluded that insufficient breast milk intake and subclinical vitamin D deficiency in the first 4 months of life constituted a significant risk factor for severe lower respiratory tract diseases.

Infections are the most frequent causes of morbidity and mortality among children worldwide. The relationship between insufficient vitamin D levels and respiratory tract infections in children and/or hospitalization has been shown

Table III. Comparision of vitamin D level and severity of disease

		Moderate-severe		**Mild**		**p**
Age	0-3 months	26	39%	21	18.9%	0.07
	4-6 months	8	25%	4	39.6%	
	6-24 months	19	36%	24	41.5%	
Gender	Girl	19	43%	28	41%	0.609
	Boy	25	57%	30	59%	
Vitamin D (ng/mL)		15.5±11.4		26.0±11.8		**0.0001**
Vitamin D	0-5	8	20%	2	3%	**0.0001**
	5-10	11	20%	3	7%	
	10-15	4	11%	2	3%	
	15-20	3	9%	8	12%	
	20-25	4	14%	13	19%	
	>30	6	25%	38	56%	
Vitamin D prophylaxis	(+)	35	80%	53	91%	0.08
	(-)	9	20%	5	9%	

in many epidemiological studies. Najada et al. (16) also reported a high incidence of nutritional rickets among infants hospitalized due to respiratory tract diseases.

In this study, the rate of the patients between the ages 0 and 2 years was significantly higher in the hospitalized group than in the non-hospitalized group. The gender distribution did not exhibit significant differences between the hospitalized and non-hospitalized patients. The vitamin D concentration was significantly lower in the hospitalized group, but the number of patients with vitamin D <20 ng/mL in this group was significantly higher than in the non-hospitalized group. The number of patients that received vitamin D prophylaxis in the hospitalized and non-hospitalized groups was not significantly different.

According to the NHANES data, the frequency of upper respiratory tract infections in patients with serum vitamin D <25 nmol/L was much higher in patients with asthma compared to those without (5). The relationship between vitamin D and asthma has been known for a long time. Although the mechanism is not known, new studies have found an association between high vitamin D levels and better lung functions, lower respiratory tract hypersensitivity and increased glucocorticoid responses (17).

In a case-control study, the vitamin D level in newborns taken to the intensive care unit due to lower respiratory tract infections was demonstrated to be significantly lower compared to healthy infants in the same age group. Additionally, the vitamin D concentration of the mothers was lower compared to the control group (18).

The mechanism by which vitamin D affects the immune system is not known. Viral infections are observed more frequently than bacterial infections in patients with vitamin D deficiency. Therefore, the relationship between vitamin D and immune functions shows the importance of supplementation in the pediatric population (19). In another study performed by Karatekin et al. (20), vitamin D concentrations were examined in the sera of newborns that did not have rickets with a lower respiratory tract infection (LRTI) diagnosis and their mothers. Based on the results, the risk of damage due to LRTI was increased in newborns with a subclinical vitamin D deficiency. The strong positive correlation between the vitamin D concentrations of the newborns and their mothers showed that sufficient vitamin D supplementation should be emphasized during pregnancy in winter months (19,20).

A large number of studies have proposed that vitamin D reduces the risk of respiratory tract infections in children (13). In one study, a 13-fold higher risk was found in Ethiopian children younger than 5 years of age whose vitamin D deficiency was shown clinically (21). In a study performed in Yemen, a 50% vitamin D deficiency was found in children who were admitted (22).

The gender and age distributions were not significantly different in the moderate-severe and mild disease severity groups in this study, but the vitamin D concentration was significantly lower in the moderate-severe disease group. Also the number of patients with vitamin D level<20 ng/mL in this group was significantly higher. There was no significant difference in the number of patients who received vitamin D prophylaxis between the two groups.

Studies have demonstrated a role for vitamin D in the expression of the human cathelicidin antimicrobial peptide, which plays a role in host defense against respiratory tract pathogens. These studies showed that the risk of respiratory system infection was increased in subjects with vitamin D deficiency. This increased risk contributes to the increase in the wheezing incidence in children and the exacerbation of asthma in adults. Vitamin D plays a modulator role in regular T cell functions and interleukin-10 production. Therefore, epidemiological and randomized studies are needed in the future to better understand the role of vitamin D in respiratory infections and asthma (8). In conclusion, our study showed that the decreased vitamin D levels in infants with bronchiolitis were associated with bronchiolitis severity.

Ethics

Ethics Committee Approval: The study was approved by the Bağcılar Training and Research Hospital Local Ethics Committee (Approval number: 2014-300), Informed Consent: Consent form was filled out by all participants.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Meltem Erol, Özlem Bostan Gayret, Özgül Yiğit, Concept: Meltem Erol, Özlem Bostan Gayret, Design: Meltem Erol, Hüseyin Kaya, Şahin Hamilçıkan, Emrah Can, Data Collection or Processing: Hüseyin Kaya, Özgül Yiğit, Şahin Hamilçıkan, Analysis or Interpretation: Meltem Erol, Hüseyin Kaya, Emrah Can, Literature Search: Meltem Erol, Hüseyin Kaya, Özlem Bostan Gayret, Özgül Yiğit, Şahin Hamilçıkan, Writing: Meltem Erol.

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References

1. Meissner HC. Viral Bronchiolitis in Children. N Engl J Med 2016;37:62-72.
2. Holick MF. Vitamin D: a D-Lightful Health perspective. Nutr Rev 2008;66(10 Suppl 2):S182-94.
3. Dong Y, Pollock N, Stallman-Jorgensen IS, et al. Low 25-hydoxyvitamin D levels in adolescents: race, season, adiposity, physical activity, and fitness. Pediatrics 2010;125:1104-11.
4. Holick MF. The D-lemma: to screen or not to screen for 25-hydroxyvitamin D concentrations. Clin Chem 2010;56:729-31.
5. Searing DA, Leung DY. Vitamin D in atopic dermatitis, asthma and allergic diseases. Immunol Allergy Clin North Am 2010;30:397-409.
6. Burns JS, Dockery DW, Neas LM, et al. Low dietary nutrient intakes and respiratory health in adolescents. Chest 2007;132:238-45.

7. Ginde AA, Mansbach JM, Camargo CA Jr. Association between serum 25-hydroxyvitamin D level and upper respiratory tract infection in the Third National Health and Nutrition Examination Survey. Arch Intern Med 2009;169:384-90.
8. Nicholson KG, Kent J, Ireland DC. Respiratory viruses and exacerbations of asthma in adults. BMJ 1993;307:982-6.
9. Dougherty RH, Fahy JV. Acute exacerbations of asthma: epidemiology, biology and The exacerbation-prone phenotype. Clin Exp Allergy 2009;39:193-202.
10. Yalçın E, Karadağ B, Uzuner N, ve ark. Türk toraks derneği akut bronşiyolit tanı ve tedavi uzlaşı raporu. Türk Toraks Dergisi 2009;10;sup1:3-7.
11. Brehm JM, Celedon JC, Soto-Quiros ME, et al. Serum vitamin D levels and markers of severity of childhood asthma in Costa Rica. Am J Respir Crit Care Med 2009;179:765-71.
12. Mansbach JM, Camargo CA Jr. Respiratory viruses in bronchiolitis and their link to recurrent wheezing and asthma. Clin Lab Med 2009;29:741-55.
13. Cannell JJ, Zasloff M, Garland CF, Scragg R, Giovannucci E. On the epidemiology of influenza. Virol J 2008;5:29.
14. Hope-Simpson RE. The role of season in the epidemiology of influenza. J Hyg (Lond) 1981;86:35-47.
15. Wayse V, Yousafzai A, Mogale K, Filteau S. Association of subclinical vitamin D deficiency with severe acute lower respiratory infection in Indian children under 5 y. Eur J Clin Nutr 2004;58:563-7.
16. Najada AS, Habashneh MS, KhadeR M. Thefrequency of nutritional rickets among hospitalized infants and its relation to respiratory diseases. J Trop Pediatr 2004;50:364-8.
17. Walker VP, Modlin RL. The vitamin D connection to pediatric infections and immune function. Pediatr Res 2009;65:106R-13R.
18. Sutherland ER, Goleva E, Jackson LP, Stevens AD, Leung DY. Vitamin D levels, lung function, and steroid response in adult asthma. Am J Respir Crit Care Med 2010;181:699-704.
19. Kovacs CS. Vitamin D in pregnancy and lactation: maternal, fetal, and neonatal outcomes from human and animal studies. Am J Clin Nutr 2008;88:520S-8S.
20. Karatekin G, Kaya A, Salihoğlu O, Balci H, Nuhoğlu A. Association of subclinical vitamin D deficiency in newborns with acute lower respiratory infection and their mothers. Eur J Clin Nutr 2009;63:473-7.
21. Muhe L, Lulseged S, Mason KE, Simoes EA. Case control study of the role of nutritional rickets in the risk of developing pneumonia in Ethiopian children. Lancet 1997;349:1801-4.
22. Banajeh SM, al-Sunbali NN, al-Sanahani SH. Clinical characteristics and outcome of children aged under 5 years hospitalized with severe pneumonia in Yemen. Ann Trop Paediatr 1997;17:321-6.

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Original Article / Orijinal Araştırma

The Results of the Treatment of Osteogenesis Imperfecta with Corkscrew Tipped Telescopic Nail

Osteogenezis İmperfektalı Olgularda Tirbuşon Uçlu Teleskopik Çivi ile Tedavi Sonuçları

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ABSTRACT

Aim: We aimed to evaluate the clinical and radiological results of an intramedullary fixation system used in surgeries for fractures and deformities of osteogenesis imperfecta where we applied a new design corkscrew tipped intramedullary nailing.

Materials and Methods: Twenty extremities of 14 osteogenesis cases, who underwent surgery and to whom corkscrew tipped intramedullary treatment was applied, were retrospectively scanned. Ambulation, discrepancies in the lenght of extremities, deformities and joint mobility range were all noted before the operation. Postoperative union rates, complications and our experience regarding the nail were also evaluated.

Results: Six tibia and 14 femurs were operated using corkscrew tipped telescopic nails. Two bones were operated due to non-union, while seven bones underwent surgery due to acute fractures and 11 bones due to deformities. All the bones were seen to have achieved the aimed union. No major complications were observed. Infection was present in two cases.

Conclusion: Corkscrew tipped telescopic nail is a safe and effective method of fixation in patients with osteogenesis imperfecta.

Keywords: Osteogenesis imperfecta, osteotomy, fracture healing

ÖZ

Amaç: Kırık ve deformiteler nedeni ile cerrahi olarak tirbuşon uçlu kanal içi çivileme yöntemi uyguladığımız osteogenezis imperfektalı olguların klinik ve radyolojik sonuçlarını değerlendirmek istedik.

Gereç ve Yöntemler: Tirbuşon uçlu kanal içi çivi uygulanarak cerrahi yapılan 14 osteogenezisli olgunun 20 ekstremitesi geriye yönelik olarak tarandı. Operasyon öncesindeki ambulasyon durumu, ekstremite boyları, deformiteler, eklem hareket genişlikleri not edildi. Operasyon sonrası kaynama oranları, komplikasyonlar ve çiviye ait deneyimlerimiz değerlendirildi.

Bulgular: Altı tibia ve 14 femur, tirbuşon uçlu teleskopik çivi ile opere edildi. İkisi kaynamama, yedi kemikte akut kırık ve 11 kemik deformite nedeniyle opere edildi. Tüm kemiklerde kaynama elde edildi. Majör komplikasyon gözlenmedi. İki olguda enfeksiyon görüldü.

Sonuç: Tirbuşon uçlu teleskopik çivi osteogenezis imperfektalı olgularda güvenli ve etkin uygulanabilecek bir yöntemdir.

Anahtar Kelimeler: Osteogenezis imperfekta, osteotomi, kırık iyileşmesi

Introduction

Osteogenesis imperfecta (OI) is an autosomal dominant or recessive connective tissue disorder caused by the deficiency of Type I collagen production associated with the deficiency of collagen Type I alpha 1 chain and collagen Type I alpha 2 chain. This disease causes problems in all tissues that contain Type 1 collagen. In addition to many systemic problems such as blue sclerosis, otosclerosis, cardiac diseases, elasticity in the joint and thinning of the fascia, it also causes the loss of the normal ossification of the endochondrial bone (1). This results in easily fragile bones.

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Although recurrent fractures show spontaneous healing trait, they lead to increased deformities developing with age (2).

In OI cases, osteosynthesis performed with intramedullary nailing is the gold standard for surgical treatment to be administered following fracture and deformity. Intramedullary nailing, together with its corrective effect on deformity, is also effective in preventing recurrent fractures. Intramedullary nailing should be performed in a way that does not hinder the growth of children and should minimize the need for repetitive surgery as the child grows.

There are a limited number of intramedullary nail types designed for this purpose and research is underway for the ideal nail. Today, telescopic nails with elastic or extendable features are frequently resorted. With the elastic nails, there was a problem of migration (retraction) over time, while the problem encountered in telescopic nails was the need to open the joint (arthrotomy) during fixation to prevent migration, which posed another challenge (3-5).

Surgical procedures performed by opening the joint pose a risk for infection development, limitation of movement and early arthrosis. In our clinic, we applied corkscrew tipped telescopic rods, which are extendable and designed in our country, to 20 extremities of 14 osteogenesis imperfecta cases operated due to fractures, failure of union, or deformity in long bones. What makes it different from the commonly resorted other nails is the fact that its distal tip is curled like a corkscrew. Thanks to these curls, the movement of the nail in the bone is prevented. Arthrotomy is not required to immobilize the distal part and no additional incision is needed.

We aimed to observe the clinical and radiological efficacy of the telescopic corkscrew tipped nail, developed in our country and started to be used in practice in long bone fractures due to osteogenesis and deformities. Ambulation, discrepancies in the lenght of extremities, deformities and joint mobility range were all noted before the operation. Postoperative union rates and our experience regarding complications and nail complication were also evaluated.

Materials and Methods

Twenty extremities of 14 OI cases to whom corkscrew tipped telescopic nail technique was applied, between the years 2014-2016 were retrospectively evaluated. In addition to clinical and radiological follow-ups, union rates, union times, complications, and nail loosening or failures, if any, were noted.

All cases were placed in the supine position under general anesthesia and the nail entry site was prepared by opening from the proximal of the bone tissue to be nailed. First of all, bone osteotomy was applied to the sites where the deformities were located by inserting thin nails (Figure 1). When thin nails were advanced and reached the epiphysis line, 1-2 pieces of Kirchner wires and the distal end of the nail were fixed. The entry site was fixed by placing the thick nail over the thin one after obtaining the proper length and cutting it in the bone.

All cases underwent casting after surgery. Ambulatory patients were allowed to bear weight after their casts were removed starting from week 6. Rehabilitation was started after the 6th week for non-ambulatory patients.

Results

Of the cases operated, five were female, nine were male. The mean age was 6.92±3.04 (minimum: 3-maximum: 13). Six of the nails were applied to the tibia, and 14 to the femur. The mean follow-up time was 23.42±5.43 months. Nailings were performed due to 2 non-unions, 7 acute fractures and 11 deformity problems. Elastic nails of the old operation were present in 6 of the cases operated due to deformity and first these were removed during the surgery. In all cases, it was observed that there was improvement in the deformities after the operation. Radiological and clinical union was achieved in all of the bones. There was no delayed union or non-union. Infection was observed in 2 of our patients, one of which was superficial. Debridement and antibiotherapy were applied to the patient with the deep infection after implant removal (Table I). Bending, loosening or migration were not observed in the nails at all. It was noticed in 2 of the cases that the tip of the nail had perforated the bone cortex and protruded out of the bone, which was fixed with additional osteotomies during the operation.

Discussion

Repeated fractures and associated severe deformities in children with OI make orthopedic surgery inevitable (Figure 2). Surgical treatment protocols in these cases have recently become clear. The type of treatment regarded as gold standard is intracanal (intramedullary) nailing (6). However, the characteristics of the nail applied in intramedullary treatment might differ from those in adults in that the growth in children continues and the bone quality in osteogenesis cases is low in addition to the fact that the channel diameter

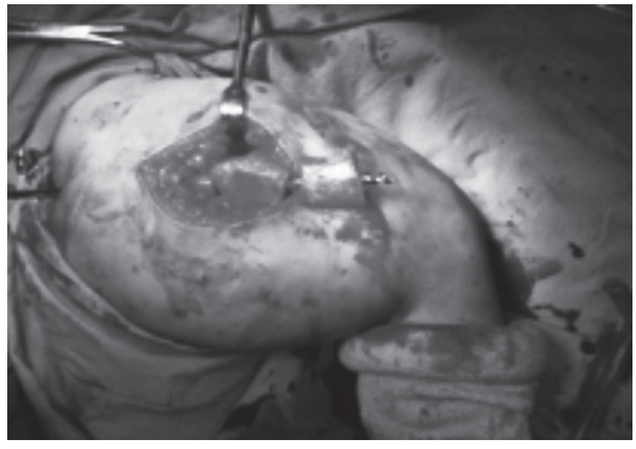

Figure 1. Osteogenesis imperfecta case of a 3-year-old. A 3-level osteotomy was applied to correct the deformity of the femur. Appearance during osteosynthesis with corkscrew-tipped nail and operation

is too narrow. For this purpose, it is necessary for the nails to be small in diameter so that they will give minimum damage to the epiphyses and extend together with the bone as the child grows older. The use of nails without extension properties may require repeated surgeries (Figure 3). To reduce the number of recurrent surgeries to a minimum, telescopic nails are used in childhood. These nails have great advantages compared to other nails in terms of reducing the risk of recurrent fractures, having extension features, allowing the prevention of new deformities.

Disadvantages can be listed as being weak for carrying the mechanical load due to narrow nail diameter, damage to the growth nuclei and damage to the joint by performing open surgery on neighboring joints for locking operations at both ends to prevent the movement of the nail in the bone (7).

The aim of all new nails being developed is to remove the existing disadvantages. In order to eliminate the disadvantages of open joint surgery, corkscrew tipped telescopic nails were developed by İnan et al. (8) and early results were reported.

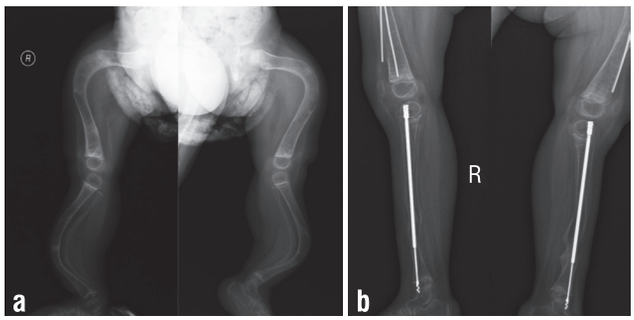

Figure 2. a) The X-rays of the gross deformities of the anteroposterior and lateral of the bilateral femur and tibia, b) Correcting the deformities of the tibia with telescopic nails

Table I. Demographic and surgical data of our cases diagnosed and operated due to osteogenesis imperfecta

Case (initials)	Age	Follow-up (month)	Ambulation	Operation site	Complication
E.B.	3	24	Assisted	2 tibia + femur	
D.B.	5	29	N/A	Femur	Infection
M.E.	4	32	Independent	Femur + tibia	
B.D.	6	17	Assisted	Femur	
S.V.S.	7	26	Independent	Femur	
E.Ö.	11	13	Independent	Femur + tibia	
E.Y.	3	23	N/A	2 femur	
E.S.	8	30	Independent	Femur	
E.K.	6	24	Assisted	Tibia	
M.Ö.	13	25	N/A	2 femur	
B.G.	7	19	N/A	Femur	
R.D.	5	22	Independent	Femur	
R.D.	11	27	N/A	Tibia	
Y.C.	8	17	N/A	Femur	Infection

We also wanted to indicate the results and observations in our clinic.

High complication rates have been reported nearly in all of the nails applied in OI cases both for the patient and the applied nails. Zionts et al. (9) reported a total of 40 problems, with 17 major 23 minor in a series of 40 cases. In 10 cases they had to replace the implants. In these studies, they observed that major problems were more common in patients under 5 years of age, and recommended that surgical applications be performed on patients over 5 years old.

In our cases, three were under 5 years of age, and no complications have been observed in the follow-up so far. Cho et al. (10) reported that nail breakage was frequently observed as the child grew up, and that there was a mechanical failure in long thin nails in their telescopic nail applications performed on 72 osteogenesis cases with a mean follow-up of 7 years. We have not encountered failure or loosening of the implants, which might be due to a relatively short follow-up period (average 2 years) in our applications. Postoperative infection was seen in 2 cases, one of which was superficial. In the cases with OI, the skin is thin, fragile and weak. That's why the skin is prone to pathologic conditions and infections (11). In one of 14 patients (7%), infection was observed at the wound site, which might call for systemic antibiotherapy and debridement (*Staphylococcus aureus*). Elastic nails used in childhood fractures and which have no extension properties may cause problems such as delayed union or non-union as they do not provide absolute stability in the fracture line. Gamble et al. (12) reported non-union in 10 of 52 OI cases. It was also observed that the non-union rate was higher in the fractures where deformity was most severe. With the deformity corrected, union was achieved following

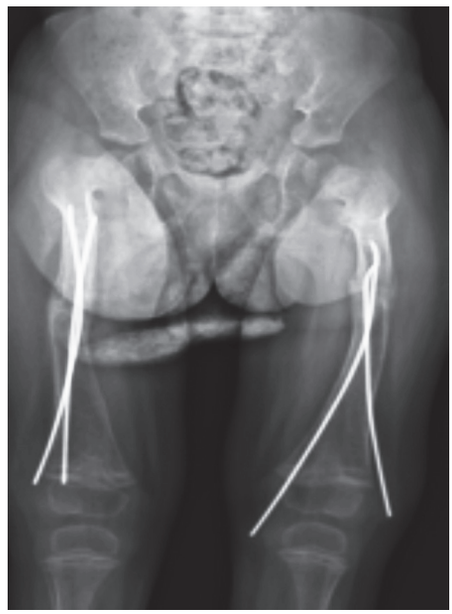

Figure 3. Radiological image of relaxation and migration of the previously applied elastic nails for correcting the bilateral femoral deformity after one year

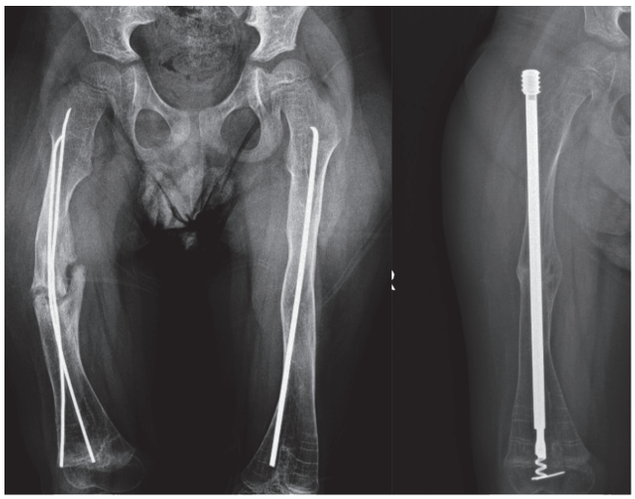

Figure 4. Second month image of the 7-year-old to whom corkscrew-tipped telescopic nail was applied due to non-union in the femur

intramedullary nailing application (12). Union occured in two of our cases to whom elastic nails were applied. We observed that in these two cases where the elastic nails were removed and telescopic nails inserted at the same time, union occured in 6 weeks (Figure 4).

In our clinic, corkscrew tipped nails have been used in the osteogenesis cases that meet operation indications since 2014. The simplicity of surgery and the fact that radiation exposure during surgery is less than other applications is a positive aspect we have observed in these applications. Sharpness of the tip of the nail is a negative feature we have observed during the operations as the nail could protrude by piercing the cortex in weaker bones, which must be carefully considered during surgery.

Conclusion

Corkscrew tipped telescopic nail application is a method that can be applied safely in OI cases. Necessity to open the joint to prevent the movement of the nail and the need for artrotomy for previously applied nails have disappeared with this method.

Ethics

Ethics Committee Approval: Retrospective study, Informed Consent: Consent form was filled out by all participants.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Hüseyin Günay, Concept: Muharrem İnan, Design: Muharrem İnan, Data Collection or Processing: Hüseyin Günay, Levent Küçük, Analysis or Interpretation: Hüseyin Günay, Levent Küçük, Literature Search: Hüseyin Günay, Levent Küçük, Writing: Hüseyin Günay.

Conflict of Interest: No conflict of interest was declared by the authors.

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References

1. Forlino A, Marini JC. Osteogenesis imperfecta. Lancet 2016;387:1657-71.
2. Forlino A, Cabral WA, Barnes AM, Marini JC. New perspectives on osteogenesis imperfecta. Nat Rev Endocrinol 2011;7:540-57.
3. Cho TJ, Choi IH, Chung CY, Yoo WJ, Lee KS, Lee DY. Interlocking telescopic rod for patients with osteogenesis imperfecta. J Bone Joint Surg Am 2007;89:1028-35.
4. Göçer H, Ulusoy S, Çıraklı A, Kaya İ, Dabak N. Osteogenezis İmperfektalı Hastada İntramedüller Çivi ile Kırık ve Deformite Tedavisi. Med Bull Haseki 2014;52:126-9.
5. Lee K, Park MS, Yoo WJ, Chung CY, Choi IH, Cho TJ. Proximal migration of femoral telescopic rod in children with osteogenesis imperfecta. J Pediatr Orthop 2015;35:178-84.
6. Reing CM. Report on new types of intramedullary rods and treatment effectiveness data for selection of intramedullary rodding in osteogenesis imperfecta. Connect Tissue Res 1995;31:77-9.
7. Nicolaou N, Bowe JD, Wilkinson JM, Fernandes JA, Bell MJ. Use of the Sheffield telescopic intramedullary rod system for the management of osteogenesis imperfecta: clinical outcomes at an average follow-up of nineteen years. J Bone Joint Surg Am 2011;93:1994-2000.
8. İnan M, Sarıkaya Aİ, Görgün B. Osteogenezis imperfekta tedavisinde tirbüşon uçlu teleskopik çivi kullanımı: Erken dönem sonuçlarımız. Acta Orthop Traumatol Turc Supp 2015 (SB 8-1):31.
9. Zionts LE, Ebramzadeh E, Stott NS. Complications in the use of the Bailey-Dubow extensible nail. Clin Orthop Relat Res 1998:186-95.
10. Cho TJ, Kim JB, Lee JW, et al. Fracture in long bones stabilised by telescopic intramedullary rods in patients with osteogenesis imperfecta. J Bone Joint Surg Br 2011;93:634-8.
11. Hansen B, Jemec GB. The mechanical properties of skin in osteogenesis imperfecta Arch Dermatol 2002;138:909-11.
12. Gamble JG, Rinsky LA, Strudwick J, Bleck EE. Non-union of fractures in children who have osteogenesis imperfecta. J Bone Joint Surg Am 1988;70:439-43.

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DOI: 10.4274/jpr.85547

Substance Use Among Adolescents, and Influencing Factors in Şanlıurfa

Şanlıurfa İlinde Adölesanlarda Madde Kullanma Durumu ve Etkileyen Faktörler

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ABSTRACT

Aim: The aim of this study was to investigate substance use by students in the 7-12th grades.

Materials and Methods: The research was conducted as a descriptive study in Şanlıurfa, which has a very young population, between September 2014 and March 2015. The research sample comprised 54.928 students in grades seven through twelve. The research data were obtained using a survey form prepared by the researchers according to the literature, and expert opinions were obtained from three specialists in the field before administering the survey. The data were evaluated using SPSS 17. Written permission from the relevant institutions and the verbal permission of the participants were obtained for conducting the study.

Results: Of the students, 51.6% were male, 23.8% were in ninth grade, 81.1% had 4 or more siblings, and 40.1% were the fourth or more in sibling order. While 23.2% were working when not at school; 6.3% smoked tobacco, 2.4% were using substances other than cigarettes, and 13.1% were acquainted with substance abusers. Those who said they used substances due to curiosity made up 31.3%, and 29.9% said they used substances to feel more powerful. The smoking and substance use rate was higher for male students, working students, students with extremely domineering families and students exposed to domestic violence.

Conclusion: These data obtained on substance use by students and the factors that affect it suggest that studies should be carried out on substance use prevention, family awareness should be increased and prevention policies should be developed and implemented.

Keywords: Adolescents, addiction, substance use

ÖZ

Amaç: Bu çalışma 7-12. sınıfta öğrenim gören öğrencilerde madde kullanma durumunun incelenmesi amacıyla yapılmıştır.

Gereç ve Yöntemler: Araştırma, nüfusunun çoğunluğunu çocuk ve gençlerin oluşturduğu Şanlıurfa ilinde Ekim 2014-Mart 2015 tarihleri arasında tanımlayıcı çalışma olarak yapılmıştır. Araştırmanın örneklemini, 7-12. sınıf 54,928 öğrenci oluşturmaktadır. Araştırmanın verileri literatür doğrultusunda araştırmacılar tarafından hazırlanan anket formu ile elde edilmiş, anketler uygulanmadan önce bu konuda çalışmaları olan üç uzmandan görüş alınmıştır. Veriler SPSS 17 programında değerlendirilmiştir. Çalışmanın yapılabilmesi için kurumlardan yazılı, katılımcılardan sözlü izin alınmıştır.

Bulgular: Öğrencilerin %51,6'sı erkek, %23,8'i 9. sınıf öğrencisi, %81,1'inin 4 ve daha fazla sayıda kardeşi bulunmakta, %40,1'i ailedeki çocuk sırasında dördüncü ve daha fazla sırada yer almaktadır. Öğrencilerin %23,2'si okul dışındaki zamanlarında bir işte çalışmaktadır. Öğrencilerin madde kullanma durumları incelendiğinde; %6,3'ü sigara, %2,4'ü sigara dışındaki diğer maddeleri kullandığını, %13,1'i çevresinde madde kullanan kişilerin olduğunu, %31,3'ü merak nedeniyle, %29,9'u kendini daha güçlü hissetmek için madde kullandığını belirtmiştir. Erkek olanlarda, çalışanlarda, baskıcı bir aileye sahip olanlarda ve şiddete maruz kalanlarda sigara ve diğer maddeleri kullanma oranı yüksektir.

Sonuç: Bu çalışmada öğrencilerin madde kullanma durumuna yönelik elde edilen veriler doğrultusunda; madde kullanımının önlenmesine yönelik tüm öğrencileri kapsayacak düzeyde çalışmaların yapılması, ailelerde farkındalığın artırılması, ülke düzeyinde önleme politikalarının geliştirilmesi önerilmektedir.

Anahtar Kelimeler: Adölesan, bağımlılık, madde kullanımı

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Introduction

As old as the history of humanity, substance use is among the most important problems around the world. The 2012 World Drug Report determined that around 153.000-300.000 people used illicit drugs (cannabis, opioids, cocaine, amphetamines, etc.) at a rate of 3.4-6.6% in 2010 (1). It was indicated in the 2012 World Health Organisation data that approximately 3.3 million people die due to alcohol use each year. These deaths constitute 5.9% of all deaths in the world. Alcohol causes more than 200 kinds of diseases and injuries (2).

Adolescents are in the risk group for substance abuse. Trying or using substances in childhood or adolescence possesses a potential risk of developing substance abuse in adulthood (3). Substance abuse in adolescence affects the future life, brain development, socialisation processes of the adolescent in a negative way. It is also closely associated with other risky behaviours and getting involved in crime (3).

Numerous studies have been performed to determine substance abuse rates in Turkey and around the world (4). Few of the studies carried out in other cities to determine substance abuse rates in Turkey include data for Şanlıurfa. The city of Şanlıurfa has the third highest child population among the cities of south eastern Turkey (5). No studies have been conducted with a comprehensive number of samples for the adolescent population of Şanlıurfa. This study was performed to examine substance abuse and the factors that affect substance abuse by students in grades seven through twelve in Şanlıurfa. It was planned to initiate prevention studies by determining the causes of substance abuse such as geographical location, population composition, being a border city and risky schools.

Materials and Methods

This descriptive study was conducted between September 2014 and March 2015.

The Research Sample

The field of the study consists of 84.000 seventh and eighth graders in middle schools and ninth through twelfth graders in high schools under the Directorate of National Education in Şanlıurfa. This study aimed to reach all the seventh through twelfth graders in the city. The research sample consisted of 54.928 students who were at school on the day of data collection and agreed to participate in the study, which intended to include all the seventh through twelfth graders in all the middle and high schools in the city and counties. However, this could not be done as some schools did not give permission to conduct the study, or due to difficulties such as transportation. The participation rate was 65%.

Data Collection Materials

The research data were obtained using a survey form prepared by the researchers after a review of the relevant literature. The opinions of specialists in this area were used to modify the survey form, and a pilot test was done with 15 students after this revision. The survey consisted of 50 questions; 22 were on substance abuse and 28 were about the factors that affect it and demographic information (gender, age, number of siblings, mother and father's age, education status, occupation, family's income status, etc). The adolescents and their teachers were informed about the study. It was explained to the entire sample group that only the volunteered would fill out the forms, and that their answers would remain confidential. They were asked to answer the questions after this step.

The surveys were administered in classrooms by pollsters and took 5-10 minutes to complete. The aim of the study was explained and information about the process was given. Questions about particular conditions participants might encounter during the study were answered beforehand. The children were assessed for the answers they gave to the questionnaire, and no laboratory tests were used to determine the presence of substance usage.

The Ethical Aspects of the Research

Written permissions from the institutions and the verbal consent of the participants were obtained for the study. This study was supported by Karacadağ Development Agency (project number: TRC2/14/DFD/0008). The ethical dimension of the study was embraced within the project evaluation process and the project was accepted.

Statistical Analysis

The data obtained from this research were evaluated using the SPSS 17. Averages, standard deviations, medians, minimum and maximum values, percentiles and chi-square were used to evaluate the data.

Results

Of the students who participated in the study, 46% were female, 51.6% were male, and 80.1% were older than 12 years of age. Of them, 23.8% were in ninth grade; 81.1% had 4 or more siblings, and 40.1% were fourth or more in sibling order. Of the participants, 64.3% said they were involved in social activities (music, sports, trips, cinema, etc.), and 84.9% claimed they loved their school while 23.2% said they worked in jobs to earn money.

The mothers of 47.8% of the students in the study were between 31 and 40 years of age; and 63.9% of the fathers were older than 40. The mothers of 47% were illiterate while the fathers of 32.6% had completed primary education. The mothers of 91.6% were housewives. Of their fathers, 41.4% were self-employed. The monthly income of 39.6% was under 1,000 Turkish Liras. Of the students, 83.4% lived in nuclear families, and 55.4% said family decisions were made jointly. Exposure to domestic violence was claimed by 8.9%.

Table I shows the data on substance abuse by the students.

It was found that 6.3% of the students were using tobacco, and 2.4% were users of substances other than tobacco. Of them, 13.1% had substance abusers among their friends or in their family (Table I). Hashish and glue were used by 36.1%, and 32.3% used bonsai among these. Propane was used by 15.5%, and 10.5% drank alcohol. Finally, 5.5% claimed use of ecstasy. The age for starting smoking was under 10 for 40.7%. Of them, 33.3% said they smoked more than one pack of cigarettes a day, 38.8% smoked due to boredom, and 36.6% started to smoke due to curiosity or to imitate their elders.

Of the substance users, 27% started before age 10, 31.3% said they began substance abuse out of curiosity, and 29.9% said they abused substances to feel better and more powerful. The percentage of students who had abused substances for seven years or more was 24.9%. To obtain substances, 63.7% paid money and 28.3% got them from their friends for free.

Of the students, 31.4% said they abused substances once or twice a month, and 55.6% said they most often used substances at night. While 39.8% of the students wanted to quit substance abuse, 46.6% believed they could quit, and 26.5% were receiving support to quit. The percentage of students receiving support to quit from their friends only was 54.2%.

Statistically significant differences were found between gender, work status, family type and domestic violence, and smoking and substance abuse by the students as seen in Table II ($p<0.01$). Smoking and substance abuse rates were higher for male students, working students, children with extremely domineering families, and children exposed to domestic violence, and these findings are presented in Table II.

Discussion

The substance use rate among adolescents attending 7th through 12th grades in Şanlıurfa was found as 2.4% for substances and 6.3% for smoking (Table I). Lifetime substance use frequency was found to be 1.5% by "Research on the Prevalence of Substance Abuse in the Young Population" carried out by Turkish Monitoring Center for Drugs and Drug Addiction (TUBİM) in 2011 with 11.812 participants studying at general and technical high schools in 32 cities (4).

Table I. Data distribution of substance use by students

Variable		n*	%
Smoking	Yes	2984	6.3
	No	44485	81.0
Use of substances (other than cigarettes)	Yes	858	2.4
	No	34897	63.5
Substance abuser Acquaintances	Yes	4347	13.1
	No	28876	86.9

*Some questions in the questionnaire were not answered by all students

Atilola et al. (3) studied alcohol and substance abuse by 2.454 adolescents in grades nine through twelve from seven developing countries: India, Serbia, Nigeria, Turkey, Indonesia, Croatia and Bulgaria. It was reported that 40.9% of the adolescents had used alcohol or substance at least once in the last year. The study's data for Turkey were collected in the city of Şanlıurfa, and the rate of adolescents abusing any substance was 24.3% during the previous 12 months. In the study of Atilola et al. (3); the sample group only consisted of high school students and the data were obtained via a measurement instrument aimed at determining the rate of using problematic substances. The rate of using problematic substances was determined as 12.9% (3). In our study, this rate was found to be lower, which is associated with the difference in the age group.

Ogel et al. (6) studied smoking, alcohol use and substance abuse with 11.989 primary school and 12.270 secondary school students in nine metropolises in 2004. Of the primary school students, 16.1% said they had smoked at least once, 15.4% said they consumed alcohol, and 1.7% said they had used inhalants and drugs (6).

Tot et al. (7) studied smoking and the use of alcohol, hashish, inhalants and other illicit substances (heroin, cocaine, tranquillisers, etc.) in the last month and the factors that affect these forms of substance abuse with 3.282 sixth graders, tenth graders and college students in Mersin. The

Table II. Variables that affect substance use

Variables	Smoking		Substance use	
	Yes (%)	No (%)	Yes (%)	No (%)
Gender				
Female	645 2.9	21731 97.1	138 0.8	16951 99.2
Male	2260 9.4	21785 90.6	688 3.7	17911 96.3
	X^2=8391 p<0.01		X^2=307.267 p<0.01	
Working status				
Working	637 16.0	3346 5.2	203 6.4	2971 93.6
Not working	1877 84.0	34287 94.8	494 1.8	26958 97.2
	X^2=711.611 p<0.01		X^2=268.076 p<0.01	
Family feature				
Whatever my father says is done	1485 7.6	16528 92.4	438 2.8	12291 97.2
Extremely domineering family	1265 15.5	25597 84.5	335 8.0	19984 92.0
Decisions are made jointly	64 5.0	457 95.0	30 1.8	356 98.2
	X^2=3.962 p<0.01		X^2=2.245 p<0.01	
Domestic violence				
Yes	764 18.2	3425 81.8	296 8.7	3091 91.3
No	2001 4.9	38903 95.1	485 1.6	30152 98.4
	X^2=5.179 p<0.01		X^2=6.969 p<0.01	

highest rates of substance abuse were as 4.7% smoking in the sixth grade, 25.3% smoking in the tenth grade, 43.9% using alcohol in college and 38.7% smoking in college (7,8).

The substance abuse rates found by this study are similar to those of TUBİM in 2011. The substance abuse rates found by Atilola et al. (3) and Ogel et al. (6) were higher.

In the current study, 6.3% of students used cigarettes. Of the students who abused other substances, 36.1% used hashish and glue, and 32.3% abused bonsai. Of this group, 15.5% used propane, 10.5% used alcohol, and 5.5% used ecstasy. The smoking rate was higher than the rate for other substances in this study. The findings of this study are similar to those of Ogel et al. (6) and TUBİM (2011) (4).

In this study, hashish, glue and bonsai had the highest usage rate among substances other than cigarettes. In the study of European School Survey Project on Alcohol and Other Drugs carried out with 6.149 students around 15-16 years of age in Adana, Ankara, Diyarbakır, İstanbul, İzmir and Samsun in 2003, the lifetime rates were 5% inhalants, 4% hashish, 2% ecstasy, 2% heroin and 2% cocaine, and it was found that 5% had used hashish in the last year, and 3% in the last month (4).

Ogel et al. (6) indicated that 15.4% of the primary school students used alcohol and 1.7% used inhalants and drugs other than tobacco. They determined that 55.9% of the secondary school students smoked tobacco, and 45% used alcohol. However, only 4% used cannabis, 5.1% used inhalants, and 2.5% had used heroin or ecstasy at least once.

The TUBİM study (2011) found that 26.7% of the students had tried a tobacco product such as cigarettes, cigars, pipes or hookahs; 19.4% had tried alcoholic drinks, and 2.2% had used drugs due to illness (4).

The highest rates of substance abuse were reported as 37.8% for alcohol, 8.6% for hashish and 8.1% for other substances in a study by Atilola et al. (3). These data for Turkey were collected in the city of Şanlıurfa, and the rate of adolescents abusing any substance was reported as 24.3%, 19.3% for alcohol, 10% for hashish, 15.7% for other substances, and the rate of hazardous or impairing use was 12.9% (3). While alcohol use disorder and alcohol addiction were 7.5% in all European countries in the 2014 data of the World Health Organisation, these rates in Turkey were 2.7% and 0.8% respectively. The use of alcohol rate was lower than that of most of the countries that participated in this study, and the rate of alcohol use was lower than most other substances. This can be attributed to the religious, cultural and geographical features of Şanlıurfa.

Studies by Corapçioglu and Ogel (8) with 18.556 tenth grade students in 1998 and 11.911 tenth grade students in 2001 showed the use of ecstasy rate as 2.65% in 1998 and 3.31% in 2001 (6,7).

According to the 2012 World Drug Report, cannabis is the world's most widely used illicit substance. There are between 119 million and 224 million cannabis users worldwide, and consumption is stable. Hashish was identified as the most used substance in a study by Yüncü et al (9). Hashish was among the most used substances in this study, too. The results of this study are similar to those of Yüncü et al. (9).

Some sociodemographic features that affect smoking and substance abuse by students were also investigated in this study. The smoking and substance abuse rate was higher for male students, working students, children with extremely domineering families, and children exposed to domestic violence (Table II). The findings obtained from the study of Kokkevi et al. (10) on the variables affecting substance abuse by adolescents in six European countries were similar to the studies of TUBİM and Corapçioglu and Ogel (8). Substance abuse was more common among male students than female students.

The substance abuse rate was higher for working students than those who didn't work. This may be due to the fact that children escape parental control when they work, encounter dangers in the streets, engage in risky behaviours, and can easily obtain substances since they earn money.

Parents' educational levels, income levels, co-habitation, and substance abuse have been investigated in studies of families (3,8,10). Regarding extremely domineering families and domestic violence, Kokkevi et al. (10) found that substance abuse was more common among adolescents who were unhappy in their relationship with their parents. When the family environment is unhealthy, the family bonds weaken, parental control of children is reduced, and children run away from home due to violence. The rate of the students who had more than four siblings and were fourth or more in sibling order, was 81.1%. Substance abuse is related to high numbers of children in families in addition to parental variables. As a result of their study including 494 students (5, 7 and 9th grades), Trick et al. (11) determined that, the rate of using substances and other risky behaviours decreased as parents' follow-up increased.

In this study; 54.6% of adolescents stated that their parents had smoking habits, whereas 13.1% stated that they had acquaintances using substances other than cigarettes among their family members and/or friends. In the study of Lobato et al., (12) it was determined that the possibility of using substances was five times greater in adolescents that had histories of substance abuse in their family than those who did not, and eight times greater in individuals who had friends using substances. A number of studies suggest that the parents' substance use is an important risk factor upon the adolescents' substance use (13).

Study Limitations

Limitations of the study are as follows:

- Students at some schools did not participate in the study due to the failure of receiving institutional permission.
- Some questions in the questionnaire were not answered by all students.
- The research data were obtained using a survey form prepared by the researchers after a review of the relevant literature. The opinions of specialists in this area were used to modify the survey form, and a pilot test was done with 15 students after this revision. Because the purpose of the

study was not to develop measuring tools to determine the use of the substances, no other tests were performed except for the expert opinions.

Conclusion

These data obtained on substance use by students and the factors that affect it suggest that studies should be carried out on substance use prevention, family awareness should be increased and prevention policies should be developed and implemented.

Ethics

Ethics Committee Approval: This study was supported by Karacadağ Development Agency (Project number: TRC2/14/DFD/0008). Ethical dimension of the study was embraced within the project evaluation process and the project was accepted, Informed Consent: Consent form was filled out by all participants.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Hülya Karataş, Selma Kahraman, Concept: Hülya Karataş, Selma Kahraman, Design: Hülya Karataş, Selma Kahraman, Data Collection or Processing: Hülya Karataş, Selma Kahraman, Zeynep Marangoz, Analysis or Interpretation: Hülya Karataş, Selma Kahraman, Literature Search: Hülya Karataş, Writing: Hülya Karataş, Selma Kahraman.

Conflict of Interest: No conflict of interest was declared by the authors.

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References

1. World Drug Report 2012. http://www.unodc.org/documents/data-and-analysis/WDR2012/WDR_2012_web_small.pdf. Erişim Tarihi: 17.08.2015.
2. World Health Organization. Global Status Report on Alcohol and Health 2014. http://www.who.int/substance_abuse/publications/global_alcohol_report/msb_gsr_2014_1.pdf?ua=1. Erişim Tarihi: 17.08.2015.
3. Atilola O, Stevanovic D, Balhara YP, et al. Role of personel and family factors in alcohol and substance use among adolescents: an international study with focus on developing countries. J Psychiatr Ment Health Nurs 2014;21;609-17.
4. Turkey Drug Report 2014. http://www.kom.pol.tr Erişim Tarihi 18.08.2015.
5. Turkish Statistical Institute 2015. www.tuik.gov.tr Erişim Tarihi: 06.10.2015.
6. Ogel K, Corapçioğlu A, Sir A, et al. Tobacco, Alcohol and substance use prevalence among elementary and secondary school students in nine cities of Turkey. Turk Psikiyatri Derg 2004;15:112-8.
7. Tot S, Yazici K, Yazici A, Metin O, Bal N, Erdem P. Psychosocial correlates of substance use among adolescents in Mersin, Turkey. Public Health 2004;118:588-93.
8. Corapçioglu A, Ogel K. Factors associated with ecstasy use in Turkish students. Addiction 2004;99:67-76.
9. Yüncü Z, Saatçioğlu H, Aydın C, Özbaran NB, Altıntoprak E, Köse S. Urban Legend: Is the onset age in substance use decreasing? Literatür Sempozyum 2014;1:43-50.
10. Kokkevi A, Richardson C, Florescu S, Kuzman M, Stergar E. Psychosocial correlates of substance use in adolescence: a cross-national study in six European countries. Drug Alcohol Depend 2007;86:67-74.
11. Trick S, Jantzer V, Haffner J, Parzer P, Resch F. Parental monitoring and its relation to behaviour problems and risk behaviour in an adolescent school sample. Prax Kinderpsychol Kinderpsychiatr 2016;65:592-608.
12. Lobato M, Sanderman R, Pizarro E, Hagedoorn M. Marijuana use and dependence in Chilean adolescents and its association with family and peer marijuana use. Int J Behav Med 2016;24:144-52.
13. Durand SC, Fogger SA, McGuinness TM. Update on substance use in adolescence. J Psychosoc Nurs Ment Health Serv 2016;54:24-7.

Survey Form for 7th-12th Graders

1. Gender: Man ( ) Woman ( )

2. Age:……

3. Grade: ……

4. Number of siblings? ……

5. What is your bird order among your siblings? ……

6. Mother's age: ……

7. Father's age: ……

8. Mother's education status:

a) Illiterate **b)** Literate **c)** Primary school graduate **d)** Middle school **e)** High school and above

9. Father's education status:

a) Illiterate **b)** Literate **c)** Primary school graduate **d)** Middle school **e)** High school and above

10. Mother's occupation:

a) Housewife-does not work **b)** Officer **c)** Worker **d)** Freelancer **e)** Other…….....

11. Father's occupation:

a) Does not work **b)** Officer **c)** Worker **d)** Freelancer **e)** Other…….....

12. Family's income status:
a) Below 1000 TL **b)** 1000 TL **c)** Above 1000 TL

13. How is your family's monetary status?
a) Very good **b)** Good **c)** Medium **d)** Bad **e)** Very bad

14. With whom do you live together?
a) Parents-children **b)** Mother or father, children **c)** Father's father, grandfather, parents, children

d) Mother's mother, grandfather, parents, children **e)** Only mother's or father's mother, parents children **f)** Other

15. Home status:
( ) Rent ( ) Our own house( ) Other…….

16. Which of the below defines your family?
a) Whatever my father says **b)** Whatever my mother saysc) Common decisions are taken in the family

d) Whatever I say **e)** I have a very oppressive family **f)** Other…….....

17. Do you like coming to school?
a) Yes **b)** No

18. Are there days you skipped school?
a) Yes **b)** No If yes, why? ……

19. Does your family give you allowance?
a) Yes **b)** No

20. How much allowance does your family give you weekly?
……

21. Was there any violence against you from your family?
a) Yes **b)** No

22. Who used violence?
……

23. If yes, how?
a) Beating/injuring **b)** Swearing/insult **c)** Sexual harassment **d)** Other…….

24. If yes, how often?
a) Once a week **b)** Once a month **c)** Less than once a month **d)** More than once a week

25. Were there times you could not go home and had to spend the night outside?
a) Yes **b)** No

26. If your answer is yes, can you explain the reason?
a) I work at night **b)** I travel and have fun at night **c)** I do not want to go home **d)** I stayed with my friends **e)** Other……

27. Do you have any social activity?
a) Yes **b)** No

28. What kind of activities do you do?
a) Sports **b)** Music **c)** Reading **d)** Travelling **e)** Movies **f)** Theater

29. What does "drug" bring into your mind? ……

30. Is there anyone smoking in your family?
a) Yes **b)** No

31. Do you smoke?
a) Yes **b)** No (If your answer is no, move to question no. 53)

32. When did you start smoking? .....
33. Why did you start smoking?
a) Peer pressure **b)** Saw it in my family **c)** Curiosity **d)** Aspiration **e)** Boredom **f)** Other

34. If yes, how often do you smoke?
a) Less than 1 package a day **b)** More than 1 package a day **c)** Other……

35. Have you used any drugs other than cigarettes?
a) Yes, I used …… **b)** I did not (if your answer is no, move to question no. 51)

36. If yes; *Why did you start using that drug?
a) I was curious **b)** To feel better **c)** Because everybody was using it **d)** My family/brother/etc. forced me

e) To do better **f)** Because I was stressed **g)** Because I had problems **h)** Because my friends asked me to

ı) To look stronger **i)** Other (please specify) ……

37. How do you acquire the drug you use?
a) My friends provide me for free **b)** I purchase it **c)** My relatives provide me for free **d)** Other……

38. Where do you think these drugs are sold the most?……
39. Where do you think these drugs are used the most?……
40. When did you start using drugs?……
41. How long have you been using it?
a) Less than 1 year **b)** 1-3 years **c)** 4-6 years **d)** 7 years and above

42. How often do you use it?
a) More than once a day **b)** Once a day **c)** 1-2 times a week **d)** 1-2 times a month **e)** Other

43. How much do you use that drug?……
44. Which hours do you use that drug?……
45. Does your family know that you use drugs?
a) Yes **b)** No

46. Do you want to quit using that drug?
a) Yes **b)** No

47. If yes, did you ask help from anyone to quit it?
a) Yes **b)** No

48. If yes, ( ) from whom? …… No ( )
49. Do you think you can quit using drugs?
a) Yes **b)** No

50. Is there anyone from your friends or family using drugs?
a) Yes **b)** No

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Lymphadenitis and Fever: First Presentation of Kawasaki Disease

Lenfadenit ve Ateş: Kawasaki Hastalığının İlk Bulgusu

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ABSTRACT

Kawasaki disease is a vasculitis of infant and childhood period. Diagnosis of Kawasaki disease is based on clinical criteria including cervical lymphadenopathy. Here we reported a 3.5-year-old boy presenting with Kawasaki disease mimicking bacterial lymphadenitis and fever. If a child presents with lymphadenopathy only which is unresponsive to antibiotics, Kawasaki disease should be considered.

Keywords: Kawasaki disease, bacterial lymphadenitis, child

ÖZ

Kawasaki hastalığı çocukluk ve infant dönemin bir vaskülitidir. Servikal lenfadenopatinin de içinde yer aldığı bazı klinik kriterler ile tanı konulur. Bu yazıda kliniğimize ateş ve bakteriyel lenfadenit kliniği ile başvuran 3,5 yaşında bir erkek olgu sunulmuştur. Antibiyotiğe cevapsız bakteriyel lenfadenit kliniği ile başvuran olgularda Kawasaki hastalığı göz önünde bulundurulmalıdır.

Anahtar Kelimeler: Kawasaki hastalığı, bakteriyel lenfadenit, çocuk

Introduction

Kawasaki disease (KD), also known as mucocutaneous lymph node syndrome and infantile polyarteritis nodosa was first described in 1967 by Tomisaku Kawasaki (1,2). The disease most frequently occurs in children aged between 6 months and 5 years. KD is the leading cause of pediatric acquired heart disease in developing countries. There is no specific diagnostic test for KD. The diagnosis is made based on the presence of clinical criteria. Typical KD criteria include fever going on for more than 5 days, cervical adenopathy, nonpurulent bilateral conjunctival injection, oral mucosal changes, hand-foot changes in extremities and polymorfic rash on body. Four of 5 clinical criteria define diagnosis (3). Of the principal diagnostic criteria, cervical adenopathy is the least common. However, KD manifests with fever and cervical adenopathy and may be misdiagnosed as bacterial cervical lymphadenitis. Therefore, specific treatment is delayed, leading to serious cardiac sequelae.

Herein, we report an unusual case of KD: a 3.5-year-old boy who presented with fever and bilateral cervical lymphadenitis as initial manifestations.

Case Report

A 3.5-year-old boy presented with a high fever, swelling in the neck, fatigue, restlessness and dehydration. He had a history of persistent fever for ten days, reaching 40.5 ºC, especially at nights. Before admission, he received an oral treatment of amoxicillin-clavulanate 90 mg/kg/24 hr for 4 days ceftriaxone 80 mg/kg/24 hr, and two doses of metronidazole 30 mg/kg/24 hr parenterally. Physical examination revealed

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three lymph node enlargements unilaterally in cervical region, the largest being, 30 mm x 30 mm in diameter; fixed, solitary, tender without fluctuation. Non-specific complaints were present including abdominal cramping, weight loss due to decreased oral intake and change in defecation. Blood testing showed white blood cell (WBC): 14.5000/mm^3 with neutrophil predominance (55%); hemoglobin: 10.4 g/dL; platelet counts: 291.000/mm^3, C-reactive protein (CRP): 7.5 mg/dL, and sedimentation ratio: 66 mm/h. Biochemical markers were normal except for slightly reduced albumin (2.9 g/dL). Soft-tissue neck ultrasonography revealed multiple lymph nodes, of fusiform shaped-reactive nature, of which the biggest was located at the right juguler triangle, 30x12 mm in diameter. Neck computerized tomography was obtained to exclude deep cervical infection. Multiple and diffuse lymphadenitis were seen at the level of superficial and deep cervical lymph nodes. The biggest one was on the right side, at jugulodigastric localization, 32x16x20 mm in diameter (Figure 1). Treatment with intravenous clindamycin 30 mg/kg/24 hr was initiated to treat possible infectious components, hydration was administered via inravenous route, but his condition did not improve, and the neck masses remained painful. The patient exhibited with enlarged cervical nodes three days after admission. The throat swab culture and blood culture samples taken at the time of admission were negative for infection. Incomplete Kawasaki syndrome was suspected due to the child's irritated, tired appearance; continuing fever, and bilateral cervical lymphadenitis an echocardiography was performed showing increased thickness of coronary artery walls (Figure 2). The patient was diagnosed with KD based on the echocardiographic findings. Immediate treatment with intravenous immnunoglobulin (2 g/kg for one day) and 80 mg/kg/day asetilesalysalic acid was initiated. Significant improvement was observed on the first day after the treatment. The lymph node sizes and acute phase response were reduced on the following day. Besides, the patient's fever subsided and the other symptoms resolved. Control laboratory analyses revealed: WBC counts 9650/mm^3, platelet counts 691.000/mm^3, sedimentation ratio 30 mm/h, and CRP 0.3 mg/dL. Follow-up echocardiography examinations after one week were normal. On the 9th day of the treatment, desquamation on the patient's fingers and toes, and macular rash on his feet were observed. Two weeks later, asetilsalysalic acid treatment dose was decreased to antiaggregant level, and the patient was discharged. Follow-up echocardiography examinations during the 2-month period after discharge were normal.

Discussion

KD is a systemic vasculitis affecting small and medium-sized vessels and is most often seen in children between 6 months and 5 years (4). It's etiology is still unknown but clinical, laboratory and epidemiological features suggest an infectious origin (5). Typical Kawasaki symptoms include fever- of more than 5 days-being unresponsive to antibiotic treatment, cervical lymphadenitis, nonpurulent bilateral conjuctival injection, oral mucosal changes, hand-foot changes in the body. In KD, lymph node enlargements are less frequent than the other findings (25-50%) (6), and their incidence is more frequent in patients of older ages. Lymphadenopathy and fever have previously been reported as initial presentations of KD in the literature. Additionaly KD has sometimes mimicked bacterial lymphadenitis by including prominent swelling, pain, and redness with high body temperatures. This may cause a delay in the diagnosis of KD and increase the risk of coronary artery involvement. Nomura et al. (7) evaluated laboratory and clinical findings of 187 patients with confirmed KD and 16 patients presented with only fever and cervical lymphadenopathy on admission. Three of sixteen patients presented with bacterial cervical lymphadenitis or deep neck infection (7). Kubota et al. (8) reported that patients with KD presenting with only fever and cervical lymphadenopathy were older and admitted earlier. Another case report by April et al. (9) demonstrated that patients with lymphadenopathy were older and had stronger inflammatory responses than those without lymphadenopathy. Yap et al. (10) reported a

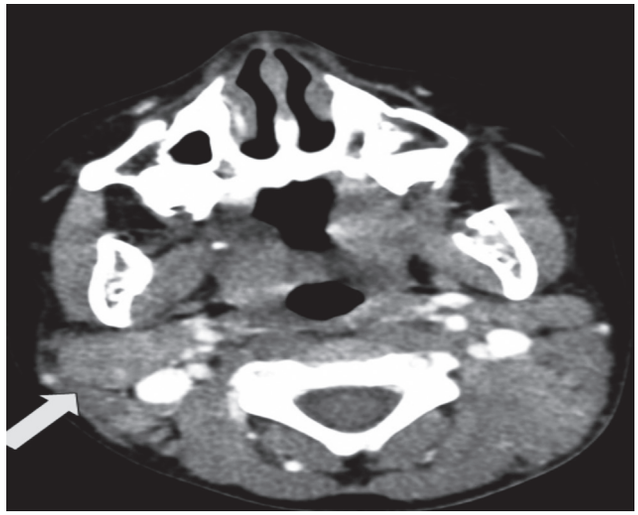

Figure 1. Computerized tomography scan of neck, both deep and superficial lymph node on the right, the largest chain of 32x16x20 mm, including common and multiple lymphadenomegalies

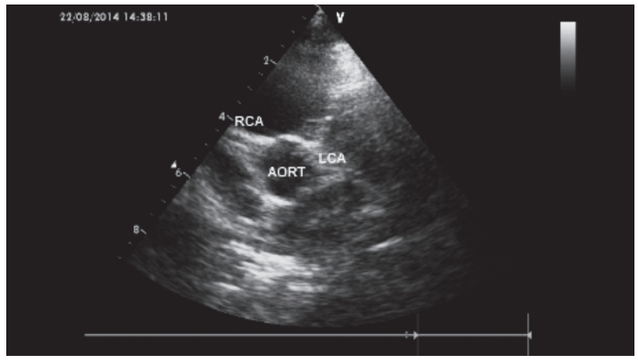

Figure 2. Echocardiography showing increased thickness of coronary artery walls especially right coronary artery
RCA: Right coronary artery, LCA: Left coronary artery

10 year-old boy, admitted with acute exudative tonsillitis and bilateral cervical lymphadenitis. The patient had a 4-day history of fever, sore throat, neck pain and an unresponsive fever despite 72 hours of intravenous antibiotic treatment. Fourteen days post admission the patient presented sheet-like desquamation of the hands, feet, and perianal region. KD was diagnosed mimicking bacterial lymphadenitis and fever. They suggested that children presenting with sole fever and cervical lymphadenopathy could be considered as having KD.

In our case we tried to emphasize that KD can present as bacterial lymphadenitis. In case of acute bacterial lymphadenitis unresponsive to antibiotic regimen, KD should be considered in differential diagnosis.

Ethics

Informed Consent: Consent form was filled out by all participants.

Peer-review: Internally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Şule Gökçe, Pelin Elibol, Güldane Koturoğlu, Concept: Güldane Koturoğlu, Zülal Ülger Tutar, Design: Şule Gökçe, Yasemin Özdemir Şahan, Data Collection or Processing: Şule Gökçe, Pelin Elibol, Analysis or Interpretation: Şule Gökçe, Literature Search: Şule Gökçe, Pelin Elibol, Güldane Koturoğlu, Yasemin Özdemir Şahan, Zülal Ülger Tutar, Writing: Şule Gökçe, Pelin Elibol, Güldane Koturoğlu.

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References

1. Son MB, Newburger JW. Kawasaki disease. In: Kliegman RM, Stanton BF, Geme JWSt III, Schor NF, Behrman RE, eds. Nelson Textbook of Pediatrics, 19th edition. Philadelphia, Pa, USA: Elsevier; 2011. pp. 862-5.
2. Takahashi K, Oharaseki T, Yokouchi Y. Pathogenesis of Kawasaki Disease. Clin Exp Immunol 2011;164:20-2.
3. Burns JC, Mason WH, Glode MP, et al. Clinical and epidemiologic characteristics of patients referred for evaluation of possible Kawasaki disease. United States Multicenter Kawasaki Disease Study Group. J Pediatr 1991;118:680-6.
4. Anderson BL, Guiot AB, Timm NL. An atypical presentation of atypical Kawasaki disease. Pediatr Emerg Care 2014;30:491-2.
5. Suddleson EA, Reid B, Woolley MM, Takahashi M. Hydrops of the gallbladder associated with Kawasaki syndrome. J Pediatr Surg 1987;22:956-9.
6. Tashiro N, Matsubara T, Uchida M, Katayama K, Ichiyama T, Furukawa S. Ultrasonographic evaluation of cervical lymph nodes in Kawasaki disease. Pediatrics 2002;109:E77-7.
7. Nomura Y, Arata M, Koriyama C, et al. A Severe Form of Kawasaki Disease Presenting with Only Fever and Cervical Lymphadenopathy at Admission. J Pediatr 2010;156:786-91.
8. Kubota M, Usami I, Yamakawa M, Tomita Y, Haruta T. Kawasaki disease with lymphadenopathy and fever as sole initial manifestations. J Paediatr Child Health 2008;44:359-62.
9. April MM, Burns JC, Newburger JW, Healy GB. Kawasaki disease and cervical adenopathy. Arch Otolaryngol Head Neck Surg 1989;115:512-4.
10. Yap CY, Lin LH, Wang NK. An atypical presentation of Kawasaki disease: a 10-year-old boy with acute exudative tonsillitis and bilateral cervical lymphadenitis. Clinics (Sao Paulo) 2012;67:689-92.

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Antipsychotic Drugs Rechallenge in Multi-antipsychotic Drug Induced Atypical Neuroleptic Malignant Syndrome: A Case of Cotard's Syndrome

Çoklu Antipsikotik İlaç Kullanımı ile Tetiklenen Atipik Nöroleptik Malign Sendromda Antipsikotik İlaç Başlama Güçlükleri: Cotard Sendromu Olgusu

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ABSTRACT

Neuroleptic malignant syndrome (NMS) is an uncommon but potentially fatal idiosyncratic reaction to neuroleptics and characterized by a distinctive clinical syndrome of mental status change, rigidity, fever, and dysautonomia. Cotard's syndrome is characterized by the appearance of nihilistic delusions concerning one's own body or life. By presenting this case, we aim to discuss the differential diagnosis and treatment plan of a patient with catatonia and Cotard's syndrome, which were noted after NMS, in light of the literature.

Keywords: Neuroleptic malignant syndrome, Cotard's syndrome, atypical neuroleptic malignant syndrome, nihilistic delusions, electro-convulsive therapy

ÖZ

Nöroleptik malign sendrom (NMS), nöroleptik ajanların kullanımıyla ortaya çıkan, bilinçte değişiklikler, rijidite, ateş yüksekliği ve otonomik değişiklikler ile karakterize olan, nadir fakat ölümcül bir idiosenkratik reaksiyondur. Cotard sendromu ise, kişinin bedeni ya da yaşamı ile ilişkili nihilistik sanrılarla karakterize bir sendromdur. Bu olgu sunumu ile, NMS sonrasında katatoni ve Cotard sendromu gelişen hastada, ayırıcı tanıları ve tedavi planını literatür eşliğinde tartışmayı amaçladık.

Anahtar Kelimeler: Nöroleptik malign sendrom, Cotard sendromu, atipik nöroleptik malign sendrom, nihilistik sanrı, elektrokonvülsif terapi

Introduction

Neuroleptic malignant syndrome (NMS) is an uncommon but potentially fatal idiosyncratic reaction to neuroleptics, characterized by a distinctive clinical syndrome of mental status change, rigidity, fever, and dysautonomia (1).

Cotard's syndrome (CS) is characterized by the appearance of nihilistic delusions concerning one's own body.

"We report a case of a 17-year-old boy with catatonia and nihilistic-paranoid delusions, which were noted after multi-antipsychotic drug treatment induced atypical NMS."

Case Report

A 17-year-old boy was admitted to the psychiatry clinic because of incoherent speech which emerged after somatic pains, irritability, temper outbursts, and initial insomnia. The patient was prescribed olanzapine (10 mg per day) in Ege University Faculty of Medicine Adult Psychiatry inpatient service. After admission to the clinic, risperidone 2 mg per day, biperiden 2 mg per day, olanzapine 5 mg per day were added to his treatment. Due to hyper salivation,

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bradykinesia, cogwheel phenomenon, and dysphagia, the dose of biperiden was increased to 6 mg per day, risperidone treatment was stopped, clozapine 12.5 mg per day was initiated. One week later, he was admitted to the intensive care unit because of hyperthermia, confusion, irregular pulse and blood pressure, vomiting, hemoptysis, dyspnea, wheezing and laboratory abnormalities such as elevation of creatine kinase (CK) (five times the upper limit of normal), lactate dehydrogenase, liver transaminases, troponin T, and leukocytosis, but urine myoglobin was negative. Posterior-anterior chest radiography and echocardiography were consistent with pulmonary edema and ventricular failure, respectively. A presumptive diagnosis of NMS was made. Anti-psychotic treatment was discontinued and supportive care was initiated afterwards (biphasic positive airway pressure, dopamine, milrinone, furosemide, intravenous hydration and alkalinization), amantadine and bromocriptine were started. His general condition improved and pulmonary edema and cardiac failure rapidly decreased within the fifth day. He was transferred to child and adolescent psychiatry inpatient service with a treatment of digoxin 0.125 mg per day and carnitine 1 g per day.

On admission, he was agitated, depressive and had nihilistic and bizarre delusions (he was convinced that he was dead and at the same time immortal; and that his body and his teeth had melted) and was suicidal. He exhibited parkinsonism signs, including tremor, bradykinesia and unsteady gait, he also had catatonic features including catalepsy, negativism, posturing, mutism. The patient was diagnosed with CS with underlying major depressive disorder with psychotic features and catatonia. During the first three days, he received a total of 2.5 mg lorazepam. On the fourth day, escitalopram 2.5 mg per day and lorazepam 2.5 mg per day were added to his treatment schedule. The following day, due to cramping pain in his neck and inappropriate laughter, increase in his motor activity, and statements like "his entire body was melting except for his penis"; acute dystonia and behavioral disinhibition were suspected and escitalopram treatment was stopped, and the dose of lorazepam was increased to 7.5 mg per day. He was rechallenged with quetiapine two weeks after the resolution of NMS without the recurrence of the symptoms. But quetiapine was not sufficient to control the symptoms, and increasing its dose rapidly could have caused the recurrence of NMS. Hence, we transferred the patient to another clinic for the purpose of electro-convulsive therapy (ECT) implementation.

Discussion

In this case report, the differential diagnosis and treatment plan of a patient with catatonia and CS, which were noted after NMS, were discussed.

NMS is a rare but potentially fatal complication of anti-psychotic pharmacotherapy (1). Incidence and mortality of NMS has declined in recent years owing to the awareness on the disorder. Estimated incidence is about 0.01-0.02% for patients treated with anti-psychotics, but it has not yet been clarified for children and adolescents (2,3).

The Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5) identified the core features of NMS as hyperthermia, rigidity and CK elevation after exposure to a dopamine antagonist within 72 hours. Neurological signs (e.g., tremor, sialorrhea, akinesia, dystonia, trismus, myoclonus, dysarthria, dysphagia, rhabdomyolysis), changes in mental status, autonomic instability (e.g., tachycardia, blood pressure elevation or fluctuation, urinary incontinence, pallor, and tachypnea) may accompany these features. It is stated in DSM-5 that NMS is often heterogeneous in onset, presentation, progression, and outcome. In our case report, hyperthermia and elevated CK levels were detected although muscle rigidity was absent, consistent with negative urine myoglobin. Pulmonary edema and ventricular failure considered as cardiorespiratory failure, are complications of NMS.

Non-specific laboratory abnormalities such as leukocytosis, elevation of transaminases, lactate dehydrogenase, and alkaline phosphatase can be observed in NMS besides CK level elevation, and we determined all of these laboratory abnormalities in our case (1,2).

The symptoms of NMS initiates between 24 hours to 30 days after anti-psychotic drug intake and resolves within 7-10 days subsequent to discontinuation of the inducing agent (4-6). In our case report NMS could have been triggered by olanzapine, risperidone or clozapine because all of these neuroleptics were initiated within 14 days before the diagnosis of NMS. Besides, NMS resolved on the fifth day after the discontinuation of anti-psychotic drugs.

Some risk factors have been reported for NMS, such as increased and rapidly titrated anti-psychotic doses, agitation, dehydration, physical restrain, iron deficiency, history of previous NMS, parenteral administration routes, excessive alcohol consumption (1,2,6-9). We detected iron deficiency, agitation, and dehydration as risk factors in our case.

NMS is a diagnosis of exclusion, so it should be differentiated from other conditions, including infection of central nervous system (CNS), agitated delirium, malignant catatonia, serotonin syndrome, malignant hyperthermia (2,10). There was neither prodromal viral illness nor other neurological signs, so we excluded the infection of the CNS. Due to the absence of history of using serotonergic agents or getting general anesthesia; serotonin syndrome and malignant hyperthermia were excluded, respectively. Although malignant catatonia is a differential diagnosis of NMS, it might be hard to distinguish the two conditions because of the associated clinical (e.g. rigidity, hyperthermia, autonomic instability, stupor) and laboratory findings (e.g., elevated CK levels, reduced serum iron level, electroencephalography abnormalities). Moreover, residual catatonia can persist for weeks after the resolution of NMS (5,11). In our case, catatonia was not detected before the patient was admitted to the intensive care unit, so we excluded malignant catatonia, and diagnosed him with atypical NMS induced with multiple neuroleptics.

Identification of the syndrome is the keystone for NMS treatment. The first step of the treatment is discontinuation of the anti-psychotic agent. After that, supportive therapy must be initiated (e.g., aggressive rehydration and restoring electrolyte balance, alkaline fluids, physical cooling, careful monitoring of complications, including aspiration pneumonia, acute renal failure, cardiac arrest, pulmonary embolism, disseminated intravascular coagulation) and specific agents such as amantadine, bromocriptine and dantrolene must be used in order to reduce the mortality rate (6,12,13).

Due to the high possibility of NMS recurrence, at least 2 weeks should elapse after recovery; low doses of low-potency anti-psychotics should be titrated gradually after a test dose; and patients should be carefully monitored for early signs of NMS when psychotic symptoms persist (6). We initiated low dosage quetiapine treatment after recovery from NMS and titrated it very slowly. If residual symptoms and catatonia persist after NMS, or supportive treatment and specific agents do not control NMS, ECT might be considered as a treatment option (12).

CS was first identified by James Cotard in 1880 as a new form of depression composed of anxious melancholia, ideas of damnation or rejection, insensitivity to pain, delusions of nonexistence concerning one's own body, and delusions of immortality. There is insufficient data about the prevalence and incidence of the syndrome (14). CS is described as a cluster of symptoms as part of an underlying disorder, mostly depressive and bipolar (15-18). Although our initial diagnosis was major depressive disorder with psychotic features, patients must be followed closely for bipolar disorder because of manic symptoms after treatment with escitalopram, and an increased risk of bipolar disorder during adolescence with CS (17). The most frequent symptoms of CS are depressive mood (89%), nihilistic delusions concerning one's own body (86%), nihilistic delusions concerning one's own existence (69%), anxiety (65%), delusions of guilt (63%), delusions of immortality (55%), and hypochondriac delusions (58%) (19). Our patient was convinced that he was dead, that his body and his teeth had melted and he felt very anxious because he was guilty, and this delusion caused extreme suffering and suicidal thoughts. However, at the same time he was convinced that he was immortal. Therefore, all symptoms were consistent with CS. Yamada et al. (20) divided the course of CS into three stages in 1999. The first stage, "the germination stage", is characterized by significant hypochondriasis and a depressive mood. The second stage, "the blooming stage", which is more specific to Cotard symptom, includes nihilistic and immortality delusions. Ultimately, the last stage, "chronic stage", is divided into two forms: a depressive and a paranoid type (20). The first two stages were present in our case. Prognosis and treatment is based on the underlying disorder, and monotherapy, combination therapy, or ECT can be used as a treatment option (14). Thus, we both prescribed anti-psychotics and planned to imply ECT in the course of our treatment.

In summary, anti-psychotic treatment is a challenge after NMS, and ECT might be considered as a treatment option in these cases.

Ethics

Informed Consent: Consent form was filled out by all participants.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Helin Yılmaz, N. Burcu Özbaran, Sezen Köse, Concept: Helin Yılmaz, N. Burcu Özbaran, Sezen Köse, Design: Helin Yılmaz, N. Burcu Özbaran, Sezen Köse, Data Collection or Processing: Helin Yılmaz, N. Burcu Özbaran, Sezen Köse, Literature Search: Helin Yılmaz, Writing: Helin Yılmaz.

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References

1. Adnet P, Lestavel P, Krivosic-Horber R. Neuroleptic malignant syndrome. Br J Anaesth 2000;85:129-35.
2. Strawn JR, Keck PE, Caroff SN. Neuroleptic malignant syndrome. Am J Psychiatry 2007;164:870-6.
3. Silva RR, Munoz DM, Alpert M, Perlmutter IR, Diaz J. Neuroleptic malignant syndrome in children and adolescents. J Am Acad Child Adolesc Psychiatry 1999;38:187-94.
4. Vihang N. Vahia. Diagnostic and statistical manual of mental disorders 5: A quick glance. Indian J Psychiatry 2013;55:220-3.
5. Bond AG. Antipsychotic Rechallenge After Neuroleptic Malignant Syndrome with Catatonic Features. 2011:1-28.
6. Picard LS, Lindsay S, Strawn JR, Kaneria RM, Patel NC, Keck PE. Atypical neuroleptic malignant syndrome: diagnostic controversies and considerations. Pharmacotherapy 2008;28:530-5.
7. Copeland WE, Angold A, Costello EJ, Egger H. Prevalence, comorbidity, and correlates of DSM-5 proposed disruptive mood dysregulation disorder. Am J Psychiatry 2013;170:173-9.
8. Keck PE. Risk Factors for Neuroleptic Malignant Syndrome. Arch Gen Psychiatry 1989;46:914.
9. Nisijima K, Ishiguro T. Neuroleptic malignant syndrome: A study of CSF monoamine metabolism. Biol Psychiatry 1990;27:280-8.
10. American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders (5th.; 2013. doi:10.1176/appi.books.
11. Koch M, Chandragiri S, Rizvi S, Petrides G, Francis A. Catatonic signs in neuroleptic malignant syndrome. Compr Psychiatry 2000;41:73-5.
12. Trollor J, Sachdev P. Electroconvulsive treatment of neuroleptic malignant syndrome: a review and report of cases. Aust N Z J Psychiatry 1999;33:650-9.
13. Reulbach U, Dütsch C, Biermann T, et al. Managing an effective treatment for neuroleptic malignant syndrome. Crit Care 2007;11:R4.

14. Debruyne H, Portzky M, Peremans K, Audenaert K. Cotard's Syndrome. Mind&Brain 2011;2:67-72.
15. Chiu H. Cotard's syndrome in psychogeriatric patients in Hong Kong. Gen Hosp Psychiatry 1995;17:54-5.
16. Debruyne H, Portzky M, Van den Eynde F, Audenaert K. Cotard's syndrome: A review. Curr Psychiatry Rep 2009;11:197-202.
17. Consoli A, Soultanian C, Tanguy ML, et al. Cotard's syndrome in adolescents and young adults is associated with an increased risk of bipolar disorder. Bipolar Disord 2007;9:665-8.
18. Cohen D, Cottias C, Basquin M. Cotard's syndrome in a 15-year-old girl. Acta Psychiatr Scand 1997;95:164-5.
19. Berrios GE, Luque R. Cotard's syndrome: analysis of 100 cases. Acta Psychiatr Scand 1995;91:185-8.
20. Yamada K, Katsuragi S, Fujii I. A case study of Cotard's syndrome: stages and diagnosis. Acta Psychiatr Scand 1999;100:396-8.

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Catheter Related *Leuconostoc Mesenteroides* Bacteremia: A Rare Case and Review of the Literature

Kateter İlişkili *Leuconostoc Mesenteroides* Bakteriyemisi: Nadir Bir Olgu ve Literatürün Derlenmesi

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ABSTRACT

Herein we report the successful treatment of catheter related blood stream infections due to *Leuconostoc mesenteroides* with antibiotic lock therapy in addition to systemic treatment. With our case, we have shown that in the presence of catheter related blood stream infections, antibiotic lock therapy can be used as a therapeutic option to get successful results if the catheter cannot be removed or there are still positive cultures despite the systemic antibiotic therapy.

Keywords: *Leuconostoc mesenteroides*, catheter related bacteremia, antibiotic lock therapy

ÖZ

Leuconostoc mesenteroides'e bağlı kateter ilişkili kan akımı enfeksiyonunun sistemik tedaviye ilaveten antibiyotik kilit tedavisi ile başarılı bir şekilde tedavi süreci anlatıldı. Sunulan olguyla kateter ilişkili kan akımı enfeksiyonu varlığında eğer kateter çıkartılamıyor ve sistemik antibiyotik tedavisine rağmen üreme devam ediyorsa antibiyotik kilit tedavisinin uygulanabileceği ve başarılı sonuç alınabileceği gösterildi.

Anahtar Kelimeler: *Leuconostoc mesenteroides*, kateter ilişkili enfeksiyon, antibiyotik kilit tedavisi

Introduction

Advances in the field of microbiology and awareness among the microbiologists led to an increase in the rate of identification of rare opportunistic microorganisms in humans. *Leuconostoc* species are catalase negative, facultative anaerobic gram positive cocci configured in doubles or chains. They are usually found in plants, dairy products, wine and food. They are initially believed to be a member of the flora of vagina and gastrointestinal system (1,2). In recent years, many case reports of serious infections led to an increased awareness and focus on risk factors, pathogeneticity and treatment options.

This case report documents the successful treatment of catheter associated blood stream infection due to *Leuconostoc* bacteria with antibiotic lock treatment in addition to systemic antibiotic treatment. This is a novel approach in cases where catheter removal is not an option because of

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the lack of alternative intravenous line. To our knowledge, it is the first known report of *Leuconostoc mesenteroides* bacteremia successfully treated with antibiotic lock therapy.

Case Report

A fifty-day-old girl was admitted to pediatric gastroenterology department with chronic diarrhea. She had complaints of diarrhea and vomiting for two weeks. Following diagnostic endoscopy, she suffered from a duodenum perforation. A central catheter was placed at the surgery. After the operation, piperacillin-tazobactam, teicoplanin and fluconazole were given as an empirical therapy. Total parenteral nutrition (TPN) was initiated. Her fever began five days later from the cessation of empirical antibiotic therapy. Laboratory workup revealed white blood cell count 13.700/mm^3, hemoglobin 9.8 g/dL, platelet count 328.000/mm^3, C-reactive protein: 22 mg/dL. Blood cultures were taken, empirical fluconazole and cefoperazone-sulbactam were initiated. As fever continued, teicoplanin was added on the second day. Amikacin was added on the third day because her appearance became worsened. Although initial blood cultures remained sterile, blood cultures taken on the third day of the therapy were positive for gram positive cocci. She had an intractable fever exceeding 39 °C. *Leuconostoc mesenteroides* were identified in addition to coagulase negative *Staphylococcus* on the seventh day of therapy. The signal for growth of *Leuconostoc* was detected at 5th and 13th hours from the catheter culture and peripheral vein culture, respectively. Teicoplanin was discontinued on the 6th day and high dose linezolid and ampicilline were initiated. Catheter lock therapy was planned but the central line was accidentally removed. As no other vascular access could be obtained, another central catheter was placed despite ongoing bacteremia. Echocardiography was negative for valvular vegetations. Abdominal ultrasonography revealed ascites. Cultures of peripheral blood and catheter remained positive for *Leuconostoc*. The signal for growth of *Leuconostoc* was detected from the catheter culture and the peripheral vein culture at 13th and 28th hours, respectively. Antibiotic lock therapy was re-initiated on the 12th day of fever. On the third day of antibiotic lock therapy, her fever resolved, and cultures remained sterile. Rectal cultures were positive for *Escherichia coli*. After systemic antibiotherapy for three weeks, antibiotic lock therapy for 2 weeks and full enteral feeding, she was discharged from the hospital.

Discussion

Leuconostoc spp. is a member of *Streptococcaceae* family. It is not an easily recognized microorganism by routine biochemistry and phenotypical identification. As it is a non-hemolytic or alfa-hemolytic gram positive coccus on sheep agar, it may be mistaken for *Enterococcus* or *Streptococcus* (3). Like *Enterococci* it may reproduce in 6.5% NaCl and hydrolysis esculin in the presence of bile. *Leuconostoc* species are incapable of producing leucine aminopeptidase and pyrrolidonyl arylamidase. CO_2 formation with glucose are distinctive properties while not a part of routine investigations in many microbiology labs. Antibiotic sensitivity tests are important for identification. *Leuconostoc* species are naturally glycopeptide resistant (1). In our case, blood cultures revealed non-hemolytic gram positive cocci, forming short chains mostly in doubles which were initially thought to be *Streptocci*. Antibiotic sensitivity test showed sensitivity to ampicillin and penicillin but resistance to vancomycin, it was suspected from *Leuconostoc* species. A verification was performed using BD phoenix 100 version 6.01A. Result was 99% consistent with *Leuconostoc mesenteroides spp. mesenteroides*.

Leuconostoc species were not considered as pathogens for humans until early eighties. The first report as human pathogen was by Buu-Hoi et al. (4) in 1985. Two cases with blood cultures which had gram positive cocci resistant to vancomycin were reported. They were initially identified as *Streptococci* by API 20 Strep system but gas production from glucose and fermentation status of various carbohydrates led to the identification of *Leuconostoc* species (4). *Leuconostoc* has been accepted as the etiologic agent for bacteremia, sepsis, catheter related blood stream infection, meningitis, endocarditis, brain and liver abscess, osteomyelitis, pulmonary and nosocomial infections (1,3,5-9). Sixty percent of *Leuconostoc* case reports between 1985 and 1996 were of children (10). Most of the patients presenting with *Leuconostoc* were immunocompromised patients (malignancy, liver transplantation, chemotherapy, immunosuppression) (10-13). However, some patients with *Leuconostoc* infection were immunocompetent (14). Other risk factors among published reports were gastrointestinal disease, prior vancomycin therapy, surgery distrupting gastrointestinal mucosal integrity, TPN usage, central venous line, prematurity (1,3,10,13,15).

Immunologic workup revealed no immune deficiency in our case. Chronic diarrhea, gastrointestinal surgery, vancomycin therapy, central venous catheter and TPN administration were important risk factors for *Leuconostoc* infection.

Even though *Leuconostoc* species are isolated from vaginal and fecal samples, they are not considered to be normal flora members. Site of entry is not clear in infections due to *Leuconostoc* species. Few reports speculate that the site of entry can be skin (16,17). Co-infection with coagulase negative staphylococci raise the suspicion of entry through skin (3,10,16). As most cases of *Leuconostoc* infection have central catheter, speculations about disrupted skin integrity during catheter insertion is the main cause (3,16,17). Others speculate that gastrointestinal system is another entry site. They state that when the gastrointestinal system is colonized with *Leuconostoc*, translocation occurs (3,15,16). In another report, 35 percent of cases have polymicrobial etiology, so intra-abdominal source may be suspected (13). Moreover, *Leuconostoc* species are isolated from infant formula, various foods, gastric aspirates, gastrostomy tubes

and these support the idea of gastrointestinal entry (15,18). Apart from that, a report by Bou et al. (6) documents TPN as the source of nosocomial *Leuconostoc* infections. It is clear that in our case there are multiple risk factors. It is hardly possible to tell which factor has the leading role, but it may be speculated that gastrointestinal route is more probable case due to chronic diarrhea. Simultaneous resolution of diarrhea and infection supports this hypothesis.

Management of *Leuconostoc* infections consists of appropriate antibiotic therapy and removal of infection source (catheter removal, draining of abscess) (3). The most preferred antibiotic for *Leuconostoc* infections is penicillin with or without gentamicin. Minimum inhibitory concentration value of penicillin is higher than *Streptococci* mandating a higher dose or combination with an aminoglycoside (3,10). In published reports, *Leuconostoc* infections are successfully treated by ampicillin, cefotaxime, carbapenem, clindamycin, erythromycin and recently by daptomycin (1,3,19). The characteristics of pediatric patients who have gastrointestinal disease with *Leuconostoc* bacteremia is shown in Table I (3).

Management of catheter related bacteremia is not clear. In most cases of catheter related bacteremia, catheter removal has been required (10,15). In three cases, bacteremia was resolved without antibiotic treatment following catheter removal (10,16,17). Guideline for catheter related blood stream infections does not contain information about *Leuconostoc* (20). No information regarding biofilm formation of *Leuconostoc* was found. Former reports show a tendency towards catheter removal for the control of infection. In our case, linezolid and ampicillin therapy was initiated following the identification of *Leuconostoc*. Vascular access problem mandated antibiotic lock therapy but the central line was accidentally removed. Another central catheter was inserted immediately. Blood cultures remained positive and the second catheter was also colonized with *Leuconostoc*. Due to catheter related blood stream infection, positive blood cultures and persistence of systemic signs required antibiotic lock therapy. The combined lock solution was prepared by mixing ampicillin 10 mg/mL and heparin 5000 units/mL for antibiotic lock therapy. Then, the solution was administered into the lumen of the catheter every 12 hours. Following lock therapy, blood cultures remained sterile and clinical signs improved. Systemic therapy with linezolid and ampicillin was continued for 21 days, while lock therapy lasted for 12 days. Three months follow-up revealed no problems.

Leuconostoc species should be kept in mind in cases of vancomycin resistant gram positive infections. Even though they are rare pathogens for humans, they may cause serious infections. In cases of catheter related blood stream infections where systemic antibiotic treatment fails and

Table I. The characteristics of pediatric patients who have gastrointestinal disease with *Leuconostoc* bacteremia (3)

C	Age	Sex	Primary disease	TPN	CVC	Previously vancomycin therapy	Treatment	Line removal
1	6 months	M	Gastroschisis and bowel infarction	+	+	-	Ampicillin (14 days)	No
2	3 years 6 months	F	Gastroschisis	+	+	Unknown	Ampicillin (14 days)	Yes (venous thrombosis)
3	6 months	M	Necrotizing enterocolitis	+	+	Unknown	Penicillin (14 days)	Yes
4	11 years	F	Midgut volvulus and congenital malrotation	+	+	-	Ampicillin and gentamicin (14 days)	Yes
5	8 months	F	Gastroschisis	+	+	-	Imipenem (14 days)	Yes
6	2 years	F	Jejunal atresia	+	+	+	Imipenem (17 days)	Yes
7	2 months	F	Necrotizing enterocolitis	-	+	+	Vancomycin clindamycin(14 days)	Yes
8	1 months	M	Necrotizing enterocolitis	+	+	+	Penicillin (14 days)	No
9	9 months	F	Jejunal atresia	+	+		Ampicillin + gentamicin (10 days)	No
10	20 months	F	Necrotizing enterocolitis	+	+	+	Amoxicillin (14 days)	Yes
11	4 years	M	Malrotation and volvulus	+	+	+	Amoxicillin (14 days)	No
12	7 months	F	Gastroschisis, small bowel and colonic atresia	+	+	+	Ampicillin + gentamicin	Yes
13	13 months	F	Gastroschisis, jejunalatresia	+	+	+	Penicillin (14 days)	Yes
14	10 months	M	Necrotizing enterocolitis	+	+	+	Ampicillin + gentamicin	No
15	1 years	F	Jejunal atresia	+	+	+	Vancomycin + cefotaxime + metronidazole	Yes
16	8 months	F	Hirschsprung and intestinal obstruction	+	+	+	Clindamycin + amikacin (16 days)	Yes
PC	1 months	F	Chronic diarrhea	+	+	+	Ampicillin + linezolid (21 days) andampicillin lock therapy (14 days)	No

TPN: Total parenteral nutrition, CVC: Central venous catheter, C: Cases, PC: Present case, F: Female, M: Male

catheter removal is impossible, antibiotic lock therapy may be a therapeutic option.

Ethics

Informed Consent: *Retrospective study.*
Peer-review: Externally peer-reviewed.

Authorship Contributions

Medical Practices: Adem Karbuz, Bilge Aldemir Kocabaş, Aytaç Yalman, Zarife Kuloğlu, Ahmet Derya Aysev, Erdal İnce, Concept: Adem Karbuz, Design: Adem Karbuz, Data Collection or Processing: Adem Karbuz, Bilge Aldemir Kocabaş, Aytaç Yalman, Analysis or Interpretation: Ergin Çiftçi, Literature Search: Adem Karbuz, Writing: Adem Karbuz.

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References

1. Arıas CA, Murray BE. Enteroccus species, Streptococcus bovis group, and Leuconostoc species, In: Mandell GL, Bennett JE, Dolin R (eds), Mandell, Douglas and Bennett's Principles and Practice of Infectious Diseases. 7th ed. Churchill Livingstone, Philadelphia. 2010: p: 2643-52.
2. Rogasa M, Sharpe ME. Species differentiation of human vaginal Lactobacilli. J Gen Microbiol 1960;23:197-201.
3. Florescu D, Hill L, Sudan D, Iwen PC. Leuconostoc bacteremia in pediatric patients with short bowel syndrome: case series and review. Pediatr Infect Dis J 2008;27:1013-9.
4. Buu-Hoi A, Branger C, Acar JF. Vancomycin-resistant streptococci or Leuconostoc sp. Antimicrob Agents Chemother 1985;28:458-60.
5. Taşkapılıoğlu, O, Bahar A, Yılmaz E, et al. Nosocomial Leuconostoc Pseudomesenteroides Meningitis: A Case Report and Review of the Literature. Balkan Med J 2011;28:216-8.
6. Bou G, Luis Saleta J, Sáez Nieto JA, et al. Nosocomial Outbreaks Caused by Leuconostoc mesenteroides subsp. mesenteroides. Emerg Infect Dis 2008;14:968-71.
7. Wonga DW, Yang, W, Vielemeyer O. Catheter-Related Leuconostoc Bacteraemia in a Pregnant HIV-Infected Woman. J Med Cases 2012:3:331-3.
8. Kocak F, Yurtseven N, Aydemir N, Yüksek A, Yavuz SS. A case of osteomyelitis due to Leuconostoc lactis. Scand J Infect Dis 2007;39:278-80.
9. Ling ML. Leuconostoc bacteraemia. Singapore Med J 1992:33:241-3.
10. Dhodapkar KM, Henry NK. Leuconostoc bacteremia in an infant with short-gut syndrome: case report and literature review. Mayo Clin Proc 1996;71:1171-4.
11. Tholpady SS, Sifri CD, Sawyer RG, Hazen KC, Pruett TL, Bonatti H. Leuconostoc pseudomesenteroides blood stream infection following liver transplantation. Ann Transplant 2010;15:61-6.
12. Ishiyama K, Yamazaki H, Senda Y, Yamauchi H, Nakao S. Leuconostoc bacteremia in three patients with malignancies. J Infect Chemother 2011;17:412-8.
13. Lee MR, Huang YT, Lee PI, et al. Healthcare-associated bacteraemia caused by Leuconostoc species at a university hospital in Taiwan between 1995 and 2008. J Hosp Infect 2011;78:45-9.
14. Casanova-Roman M, Rios J, Sánchez-Porto A, Gomar JL, Casanova-Bellido M. Leuconostoc bacteremia in a healthy infant. Minerva Pediatr 2003;55:83-6.
15. Janow G, Lambert B, Scheiner M, Rosen O, Goldman DL, Soghier L. Leuconostoc septicemia in a preterm neonate on vancomycin therapy: case report and literature review. Am J Perinatol 2009;26:89-91.
16. Handwerger S, Horowitz H, Coburn K, Kolokathis A, Wormser GP. Infection due to Leuconostoc species: six cases and review. Rev Infect Dis 1990;12:602-10.
17. Bernaldo de Quiros JC, Munoz P, Cercenado E, Hernandez Sampelayo T, Moreno S, Bouza E. Leuconostoc species as a cause of bacteremia: two case reports and a literature review. Eur J Clin Microbiol Infect Dis 1991;10:505-9.
18. Carapetis J, Bishop S, Davis J, Bell B, Hogg G. Leuconostoc sepsis in association with continuous enteral feeding: two case reports and a review. Pediatr Infect Dis J 1994;13:816-23.
19. Golan Y, Poutsiaka DD, Tozzi S, Hadley S, Snydman DR. Daptomycin for line-related Leuconostoc bacteraemia. J Antimicrob Chemother 2001;47:364-5.
20. Mermel LA, Allon M, Bouza E, et al. Clinical Practice Guidelines for the Diagnosis and Management of Intravascular Catheter-Related Infection: 2009 Update by the Infectious Diseases Society of America. Clin Infect Dis 2009;49:1-45.

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Endocrine Disrupting Chemicals: A Challenge to Child Health

Endokrin Sistemi Bozan Kimyasallar: Çocuk Sağlığına Bir Tehdit

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Keywords: Endocrine disrupting chemicals, endocrine disruptors, diethylstilbestrol, persistent organic pollutants

Anakhar Kelimeler: Endokrin bozan kimyasallar, endokrin bozucular, dietilstilbestrol, kalıcı organik kirletici

Dear Editor,

Endocrine-disrupting Chemicals: Definition

The World Health Organization (WHO), defines endocrine disrupting chemicals (EDC) as substances that alter one or more functions of the endocrine system and consequently cause adverse health effects in an intact organism, or its progeny, or (sub) populations (WHO, International Programme on Chemical Safety) (1). They are broadly classified as estrogenic, anti-estrogenic, anti-androgenic and thyroid hormone disrupting compounds (1). The toxic effects of in-utero exposure to thalidomide and diethylstilbestrol (DES) in the 1960s and 1970s furnished the earliest knowledge on the fragility of the developing fetus, and led to the framing of "fragile fetus" concept by Howard Ben to depict the vulnerability of the developing fetus on exposure to environmental chemicals with endocrine properties (2). Theo Colborn, a wildlife biologist and the author of "Our stolen future", made the first modern day observations on health effects of EDCs on wildlife and human population and inferred that endocrine changes affecting fetus in-utero may have effects on many future generations to come (3).

Endocrine Disrupting Chemicals are Ubiquitous

An approximate 800 chemicals are suspected to adversely affect hormone synthesis, hormone receptors, or hormone conversion (1,2,4). EDCs may be natural (phytoestrogens in plants or fungal estrogens), or manmade (3,5). Examples of natural EDCs include isoflavonoids in soybeans, legumes; ligananes in grains, fruits and vegetables and coumestans in clover, and alfalfa among others (3,5). The synthetic compounds have assorted applications in today's world as pesticides, flame retardants, plastic additives, active ingredients in pharmaceuticals, food additives and contaminants, plastics, textiles, construction materials, hormonal therapies, personal care products and cosmetics which may result in residues and contaminants in food and other products (3,5). DES, a synthetic estrogen prescribed for

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preventing miscarriage between the 1940s-1970s was one of the first established examples of EDCs when a cluster of a new rare form of cancer, vaginal clear cell adenocarcinoma was identified in adolescent daughters of women who had taken the drug during pregnancy. It was also determined to result in various other benign reproductive tract abnormalities in prenatally exposed males and females (2).

Exposure to Endocrine Disrupting Chemicals

Humans are exposed to EDCs via ingestion of food, dust and water, inhalation of gases and particles in the air and dermal uptake (3). EDCs are also transferred to a developing fetus, or infant by transplacental route and breast milk (3). EDCs mimic or antagonize the effects of endogenous hormones, disrupt the synthesis of endogenous hormones or their receptors, or may alter target cell sensitivity (6). The mechanisms by which EDCs affect developmental events are identified to be numerous and include changes in the neuro-endocrine system, epigenetic mechanisms and/or direct effects on gene expression (2).

Vulnerability of Children and Their Windows of Exposure

Children are at risk of higher exposures to these chemicals due to their hand-to-mouth activity and higher metabolic rate (3). They also have immune systems which are yet to develop completely (3). Their exploratory or probing behavior and ignorance of impending risks maximize their contact with harmful chemicals in the environment during developmental period (3). Recent decades have witnessed an unusual spurt in the incidence of genital malformations, infertility due to low semen quality, adverse pregnancy outcomes, neuro-behavioral disorders associated with thyroid disruption, endocrine-related cancers (breast, endometrial, ovarian, prostate, testicular, thyroid), premature thelarche, obesity and Type II diabetes mellitus (4). Congenital disorders such as cryptorchidism, hypospadias, early puberty and thyroid dysfunction have also been shown to have clear endocrine association (1-3).

Common examples of exposure to EDCs include Bisphenol A used in the manufacture of polycarbonate plastics and epoxy resins which have been found to be causative for obesity and polycystic ovarian syndrome (2,7). Phthalates used in nail polish, hair spray, deodorants and shampoos have been found to be associated with impaired genital development in male children (7). Lavender and tea tree oil on repeated use can stimulate estrogenic activity and cause male pubertal gynaecomastia (7). Flame retardants used in car seats and table pads may be inhaled by or absorbed through the skin of the baby and cause a greater risk of tumors (7). Early exposure to lead has been found to cause significant changes in hypothalamo-pituitary-adrenal axis while maternal smoking has been associated with obesity (7).

Apart from the above examples, some of these chemicals such as persistent organic pollutants (POPs), methylmercury which enter the bodies of younger children persist for a longer time due to their long half-lives and present their harmful effects later in life or cause multigenerational effects (2,3). The Stockholm Convention (2011) ratified by the international community recommended the elimination or phasing out of POPs (3). Some EDCs like dichloro-diphenyl-trichloroethane banned years ago in some countries, still persist in the environment and human bodies and manifest in older age groups (7).

Characteristics of Endocrine Disrupting Chemical Exposure

EDCs often produce their impacts with relatively low doses (3,7). Most EDCs do not have traditional dose-response curves (3,7,8). The timing of exposure decides the magnitude of impact (3,7,8). Effects such as learning difficulties, increased susceptibility and sensitivity to infections, testicular dysgenesis syndrome, infertility, fibroids, premature menopause, obesity, atherosclerosis, cardiovascular disease, Alzheimer's disease, Parkinson's disease, breast and prostate cancers manifest after a variable latent period depending on the time and the specific tissue exposed (3,7). Multi-chemical exposures are frequent and often have additive or synergistic potential (3,8).

Challenges

A large number of EDCs and their sources are yet to be identified. The effects of human exposure and their mechanisms of action have not been clearly understood for many EDCs. There are no clear guidelines for testing the effects of EDCs. The most challenging aspects of EDCs are their ubiquitous nature and exposure, their ability to cause a wide range of health effects with minimal doses, and the persistence of the resulting biological effects which are sometimes multigenerational (7).

Actions Required

Prevention of exposure is the single most effective measure to protect children against these toxic chemicals. But prevention of exposure may not be an immediate possibility due to the ubiquitous and often masked presence of EDCs in the environment. Exposure control could be effectively implemented as a short-term measure. Exposure control of lead has been found to have proven favorable consequences (4). Table I discusses the strategies and actions required to efficiently control EDCs.

All the above measures would be incomplete without sensitizing the health care workers and health professionals who encounter children in their day-to-day practice.

Pediatricians, general health practitioners and other health-care personnel are a significant resource-group with frequent contact with individual families. They have to be sensitized to enquire about the child's environment with specific queries for EDCs apart from regular arthropods and insects (3). History-taking at the onset of reproductive disorders should include careful assessment of occupational and environmental exposure of the individual and the close family (8). Pediatricians can play a role in promoting the development of model programs and

Table I. Overview of strategies and actions required to control endocrine disrupting chemicals

Strategies	Actions required
Strengthening knowledge on EDCs (4)	-Knowledge on EDCs should be imparted to all streams of students in schools and colleges. -The general population should be educated through mass media and local representatives. -Community level decisions and strict adherence are required for safe use of substances involving EDCs. -Production and consumption of organic food products should be encouraged.
Enabling research environment (4)	-To identify EDCs (4). -To identify methods to evaluate evidence (4). -To develop validated screening and testing systems (4). -To promote development of validated biomarkers (8). -To strengthen existing systems in accurate hazard identification and toxicity prediction (4). -Populations and communities with EDC exposure should be identified and exposure response studies should be conducted (8). Hotspots with unexpectedly high prevalence of suspected disorders should be watched for and identified early (8).
Reducing exposure and vulnerability (4)	-Community education, education of parents and teachers is an essential need. -Stringent regulatory measures for manufacturing and processing units to curb the use of toxic chemicals and promote the use of harmless, neutral substitutes. -Restrain indiscriminate discharge of effluents into the environment Implement strict measures to motivate manufacturers in proper labelling and declaration of constituent ingredients of products (4). -Consumers should be educated to peruse the relevant ingredient details (4). -Promote safe agricultural practices with the restriction of the use of pesticides and other chemicals.
Commitment from policy-makers	-EDCs with proven toxic effects have to be banned. -For EDCs where total ban is impossible, an initial ban of residential use can be promoted. -Substitution with non-toxic or less toxic substances should be promoted. -Develop guidelines for the disposal of industrial, agricultural and pharmaceutical waste and ensure their strict implementation (3). -Enabling intersectoral coordination for efficient implementation of programmes. -Poverty alleviation measures to enable improved housing, cooking fuel to help avoid environmental exposure. -Provide simple, specific messages and guidelines for general public to help them protect themselves.

EDCs: Endocrine disrupting chemicals

practices in the communities and schools of their patients (9). An integrated team approach involving pediatricians, clinical endocrinologists, environmental toxicologists and epidemiologists will strengthen study designs and help in the increased understanding of the associations (10).

While it is a substantial challenge to identify and regulate the use of these chemicals, immediate and effective measures against EDCs provide a tremendous opportunity to improve child health and contribute to a healthy human resource. It is the supreme responsibility of today's citizens, national and international governing bodies to recognize and act upon this imminent threat to child health and provide our children with a healthy future.

Ethics

Peer-review: Internally peer-reviewed.

Authorship Contributions

Concept: Geetha Mani, Design: Geetha Mani, Raja Danasekaran, Literature Search: Geetha Mani, Kalaivani Annadurai, Writing: Geetha Mani, Review and Approval of the Final Draft: Geetha Mani, Raja Danasekaran, Kalaivani Annadurai.

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References

1. World Health Organization. Possible developmental early effects of endocrine disrupters on child health. Geneva, World Health Organization. 2012. Accessed from www.who.int/phe/en Last accessed on November 1, 2016.
2. Newbold RR. Developmental exposure to endocrine-disrupting chemicals programs for reproductive tract alterations and obesity later in life. Am J Clin Nutr 2011;94(6 Suppl):1939S-42S.
3. World Health Organization. Children's Health and Environment. WHO Training package for the Health Sector. World Health Organization, Geneva. 2011. Available at www.who.int/ceh Accessed on November 2, 2016.
4. United Nations Environment Programme and World Health Organization. State of the Science of Endocrine Disrupting Chemicals 2012. Summary for Decision-Makers. Geneva, World Health Organization. 2013. Available at www.who.int/phe/en Accessed on November 1, 2016.
5. World Health Organization (WHO). Global assessment of the state-of-the-science of endocrine disruptors. Geneva, WHO, 2002. Available at www.who.int/ipcs/publications/new_issues/endocrine_disruptors/en/index.html Accessed on April 15, 2014.
6. Amaral Mendes JJ. The endocrine disrupters: a major medical challenge. Food Chem Toxicol 2002;40:781-8.
7. National Institute of Environmental Health Sciences. Child development and environmental toxins. Available at http://www.niehs.nih.gov/health/assets/docs_a_e/child_development_and_environmental_toxins_508.pdf Accessed on November 2, 2016.
8. Diamanti-Kandarakis E, Bourguignon J, Giudice LC, et al. Endocrine-disrupting chemicals: an Endocrine Society scientific statement. Endocr Rev 2009;30:293-342.
9. Pesticide exposure in children- Policy statement. American Academy of Paediatrics. Paediatrics 2012;130:1757-63.
10. Fleisch AF, Wright RO, Baccarelli AA. Environmental epigenetics: a role ain endocrine disease? J Mol Endocrinol 2012;49:R61-7.

THE JOURNAL OF PEDIATRIC RESEARCH

Yazarlık, Yayın Hakkı Devri, Maddi Yardım ve Teşekkür-Kabul İzni

TEŞEKKÜR VE KABUL BEYANI BÖLÜMÜ, SORUMLU YAZAR TARAFINDAN İMZALANMALI. SON BÖLÜM İSE MAKALEDE İSMİ GEÇEN BÜTÜN YAZARLAR TARAFINDAN İMZALANMALIDIR.

MAKALE BAŞVURUSUNDA FORM DOLDURULARAK ONLİNE SİSTEME YÜKLENMELİDİR.

BU FORM GEREKİRSE, İMZA İÇİN HER BİR YAZAR TARAFINDAN DOLDURULMAK ÜZERE FOTOKOPİ İLE ÇOĞALTILABİLİR.

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