Larissa Ferreira Silva

Análise do perfil leucocitário de pacientes com periodontite crônica e diabetes mellitus tipo 2

Brasília
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Trabalho de Conclusão de Curso apresentado ao Departamento de Odontologia da Faculdade de Ciências da Saúde da Universidade de Brasília, como requisito parcial para a conclusão do curso de Graduação em Odontologia.

Orientador: Prof. Dra. Maria do Carmo Machado Guimarães

Co-orientador: Ms. Priscilla Naiff

Brasília
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Aos meus pais, por simplificarem a trajetória da vida com sabedoria e grandeza de coração.

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EPÍGRAFE

“E ali logo em frente, a esperar pela gente, o futuro está.
E o futuro é uma astronave que tentamos pilotar,
Não tem tempo nem piedade, nem tem hora de chegar.
Sem pedir licença muda nossa vida, depois convida a rir ou chorar.”
Toquinho

RESUMO

Silva, Larissa Ferreira. Análise do perfil leucocitário de pacientes com periodontite crônica e diabetes mellitus tipo 2. 2016. Trabalho de Conclusão de Curso (Graduação em Odontologia) – Departamento de Odontologia da Faculdade de Ciências da Saúde da Universidade de Brasília.

A periodontite e o diabetes mellitus são doenças crônicas que possuem uma relação bidirecional, apresentando repercussões clínicas, onde o decurso de uma influencia negativamente a outra. Material e Métodos: Esse estudo contou com a participação de 25 pacientes, divididos em três grupos: periodontite (n=10), periodontite e diabetes mellitus tipo 2 (n=10) e controle, saudável (n=5). Foram realizados exame periodontal completo e coleta de sangue periférico de todos os grupos para análise do leucograma. Resultados: A contagem total de leucócitos foi maior no grupo controle (7200 ± 1927 /mm^3), seguida pelo grupo periodontite e diabetes (6600 ± 3398 /mm^3) e foi menor no grupo periodontite (5970 ± 2653 /mm^3). Nenhum grupo apresentou diferenças em relação aos valores de referência (3600 ± 11000 /mm^3). A contagem diferencial revelou uma tendência ao aumento de monócitos no grupo periodontite e diabetes (550 ± 183 / mm^3), ainda assim não foi significativa ($p > 0,05$). Nenhum grupo apresentou quadros de leucocitose ou leucopenia. Conclusão: A periodontite associada ou não ao diabetes mellitus não causou aumento no número de células de defesa em nível sistêmico.

ABSTRACT

Silva, Larissa Ferreira. White cell count of patients with chronic periodontitis and diabetes mellitus type 2. 2016. Undergraduate Course Final Monograph (Undergraduate Course in Dentistry) – Department of Dentistry, School of Health Sciences, University of Brasília.

Background: Periodontitis and diabetes mellitus are chronic diseases with a bidirectional relationship, presenting clinical impact, where one's courses affects the other negatively. Methods: 25 patients were included in the present study, who were allocated into three groups: periodontitis (n=10), periodontitis and diabetes type 2 (n=10) and control, healthy (n=5). Clinical periodontal examination was performed and venous peripheral blood was collected for leukocyte analyses. Results: The total leukocyte count was higher in the control group (7200 ± 1927 / mm^3), followed by the periodontitis and diabetes group (6600 ± 3398 / mm^3) and was lower in the periodontitis group (5970 ± 2653 / mm^3). Differential leukocyte count showed a tendency to an increase of monocyte in the periodontitis and diabetes group (550 ± 183/ mm^3), with no statistically significant differences among the others groups ($p>0.05$). Diagnosis of leukocytosis or leukopenia was not observed in any group. Conclusion: Periodontitis did not increase the number of defense cells even when associated with diabetes.

SUMÁRIO

ARTIGO CIENTÍFICO

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SILVA, Larissa Ferreira; NAIFF, Priscilla Farias; GUIMARÃES, Maria do Carmo Machado. Análise do perfil leucocitário de pacientes com periodontite crônica e diabetes mellitus tipo 2.

Apresentado sob as normas de publicação do Journal of Periodontology.

FOLHA DE TÍTULO

Análise do perfil leucocitário de pacientes com periodontite crônica e diabetes mellitus tipo 2

White cell count of patients with chronic periodontitis and diabetes mellitus type 2

Larissa Ferreira Silva[1]
Priscilla Farias Naiff[2]
Maria do Carmo Machado Guimarães[3]

[1] Aluna de Graduação em Odontologia da Universidade de Brasília.
[2] Doutoranda do programa de Ciências da Saúde da Universidade de Brasília.
[3] Professora Adjunta de Periodontia da Universidade de Brasília.

Correspondência: Prof. Dr. Maria do Carmo Machado Guimarães
Campus Universitário Darcy Ribeiro - UnB - Faculdade de Ciências da Saúde - Departamento de Odontologia - 70910-900 - Asa Norte - Brasília - DF
E-mail: mmgcarmo@gmail.com / Telefone: (61) 31071849

RESUMO

Análise do perfil leucocitário de pacientes com periodontite crônica e diabetes mellitus tipo 2

Resumo

A periodontite e o diabetes mellitus são doenças crônicas que possuem uma relação bidirecional, apresentando repercussões clínicas, onde o decurso de uma influencia negativamente a outra. Material e Métodos: Esse estudo contou com a participação de 25 pacientes, divididos em três grupos: periodontite (n=10), periodontite e diabetes mellitus tipo 2 (n=10) e controle, saudável (n=5). Foram realizados exame periodontal completo e coleta de sangue periférico de todos os grupos para análise do leucograma. Resultados: A contagem total de leucócitos foi maior no grupo controle (7200 ± 1927 /mm3), seguida pelo grupo periodontite e diabetes (6600 ± 3398 /mm3) e foi menor no grupo periodontite (5970 ± 2653 /mm3). Nenhum grupo apresentou diferenças em relação aos valores de referência (3600 ± 11000 /mm3). A contagem diferencial revelou uma tendência ao aumento de monócitos no grupo periodontite e diabetes (550 ± 183 / mm3), ainda assim não foi significativa (p> 0,05). Nenhum grupo apresentou quadros de leucocitose ou leucopenia. Conclusão: A periodontite associada ou não ao diabetes mellitus não causou aumento no número de células de defesa em nível sistêmico.

Palavras-chave

Contagem de leucócitos, Diabetes mellitus tipo 2, Periodontite crônica

Relevância Clínica

Uma vez que diabetes mellitus e periodontite consistem em doenças crônicas resultantes de vias inflamatórias, a associação entre ambas tem sido investigada. A influência da inflamação local do periodonto sobre os níveis séricos de marcadores inflamatórios e das células de defesa do sangue periférico constitui um importante objeto de estudo dessa associação.

ABSTRACT

White cell count of patients with chronic periodontitis and diabetes mellitus type 2

Abstract

Background: Periodontitis and diabetes mellitus are chronic diseases with a bidirectional relationship, presenting clinical impact, where one's courses affects the other negatively. Methods: 25 patients were included in the present study, who were allocated into three groups: periodontitis (n=10), periodontitis and diabetes type 2 (n=10) and control, healthy (n=5). Clinical periodontal examination was performed and venous peripheral blood was collected for leukocyte analyses. Results: The total leukocyte count was higher in the control group (7200 ± 1927 / mm3), followed by the periodontitis and diabetes group (6600 ± 3398 / mm3) and was lower in the periodontitis group (5970 ± 2653 / mm3). Differential leukocyte count showed a tendency to an increase of monocyte in the periodontitis and diabetes group (550 ± 183/ mm3), with no statistically significant differences among the others groups ($p>0.05$). Diagnosis of leukocytosis or leukopenia was not observed in any group. Conclusion: Periodontitis did not increase the number of defense cells even when associated with diabetes.

Keywords

Leucocyte count; Diabetes mellitus type 2; Chronic periodontitis

INTRODUÇÃO

A periodontite e o diabetes mellitus são doenças crônicas resultantes de vias inflamatórias, que possuem uma relação bidirecional que vem sendo extensivamente pesquisada.

O diabetes mellitus constitui um grupo de doenças metabólicas caracterizadas por níveis elevados de açúcar no sangue (hiperglicemia), resultantes do defeito na secreção e/ou ação da insulina. O diabetes tipo 2 é o tipo mais comum e resulta da resistência à insulina; isto é, há redução na resposta das células do organismo à insulina, resultando em capacidade diminuída para se transferir glicose da circulação para o meio intracelular. [1]

Pacientes com diabetes possuem altos níveis de marcadores imunológicos no sangue, o que dificulta o controle glicêmico, devido a resistência à insulina. Alguns estudos mostram presença de diabetes como um fator que aumenta em três vezes o risco à doença periodontal. [2, 3] Pode-se observar que pacientes com diabetes e dificuldade no controle glicêmico têm maior probabilidade de desenvolver doença periodontal grave, ao passo que a doença periodontal pode dificultar o controle glicêmico destes pacientes. [4,5,6]

As alterações mais notáveis no diabetes não controlado são a redução nos mecanismos de defesa e suscetibilidade aumentada a infecções, levando à doença periodontal destrutiva. De fato, a doença periodontal é considerada a sexta complicação do diabetes. [7]

A periodontite crônica é uma doença de caráter infeccioso e inflamatório que acomete o tecido periodontal de suporte dos indivíduos acometidos e sua progressão envolve reabsorção do osso alveolar, destruição do ligamento periodontal, além da formação de bolsas, podendo levar à perda dentária. [8]

Os neutrófilos representam mais de 65% dos leucócitos do sangue periférico e, no processo de defesa do hospedeiro, são as primeiras células envolvidas. A função neutrofílica pode estar alterada na periodontite e, a fisiopatologia da doença periodontal pode estar centrada na disfunção dos neutrófilos. [9]

A periodontite crônica tem como fator etiológico primário a infecção dos tecidos periodontais por algumas bactérias, principalmente *Aggregatibacter actinomycetemcomitans*, *Porphyromonas gingivalis*, Tannerella forsythia, *Prevotella intermedia*, *Prevotella nigrescens*, *Fusobacterium nucleatum*. Tais patógenos, dependendo de outras condições, como aquelas relacionadas à resposta do hospedeiro, são capazes de estimular o sistema imune, que tenta conter o processo infeccioso. Os neutrófilos, leucócitos que participam da primeira linha de defesa durante o processo inflamatório, produzem moléculas específicas, que podem levar a uma resposta inflamatória exacerbada local ou sistêmica, dependendo das condições de saúde do indivíduo.[10,11] Alterações na contagem total de leucócitos provenientes de sangue periférico e, especificamente, na contagem de neutrófilos, podem indicar a presença de processos ou doenças infecciosas e/ou inflamatórias.[12,13,14] A importância da contagem de leucócitos em diferentes níveis de gravidade da periodontite ainda não foi suficientemente investigada, sendo os estudos recentes e escassos.

Este trabalho tem como objetivo determinar a relação entre as células de defesa presentes no sangue periférico com a periodontite crônica associada ou não ao diabetes mellitus tipo 2.

MATERIAL E MÉTODOS

Os critérios estabelecidos para a execução deste estudo foram de acordo com as normas da Resolução 466/2012 do Conselho de Saúde do Ministério da Saúde Pública e Código de Ética Profissional Odontológico, segundo a Resolução CFO-

118/2012. Todos os indivíduos foram informados verbalmente e por escrito do objetivo do estudo e, ao concordarem em participar da pesquisa, assinaram o termo de consentimento Livre e Esclarecido (TCLE). O projeto foi submetido ao Comitê de Ética em Pesquisa da Universidade de Brasília (UnB) e foi aprovado sob CAAE 46609515.7.0000.0030 e parecer 1.257.137.

A seleção das amostras foi conduzida na Clínica do Curso de Odontologia da Universidade de Brasília (UnB), no Hospital Universitário de Brasília (HUB), em Brasília/DF. O tipo de amostragem foi por conveniência, de forma que foram constituídos três grupos: 1- indivíduos com diagnóstico de periodontite crônica sem doenças sistêmicas, 2- indivíduos com periodontite crônica associada ao diabetes mellitus do tipo 2 e 3- Indivíduos saudáveis sistemicamente e com periodonto saudável clinica e radiograficamente (controle).

Para os grupos testes (1 e 2) foram selecionados indivíduos baseando-se nos seguintes critérios: presença de, no mínimo, 10 dentes e, no mínimo dois sítios periodontais interproximais, em dentes diferentes, com profundidade de sondagem ≥ 4 mm e, ao menos, dois sítios interproximais com perda de inserção ≥ 3 mm; idade ≥30 anos.

O grupo controle compreendeu indivíduos com idade ≥ 30 anos, com o periodonto saudável clínica e radiograficamente e, com presença de, no mínimo, 20 dentes (com profundidade de sondagem clínica ≤3 mm, nível de inserção clínica ≤3 mm, ≤10% dos sítios com sangramento à sondagem).

Foram excluídos do estudo pacientes que haviam realizado tratamento periodontal nos últimos seis meses; que tivessem feito uso continuado de medicamentos como antimicrobianos, imunomoduladores, anti-inflamatórios ou corticosteroides, nos últimos três meses; indivíduos com doenças sistêmicas que interferiam na condição periodontal, com exceção do diabetes mellitus tipo 2 (HIV, cardiopatias, câncer, desordens imunes, infecções agudas, alergias graves, doenças

gastrointestinais ou renais, obesidade mórbida [IMC 40 kg/m2] ou abaixo do peso [mal nutridos IMC <18,5 kg/m2], etc); gestantes ou lactantes; histórico positivo de tabagismo; idade < 30 anos; etnias indígenas.

Os parâmetros periodontais clínicos analisados foram: profundidade de sondagem (PS), nível de inserção clínico (NIC), índice de placa visível (IP) [15] e índice de sangramento à sondagem (ISS)[16]. No exame clínico foram registrados seis sítios por dente (vestibular, mésio-vestibular, disto-vestibular, lingual, mésio-lingual e disto-lingual) com o auxílio de uma sonda periodontal Michigan 0 com marcações de Williams (Millenium, Brasil). Com exceção dos terceiros molares, todos os dentes presentes na boca foram examinados. O exame clínico foi realizado por um único examinador (P.F.N).

Para análise bioquímica do sangue, foi realizada a coleta de sangue venoso no laboratório de análises clinicas Sabin-DF para avaliação dos seguintes dados: hemograma completo, HbA1C e glicemia em jejum .

Após a coleta das amostras, todos os indivíduos com periodontite foram submetidos a tratamento e acompanhamento periodontal e os indivíduos sem doença periodontal receberam orientação de higiene bucal e profilaxia profissional, além de outro tratamento odontológico, quando necessário.

Para análise estatística foi utilizado o programa GraphPad Prism 5 e todos os dados foram submetidos ao teste Kruskal-Wallis.

RESULTADOS

As características sociodemográficas dos pacientes estão na tabela 1. A amostra do estudo incluiu 25 indivíduos, que foram divididos como grupo controle (cinco pacientes), grupo periodontite (dez pacientes) e grupo periodontite e diabetes (dez pacientes). O grupo periodontite e diabetes foi o grupo com a

maior média de idade (51± 5,5 anos), e IMC (30,82 ± 5,2 Kg/m^2). Quanto aos anos de estudo, o grupo controle foi o que apresentou maior escolaridade (16 ± 3 anos).

Tabela 1. Dados sociodemográficos dos pacientes

	PERIODONTITE	PERIODONTITE E DIABETES	CONTROLE
SEXO (F/M)	6/4	5/5	4/1
IDADE (anos*)	43,5 ± 9,5	51 ± 5,5	43,6 ± 12,6
IMC (kg/m^2*)	30,82 ± 6,1	26,27 ± 5,2	22,24 ± 1,7
ESCOLARIDADE (ANOS*)	13 ± 3	13 ±2	16 ± 3

*Valores expressos em média/desvio padrão

Os parâmetros periodontais mostram diferença significativa nos valores de ISS, IP, NIC e PS entre os grupos avaliados. Essa diferença está relatada na Tabela 2.

Tabela 2. Dados clínicos periodontais

	PERIODONTITE	PERIODONTITE E DIABETES	CONTROLE	p valor
ISS	43%	36,7%	≤ 10	0,0097
IP	97,5%	78,5%	≤ 15	0,0017
NIC	4,7 mm	4,3 mm	≤ 2	0,0022
PS	5 mm	4,8 mm	≤ 3	0,0021

*Valores expressos em mediana. p significativo quando <0,05

Os valores de glicose e hemoglobina glicada (HbA1C) encontram-se na Tabela 3.

Tabela 3. Glicose e hemoglobina glicada dos pacientes

	PERIODONTITE	PERIODONTITE E DIABETES	CONTROLE	p valor
Glicose (mg/dl)	93 ± 12	134 ± 27	97 ± 10	0,0016
Hemoglobina glicada (%)	5,4 ± 0,4	7,4 ± 1,1	5,5 ± 0,4	0,0074

Valores expressos em mediana/desvio padrão. p significativo quando <0,05

O valor da mediana referente à contagem de leucócitos totais (CTL) dos pacientes do estudo foi de 6520 ± 2524/mm^3. O grupo periodontite apresentou a menor mediana (5970± 2653/mm^3) de contagem total de leucócitos. Os valores não demonstram diferença em relação aos valores de referência, indicando que não houve quadros de leucocitose ou leucopenia entre os participantes do estudo. O resultado dos valores referentes às medianas das contagens de leucócitos totais entre os grupos encontra-se na Tabela 4. Os bastonetes tiveram o valor 0 em todos os grupos. O grupo periodontite e diabetes apresentou uma discreta tendência ao aumento no número de monócitos, em comparação aos grupos periodontite e controle, contudo essa diferença não foi estatisticamente significativa (p> 0,05) Figura 1. Os valores para segmentados, basófilos, eosinófilos, linfócitos e monócitos encontram-se na Tabela 5.

Tabela 4. Contagem Total de Leucócitos (milhões/mm^3)

PERIODONTITE	PERIODONTITE E DIABETES	CONTROLE	VALORES DE REFERÊNCIA
5970 ± 2653	6600 ± 3398	7200 ± 1927	3600 A 11000

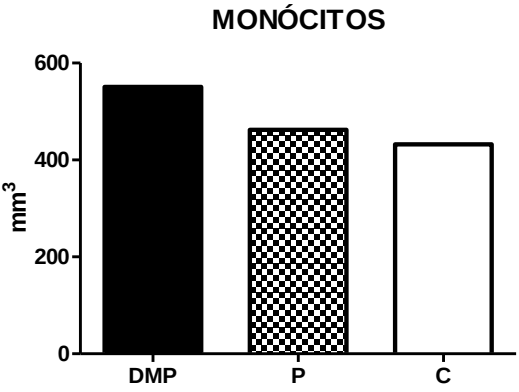
*Valores expressos em mediana/desvio padrão

Tabela 5. Contagem Diferencial de Leucócitos (milhões/mm^3)

	PERIODONTITE	PERIODONTITE E DIABETES	CONTROLE	VALORES DE REFERÊNCIA	p valor
Bastonetes	0 ± 0	0 ± 147	0 ± 0	0 a 550	0,4724
Segmentados	3468 ± 1132	3855 ± 2683	4218 ± 1049	1480 a 7700	0,8571
Eosinófilos	159 ± 150	155 ± 143	144 ± 130	0 a 550	0,6171
Basófilos	0 ± 32	0 ± 28	0 ± 26	0 a 220	0,9006
Linfócitos	2086 ± 558	2253 ± 608	2378 ± 712	740 a 5500	0,6403
Monócitos	462 ± 80	550 ± 183	432 ± 219	37 a 1100	0,4050

*Valores expressos em mediana/desvio padrão. p significativo quando <0,05.

Figura 1. Diferenças na contagem de monócitos entre os grupos.
DMP: Diabetes e periodontite, P: periodontite, C: controle.



DISCUSSÃO

O diabetes consiste em uma doença crônica que leva ao aumento da glicose no sangue, estando o paciente sob constante estado de hiperglicemia. Na infecção periodontal, a atividade fagocitária dos monócitos pode suscitar um estado de resistência à insulina, gerando uma hiperglicemia.[17] A periodontite e o diabetes associados alimentam uma via de mão dupla, onde o decurso de uma doença pode influenciar o quadro clínico da outra.[1,18] Os mecanismos que relacionam as doenças periodontais e o diabetes não estão totalmente elucidados, mas envolvem aspectos da inflamação, ativação imunológica, atividade dos neutrófilos, além de outros leucócitos e a produção de diversas citocinas.[19]

As células de defesa do corpo, também conhecidas como leucócitos, desempenham os principais mecanismos de defesa do organismo frente à invasão microbiana.[20] Em uma infecção bacteriana, os neutrófilos são as células predominantes inicialmente, além de desempenharem um papel importante no decurso da inflamação e patogênese.[10]

O presente trabalho não encontrou diferença estatisticamente significativa ($p>0,05$) entre os grupos, em

relação aos parâmetros hematológicos, em oposição ao que encontramos na literatura. Kweider et al., 1993, mostraram que pacientes com periodontite possuíam níveis de fibrinogênio e número de leucócitos significativamente maiores quando comparados a indivíduos do grupo controle. Esse estudo sugere, ainda, que a alteração nesses parâmetros relacionados à doença periodontal pode indicar um maior risco de infarto do miocárdio.[21]

Um estudo conduzido em 2011 por Pejčić et al. concluiu que um aumento na contagem total de leucócitos e diferença de neutrófilos em pacientes com periodontite, especialmente em sua forma grave, pode ser um indicador da possível exposição do corpo para alguma doença sistêmica, podendo representar um importante alerta para os médicos encaminharem seus pacientes a um cirurgião-dentista.[14]

Nossos dados corroboram com os resultados encontrados no estudo de Banthia et al., 2013, que, ao comparar um grupo saudável e um grupo com periodontite, não observou diferença significativa estatisticamente entre os mesmos. Siddeshappa et al., 2016, compararam os resultados do leucograma de pacientes com periodontite, antes e depois do tratamento periodontal não cirúrgico, e também não encontraram diferença estatística significante. Esses estudos incluíram a fase de tratamento periodontal não cirúrgico, o que gerou um decréscimo na contagem de leucócitos total e de células individuais, ainda que essa diferença não tenha sido significativa.[11,22]

O trabalho de Carneiro et al, 2011, mostra, em uma análise longitudinal, que não houve diferença estatisticamente significativa entre as células de defesa do sangue periférico avaliadas antes e após terapia periodontal não cirúrgica. Em uma análise transversal do mesmo, averiguamos que as células de defesa dos pacientes são semelhantes aos achados do nosso estudo, não havendo ainda diferença no perfil leucocitário entre os grupos.[9]

O presente estudo objetivou avaliar as células de defesa do sangue periférico, no entanto, na literatura há trabalhos que objetivaram avaliar os leucócitos polimorfonucleares presentes na saliva, como os estudos de Landzberg et al., 2015, Bhadbhade et al., 2012 e Andersen & Cimasoni, 1993. Esses estudos concluíram que existe uma alta correlação positiva entre os leucócitos polimorfonucleares bucais e a gravidade da doença periodontal.[23,24,25] Ainda são necessários testes estatísticos específicos para verificar se a mesma relação é presente avaliando o sangue dos indivíduos.

CONCLUSÃO

O presente estudo concluiu que a inflamação gerada pela periodontite crônica não gerou leucocitose em nível sistêmico. As contagens de tais células se mantiveram dentro dos valores de referência e não apresentaram diferença estatisticamente significativa entre os grupos. No entanto, há necessidade de se ampliar a amostra do estudo para se confirmar tais observações. A tendência ao aumento de monócitos ainda precisa ser investigada e testes específicos devem ser realizados a fim de estabelecer a relação entre essas células e a doença periodontal associada ou não ao diabetes mellitus tipo 2.

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ANEXOS

NORMAS DA REVISTA

This update includes a general reorganization of the author guidelines as well as improved navigation. Other changes include online-only publication, conflict of interest form collection, and updates to clinical trial registration requirements.

The Journal of Periodontology publishes articles relevant to the science and practice of periodontics and related areas. Manuscripts are accepted for consideration with the understanding that text, figures, photographs, and tables have not appeared in any other publication, except as an abstract prepared and published in conjunction with a presentation by the author(s) at a scientific meeting, and that material has been submitted only to this journal.

The Journal of Periodontology accepts manuscript submissions online at ScholarOne Manuscripts. To start a new submission, enter the Author Center and click "Click here to submit a new manuscript." Details regarding each submission step are located at the top of the page in ScholarOne Manuscripts. Authors should prepare manuscripts in accordance with the instructions below. Failure to do so may result in delays or manuscript unsubmission.

MANUSCRIPT CATEGORIES AND SPECIFIC FORMATS

Submissions to the Journal of Periodontology should be limited to one of the categories defined below. Specific information regarding length and format is provided for each category. Please

also refer to the instructions provided under General Format and Style. All manuscripts will be reviewed by the Editors for novelty, potential to extend knowledge, and relevance to clinicians and researchers in the field. Some manuscripts will be returned without review, based on the Editors' judgment of the appropriateness of the manuscript for the Journal of Periodontology.

ORIGINAL ARTICLES

These are papers that report significant clinical or basic research on the pathogenesis, diagnosis, and treatment of the different forms of periodontal disease. Papers dealing with design, testing, and other features of dental implants are also included.

Format
Original articles must be limited to 4,000 words (excluding the abstract, references, and figure legends). The reference list should not exceed 50 references, and the total combined number of figures and tables must be six or fewer. Multi-panel figures are acceptable.

Abstract
All original articles should be submitted with a structured abstract, consisting of no more than 250 words and the following four paragraphs:
Background: Describes the problem being addressed.
Methods: Describes how the study was performed.
Results: Describes the primary results.
Conclusion(s): Reports what authors have concluded from these results, and notes their clinical implications.
Introduction
The Introduction contains a concise review of the subject area and the rationale for the study. More detailed comparisons to

previous work and conclusions of the study should appear in the Discussion section.

Materials and Methods

This section lists the methods used in the study in sufficient detail so that other investigators would be able to reproduce the research. When established methods are used, the author need only refer to previously published reports; however, the authors should provide brief descriptions of methods that are not well known or that have been modified. Identify all drugs and chemicals used, including both generic and, if necessary, proprietary names and doses. The populations for research involving humans should be clearly defined and enrollment dates provided.

Results

Results should be presented in a logical sequence with reference to tables, figures, and supplemental material as appropriate.

Discussion

New and possible important findings of the study should be emphasized, as well as any conclusions that can be drawn. The Discussion should compare the present data to previous findings. Limitations of the experimental methods should be indicated, as should implications for future research. New hypotheses and clinical recommendations are appropriate and should be clearly identified. Recommendations, particularly clinical ones, may be included when appropriate.

Publication of Accepted Original Articles

Please note that accepted manuscripts which are classified by the Editors as "Discovery Science" will be placed on an accelerated schedule for online-only publication. See Online-Only Publication below.

REVIEW ARTICLES

These are focused reviews of basic and clinical science related to periodontics and implant dentistry. These reviews should be concise and address an important and timely clinical question. Authors should discuss clinical relevance and the impact on future understanding and practice. The review should be based on a critical assessment of the literature and should use the format and methods of a "systematic review." Detailed descriptions of the systematic review methodology are available in the Cochrane Handbook for Systematic Reviews of Interventions.1 There are many excellent published examples of systematic reviews, including "Periodontal Disease and Coronary Heart Disease Incidence: A Systematic Review and Meta-Analysis" by Humphrey et al.2

Authors of systematic reviews that include a meta-analysis should refer to the QUOROM statement.3 Authors of systematic reviews without meta-analysis should refer to the papers by Cook et al.4 and Mulrow et al.5

Higgins JPT, Green S. Cochrane Handbook for Systematic Reviews of Interventions [serial on the Internet]. September 2008; version 5.0.1. Available at: www.cochrane-handbook.org.

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Mulrow C, Langhorne P, Grimshaw J. Integrating heterogeneous pieces of evidence in systematic reviews. Ann Intern Med 1997;127:989-995.

Format

The abstract should summarize the main conclusions of the review in 350 words or less. Systematic review articles should: define a clear and clinically relevant research question; retrieve and describe the limitations of previously published reviews on this topic; and justify the need for a systematic review. The review should then define the search strategy used to identify primary articles; describe the methods used to select primary studies; specify inclusion and exclusion criteria (criteria for selecting primary studies should be based on population studied, intervention or exposure, study outcomes, and study methodology); perform a blinded assessment of the quality of the selected articles; describe the reliability of this process in terms of agreement between two evaluators; account for all studies identified by the search and justify exclusions; state their conclusions; compare their conclusions to the literature and current standard of care; outline the limitations of the review; and suggest areas for future research.

Papers should be balanced, literature-based reviews that are concise (2,000 to 3,000 words) with about 100 key references. Tables and figures should be limited to those essential to convey the results of the review, and the total combined number of figures and tables should not exceed six. Since critical reviews require selection of reports and interpretation of data, authors should disclose financial interest in the companies making products or providing services described in the review.

COMMENTARIES

The purpose of these papers is to provide a forum for discussion of controversies and other issues as they relate to the practice of periodontics and implant dentistry. Full and balanced discussion of controversies on important issues is encouraged. This may result in several authors each presenting a relevant viewpoint. Commentaries should be concise (2,000 to 3,000 words) with no more than 50 references; however, they should be complete and balanced, which may require that the issue or controversy addressed be highly focused.

Introduction
This section should clearly state the clinical question or issues to be discussed and document their importance and timeliness.

Body
The body should present the information supporting all aspects of the issues. This portion of the Commentary may be subdivided as appropriate with headings. Figures, tables, and other illustrative materials may be incorporated. The total combined number of figures and tables should not exceed six.

Summary
The summary should place the issue in perspective and point a way for future directions in addressing the controversy.

Acknowledgment(s)
Since these papers allow authors to express their opinions on a subject, it is extremely important that authors disclose any and all affiliations, financial position, or any other information that constitutes a real or perceived conflict of interest.

CASE SERIES

The Journal of Periodontology no longer publishes Case Reports. Authors are encouraged to submit Case Reports to Clinical Advances in Periodontics. The Journal of Periodontology publishes selected Case Series that describe unusual case presentations, complex diagnoses, and novel approaches to treatment within the scope of practice of periodontology. These Case Series provide valuable information for clinicians and teachers in the field.

Case Series report a sufficient number of consecutive or randomized cases to make a persuasive argument for or against the procedure, technique, or concept under discussion. Cases should be relatively homogeneous so that a systematic evaluation of one type of disease, lesion, or condition is made for the procedure under consideration. Also, treatment and documentation should be consistent and standardized for all cases. It is recognized that definitive evidence for the safety and efficacy of any procedure, drug, or device comes primarily from well-designed, randomized, controlled trials. However, well-executed Case Series may lead to hypotheses about the usefulness of new and innovative procedures, drugs, or devices and may therefore be of value to the progress of clinical science.

The requirements for patient consent, privacy, and institutional approval are well defined for manuscripts describing research on human subjects. These basic requirements are described by the International Committee of Medical Journal Editors (ICMJE) in their Uniform Requirements for Manuscripts Submitted to Biomedical Journals (available at: www.icmje.org) and are interpreted in the instructions to authors of all peer-reviewed biomedical journals, including the Journal of Periodontology.

Due to the changing ethical and legal environment around the use of patient information, the editorial team has received multiple questions about the need for subject consent from patients described in Case Series submitted for publication.

The following applies to most Case Series. It should be noted that the Editors will determine whether specific Case Series require additional approvals beyond what is described below.

Requirement for Ethics Board Approval

Most Case Series are a retrospective description of clinical findings in cases or an observed course of events that document a new aspect of patient management during the normal course of clinical treatment. Since there is no hypothesis testing, no systematic data collection beyond that which is part of routine clinical practice, no data analysis, and the work has already been done, Case Series do not usually qualify as "research" requiring approval from ethical boards designed to protect humans involved in clinical research.

(U.S. Fed. definition: "RESEARCH is any systematic investigation, including research development, testing and evaluation, designed to develop or contribute to generalizable knowledge.")

Example 1: Series of private practice implant cases in patients who have been taking bisphosphonates. Authors describe the findings in each case, which are collected and reported in a table format.

Example 2: Authors collect series of private practice implant cases in patients who have or have not been taking bisphosphonates. The sample size is sufficient for data analysis, and authors analyze and report the incidence of complications.

Example 1 does not qualify as "research," but example 2 does qualify and requires ethical approval.

Please see "Does My Case Series Need IRB Approval?" for more information.

Privacy in Case Series

No patient identifiers should be included in Case Series. If the authors choose to include any subject identifiers, the authors

must include the patient's informed written consent to publish the information.

Our policy conforms to the Uniform Requirements, which states: "Patients have a right to privacy that should not be violated without informed consent. Identifying information, including names, initials, or hospital numbers, should not be published in written descriptions, photographs, or pedigrees unless the information is essential for scientific purposes and the patient (or parent or guardian) gives written informed consent for publication. Informed consent for this purpose requires that an identifiable patient be shown the manuscript to be published. Authors should disclose to these patients whether any potential identifiable material might be available via the Internet as well as in print after publication."

It should be noted that patients may have given a signed "consent to treat," but that does not constitute permission to publish their case with personal identifiers unless they have explicitly approved the manuscript. Likewise, patient consent under government privacy rules, such as the Health Insurance Portability and Accountability Act (HIPAA) in the United States, does not constitute permission to publish their case with personal identifiers unless they have explicitly approved the manuscript.

Format

Case Series must be limited to 2,000 to 3,000 words (excluding the abstract, references, and figure legends). The reference list should not exceed 50 references, and the total combined number of figures and tables must be six or fewer. Multi-panel figures are acceptable.

Abstract

Case Series should be submitted with a structured abstract, consisting of no more than 250 words and the following four paragraphs:

Background: Describes the clinical situation being discussed.
Methods: Describes the clinical procedures (surgical and non-surgical) performed.
Results: Describes the clinical results.
Conclusion(s): Reports what authors have concluded, specifically clinical implications in practice situations.

Introduction

This section should include a critical review of the pertinent literature.

Case Description and Results

This section describes the cases, including all relevant data. For ease of presentation, tables describing longitudinal data in a chronological form may be useful. Carefully selected, high-quality clinical photographs in full color, as well as radiographs, are encouraged.

Discussion

This should include findings, put into perspective with respect to the field and literature. Unique arguments and new information gained should be summarized. Consideration of the clinical significance of the cases should be emphasized in all sections.

GUEST EDITORIALS

Guest Editorials may be invited or may be submitted from authorities in certain areas as a means of offering their perspective on one or more articles published in the Journal, or on other items of interest to the readership.

LETTERS TO THE EDITOR

Letters may comment on articles published in the Journal and should offer constructive criticism. If a letter comments on a

published article, the author(s) will be provided 30 days to respond to the observations.

Letters to the Editor may also address any aspect of the profession, including education and training, new modes of practice, and concepts of disease and its management.

Letters should be brief (<1,000 words), focused on one or a few specific points or concerns, and can be signed by no more than five individuals.

Citations should be handled as standard references.

GENERAL FORMAT

Manuscripts must be submitted in Microsoft Word. Margins should be at least 1" on both sides and top and bottom and all text should be double-spaced. Materials should appear in the following order:

Title Page
Abstract (or Introduction) and Key Words
Text
Footnotes
Acknowledgment(s)
References
Figure Legends
Tables

Figures should not be embedded in the manuscript. Please see the Journal of Periodontology Digital Art Guidelines for more information on submitting figures.

Authors should retain a copy of their manuscript for their own records.

TITLE PAGE

The title page should contain:
a concise but informative title;

first name, middle initial, and last name of each author, with the highest academic degree and the current institutional affiliation, including department, for each (please use footnote symbols in the sequence *, †, ‡, §, ‖, ¶, #, **, etc. to identify authors and their corresponding institutions);
disclaimers, if any;
the name and address (including fax number and e-mail) of the author responsible for correspondence (please indicate whether fax number and e-mail can be published);
word count and number of figures, tables, and references in the manuscript;
a short running title of no more than 60 characters, including spaces;
a one-sentence summary describing the key finding(s) from the study.
KEY WORDS

A maximum of six key words or short phrases, drawn from MeSH documentation, to facilitate indexing should be listed below the abstract.

ACKNOWLEDGMENT(S) AND CONFLICTS OF INTEREST

Acknowledgment(s)
Following the Discussion, acknowledgments may be made to individuals who contributed to the research or the manuscript preparation at a level that did not qualify for authorship. This may include technical help or participation in a clinical study. Authors are responsible for obtaining written permission from persons listed by name. Acknowledgments must also include a statement that includes the source of any funding for the study, and defines the commercial relationships of each author.

Conflicts of Interest

In the interest of transparency and to allow readers to form their own assessment of potential biases that may have influenced the results of research studies, the Journal of Periodontology requires that all authors declare potential competing interests relating to papers accepted for publication. Conflicts of interest are defined as those influences that may potentially undermine the objectivity or integrity of the research, or create a perceived conflict of interest.

Authors are required to submit:
A statement in the acknowledgments section of the manuscript that includes the source of any funding for the study, and defines the commercial relationships of each author. If an author has no commercial relationships to declare, a statement to that effect should be included. This statement should include financial relationships that may pose a conflict of interest or potential conflict of interest. These may include financial support for research (salaries, equipment, supplies, travel reimbursement); employment or anticipated employment by any organization that may gain or lose financially through publication of the paper; and personal financial interests such as shares in or ownership of companies affected by publication of the research, patents or patent applications whose value may be affected by this publication, and consulting fees or royalties from organizations which may profit or lose as a result of publication. An example is shown below.
A conflict of interest and financial disclosure form for each author. A link to this electronic form will be e-mailed to each author after manuscript submission.
Conflict of interest information will not be used as a basis for suitability of the manuscript for publication.
Example of Conflict of Interest Statement
This study was supported by a grant from the Acme Implant Corporation, Seoul, Korea. Dr. Lee is on the scientific advisory

board for Acme Implant Corporation and gives lectures sponsored by the company. Dr. Smith is a consultant and shareholder of the Brownstone Implant Corporation, Boston, Massachusetts. Dr. Wang is employed full-time as chief technical officer of the Acme Implant Corporation. Drs. Able, Kim, and Bruce report no conflicts of interest related to this study.

REFERENCES

References should be numbered consecutively in the order in which they appear in the text. A journal, magazine, or newspaper article should be given only one number; a book should be given a different number each time it is mentioned, if different page numbers are cited.

All references are identified, whether they appear in the text, tables, or legends, by Arabic numbers in superscript. Journal title abbreviations should be those used by the U.S. National Library of Medicine. If you are uncertain about the correct abbreviation for a journal title, please search for the journal at http://www.ncbi.nlm.nih.gov/nlmcatalog.

The use of abstracts as references is strongly discouraged. Manuscripts accepted for publication may be cited and should include the manuscript's DOI, if known. Material submitted, but not yet accepted, should be cited in text as "unpublished observations." Written and oral personal communications may be referred to in text, but not cited as references. Please provide the date of the communication and indicate whether it was in a written or oral form. In addition, please identify the individual and his/her affiliation. Authors should obtain written permission and confirmation of accuracy from the source of a personal communication. Presented papers, unless they are subsequently published in a proceedings or peer-reviewed journal, may not be cited as references. In addition, Wikipedia.org may not be cited as a reference. For most manuscripts, authors should limit

references to materials published in peer-reviewed professional journals. In addition, authors should verify all references against the original documents. References should be typed double-spaced. Examples of references are given below. Authors are encouraged to consult EndNote for the Journal of Periodontology's preferred reference style.

Journals

Standard journal reference. Note: list all authors if six or fewer; when seven or more, list only first three and add et al. Kurita-Ochiai T, Seto S, Suzuki N, et al. Butyric acid induces apoptosis in inflamed fibroblasts. J Dent Res 2008;87:51-55.

Corporate author. Federation Dentaire Internationale. Technical report no. 28. Guidelines for antibiotic prophylaxis of infective endocarditis for dental patients with cardiovascular disease. Int Dent J 1987;37:235.

Journal paginated by issue. Card SJ, Caffesse RG, Smith BA, Nasjleti CE. New attachment following the use of a resorbable membrane in the treatment of periodontitis in dogs. Int J Periodontics Restorative Dent 1989;9(1):59-69.

Non–English-language titles translated into English. Buchmann R, Khoury F, Hesse T, Müller RF, Lange DE. Antimicrobial therapy of peri-implant disease (in German). Z Zahnärztl Implantol 1996;12:152-157.

Books and Other Monographs

Personal author(s). Tullman JJ, Redding SW. Systemic Disease in Dental Treatment. St. Louis: The CV Mosby Company; 1983:1-5.

Chapter in a book. Rees TD. Dental management of the medically compromised patient. In: McDonald RE, Hurt WC, Gilmore HW, Middleton RA, eds. Current Therapy in Dentistry, vol. 7. St. Louis: The CV Mosby Company; 1980:3-7.

Agency publication. Miller AJ, Brunelle JA, Carlos JP, Brown LJ, Loë H. Oral Health of United States Adults. Bethesda, MD:

National Institute of Dental Research; 1987. NIH publication no. 87-2868.

Dissertation or thesis. Teerakapong A. Langerhans' cells in human periodontally healthy and diseased gingiva. [Thesis]. Houston, TX: University of Texas; 1987. 92 p.

Electronic Citations

Note: DOIs are preferred for journal articles. If a DOI is not available, please provide a URL and access date.

Online-only article. Rasperini G, Acunzo R, Limiroli E. Decision making in gingival recession treatment: Scientific evidence and clinical experience. Clin Adv Periodontics 2011;1:41-52. doi:10.1902/cap.2011.100002.

Ahead of print. McGuire MK, Scheyer ET, Nevins M, et al. Living cellular construct for increasing the width of keratinized gingiva. Results from a randomized, within-patient, controlled trial [published online ahead of print March 29, 2011]. J Periodontol; doi:10.1902/jop.2011.100671.

Web sites. Centers for Disease Control and Prevention. Periodontal Disease. Available at: http://www.cdc.gov/OralHealth/topics/periodontal_disease.htm. Accessed September 29, 2010.

TABLES

Tables should be numbered consecutively in Arabic numbers in the order of their appearance in the text. A brief descriptive title should be supplied for each. Explanations, including abbreviations, should be listed as footnotes, not in the heading. Every column should have a heading. Statistical measures of variations such as standard deviation or standard error of the mean should be included as appropriate in the footnotes. Do not use internal horizontal or vertical rules. The submission system will easily read tables created with Word's table utility or when inserted into Word from Excel.

FIGURES

Please see the Journal of Periodontology Digital Art Guidelines for detailed instructions on submitting high-quality images.

Figure Legends

Legends should be typed double-spaced with Arabic numbers corresponding to the figure. When arrows, symbols, numbers, or letters are used, explain each clearly in the legend; also explain internal scale, original magnification, and method of staining as appropriate. Panel labels should be in capital letters. Legends should not appear on the same page as the actual figures.

FOOTNOTES

Footnotes should be used only to identify author affiliations; to explain symbols in tables and illustrations; and to identify manufacturers of equipment, medications, materials, and devices. Use the following symbols in the sequence shown: *, †, ‡, §, ‖, ¶, #, **, ††, etc.

SUPPLEMENTARY MATERIAL

The Journal of Periodontology includes supplementary material in the online Journal (www.joponline.org). All supplemental material should be called out in the text.

Supplementary Figures and Tables

Journal of Periodontology articles are limited to a combined total of six figures and tables in the print publication. Any additional figures and tables should be submitted as supplementary files. Each supplementary figure or table should be submitted as a separate file. Please follow the guidelines regarding resolution, format, etc. for printed figures (see Digital Art Guidelines) and

tables (see above) when preparing supplementary figures and tables. In summary, each figure, table, or multimedia file should be uploaded separately and the file names should clearly identify the file (i.e., SupplementaryFigure1.tif, SupplementaryTable1.xls, etc.). If file size limitations prevent you from uploading your supplemental material, please e-mail jerry@perio.org.

Supplementary Videos

The Journal of Periodontology publishes short videos to supplement a paper when appropriate. Most standard video formats are accepted. Videos should be edited to remove extraneous material. Authors should adhere to OSHA regulations when preparing their videos. Please e-mail bethanne@perio.org for information on how to submit videos. If your video is accepted for publication, all authors will need to submit a video copyright form. This form can be found on ScholarOne Manuscripts in the upper right-hand corner under "Instructions & Forms."

STYLE

Please follow the guidelines below when preparing a manuscript:

Be sure to put the genus and species of an organism and journal names in the reference section in italics.

Do not italicize common Latin terms such as in vitro, in vivo, e.g., or i.e.

Use a block style; do not tabulate or indent material.

Refer to the newest edition of the Glossary of Periodontal Terms published by the American Academy of Periodontology for preferred terminology.

Authors are encouraged to use the disease classification as outlined in the Annals of Periodontology, volume 4 (1999 International Workshop for a Classification of Periodontal Diseases and Conditions).

Create equations as text, treating any mathematical symbols as special characters and assigning them the Symbol font.

Measurements of length, height, weight, and volume should be reported in metric units or their decimal multiples. Temperatures should be given in degrees Celsius and blood pressure in millimeters of mercury. All hematologic and clinical chemistry measurements should be reported in the metric system in terms of the International System of Units (SI). Description of teeth should use the American Dental Association (i.e., Universal/National) numbering system.

Statistical methods should be described such that a knowledgeable reader with access to the original data could verify the results. Wherever possible, results should be quantified and appropriate indicators of measurement error or uncertainty given. Sole reliance on statistical hypothesis testing or normalization of data should be avoided. Data in as close to the original form as reasonable should be presented. Details about eligibility criteria for subjects, randomization, methods for blinding of observations, treatment complications, and numbers of observations should be included. Losses to observations, such as dropouts from a clinical trial, should be indicated. General-use computer programs should be listed. Statistical terms, abbreviations, and symbols should be defined. Detailed statistical, analytical procedures can be included as an appendix to the paper if appropriate.

AUTHORSHIP

Individuals identified as authors must meet all of the following criteria established by the International Committee of Medical Journal Editors: 1) substantial contributions to conception and design, or acquisition, analysis, or interpretation of data; 2) drafting the article or revising it critically for important intellectual content; 3) final approval of the version to be published; and 4) agreement to be accountable for all aspects of the work in

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Once the Journal has received a manuscript, any changes in authorship must be faxed to the editorial office at 312/573-3225 (attn: Jerry Eberle, Editorial Specialist) or e-mailed to jerry@perio.org and must contain the signature of the author who has been added or removed from the paper. Authors who are added must submit a conflict of interest and financial disclosure form (see below).

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